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Phase II Study of the Combination of Low-Intensity Chemotherapy and Blinatumomab in Patients with Philadelphia Chromosome Negative Relapsed/Refractory Acute Lymphoblastic Leukemia (ALL)

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1. INTRODUCTION

1.1 Outcomes of Acute Lymphoblastic Leukemia (ALL) in Adults

Although >90% of adult patients with ALL achieve a complete remission (CR) with intensive chemotherapy, many patients ultimately relapse, leading to long-term event-free survival (EFS) rates of 30-40%.[1-5] Current outcomes of salvage chemotherapy for ALL are poor, with less than half of patients achieving a second CR, with rates varying based on prior therapy and duration of first remission.[6-8] Overall, the long-term survival for patients with ALL after first relapse is very poor with only 10-20% achieving cure.[8] CR and long-term survival rates are even lower for patients with multiply refractory disease. Given the poor outcomes for patients with relapsed/refractory ALL, novel agents and combinations capable of achieving remission to allow for potentially curative allogeneic stem cell transplantation SCT are needed.

1.2 Low-Intensity Chemotherapy in Relapsed or Refractory ALL

For patients with relapsed or refractory ALL, further intensification of chemotherapy has not proved to be effective.[9] We have recently shown the benefit of dose-reduced chemotherapy in combination with novel therapies for these patients. In patients with relapsed or refractory Philadelphia chromosome (Ph)-negative ALL, the combination of inotuzumab ozogamicin, an anti-CD22 drug-conjugate, with a “mini-hyper-CVD” regimen has shown promising clinical activity with reduced toxicity.[10] In the most recent update of these data, 69% of patients achieved CR or CR with inadequate platelet recovery (CRp). Among patients in first salvage, the 1-year overall survival (OS) rate was 59% with a median OS of 17 months. These promising results in elderly patients with ALL provide the rationale for combining the mini-hyper-CVD backbone with other novel agents.

1.3 Blinatumomab

Blinatumomab is a bi-specific T-cell engager (BiTE) antibody against CD3 and CD19 which is designed to direct cytotoxic T cells to CD19-expressing leukemic cells.[11] CD19 is nearly universally expressed on the cell surface of precursor B-ALL leukemic blasts and therefore is a rationale target for antibody-directed therapy for this disease.[12] The drug was initially administered as an intermittent infusion two to three times weekly, but lack of activity and serious neurologic toxicity caused the schedule of administration to be abandoned.[13] Based on the short half-life of the drug and the mechanism of action, continuous infusion over several weeks were subsequently investigated. This drastically improved the activity of the drug and helped reduce adverse effects.

In a phase 2 study of 189 heavily pre-treated patients with relapsed/refractory Ph-negative ALL, blinatumomab given as a continuous intravenous for 4 consecutive weeks on a 6-week cycle was associated with a CR plus CR with partial hematologic recovery (CRh) rate of 43% and a median response duration and OS of 9 months and 6 months, respectively.[14] These promising results have provided the rationale for a phase 3 randomized trial (TOWER study) of blinatumomab versus investigator’s choice chemotherapy for patients with relapsed/refractory Philadelphia-negative ALL. The overall response rates were 45% and 30% (p=0.007), respectively. Molecular remission rates among responders, defined as < 10-4 blasts in the first 12 weeks, were 75% and 48%, respectively. Blinatumomab prolonged survival, the primary study endpoint: the median survival was 7.7 months (5.6-9.6) with blinatumomab and 4.0 months (2.9-5.3) with standard of care chemotherapy, respectively (p=0.012).[15]

In another study of 116 patients with Ph-negative ALL who remained minimal residual disease (MRD)-positive after initial chemotherapy and subsequently received blinatumomab, MRD negativity was achieved in 76% of patients.[15] The median OS was significantly longer in patients who subsequently achieved MRD negativity compared to those who remained MRD-positive (40 months vs. 12 months, respectively; P=0.001). Notably, allogeneic stem cell transplantation (ASCT) did not confer a survival benefit for patients who achieved MRD negativity in first remission. These results provide evidence that blinatumomab is highly effective in eradicating MRD and that this translates into better long-term survival for MRD responders.

1.4 Summary

In summary, the outcomes of patients with relapsed/refractory ALL are poor, and novel treatment strategies are needed in this population. There is strong clinical evidence for blinatumomab as a potent therapeutic agent in patients with ALL. Given previous experience that intensification of chemotherapy in the relapsed/refractory setting does not improve outcomes, there is a rationale to combine novel agents with a less intensive chemotherapy backbone. Previous data of the combination of mini-hyper-CVD with inotuzumab ozogamicin in patients with ALL suggests that this dose-reduced chemotherapy backbone can result in excellent response rates with minimal toxicity, with an improved outcome compared to Inotuzumab alone. The median survival was 11 and 6 months respectively. Taken together, there is a strong rationale for a phase II trial combining blinatumomab with mini-hyper-CVD in patients with relapsed/refractory ALL.

2. STUDY OBJECTIVES

- The primary objective is to evaluate the combined effect of blinatumomab and mini-hyper-CVD on event-free survival
- The secondary objectives include evaluating other clinical efficacy endpoints (minimal residual disease [MRD] negativity, duration of response, the overall response rate [CR + CR with inadequate count recovery (CRi)]) of the regimen occurred any time during the treatment and overall survival and determining the safety of the combination regimen.

3. SELECTION OF PATIENTS

Patients will be selected from those referred to the Leukemia department at MD Anderson Cancer Center through the normal of process of referral. Eligible patients will be registered after the process of consenting on the MD Anderson protocol and data monitoring system.

3.1 Inclusion Criteria

1. Patients ≥ 18 years of age with first or second relapsed/refractory B-cell ALL

2. Performance status ≤3 (ECOG Scale)

3. Adequate liver function as defined by the following criteria:
   - Total serum bilirubin ≤ 2 x upper limit of normal (ULN), unless due to Gilbert's syndrome, hemolysis or the underlying leukemia approved by the PI
   - Alanine aminotransferase (ALT) ≤ 3 x ULN, unless due to the underlying leukemia approved by the PI OR

- Aspartate aminotransferase (AST) ≤ 3 x ULN unless due to the underlying leukemia approved by the PI

4. Signed informed consent

5. Women of childbearing potential (WOCBP) or male subjects with a partner who is WOCBP must agree to use contraception during the study, if sexually active.

3.2 Exclusion Criteria

1. Patients with Ph-positive ALL or Burkitt leukemia

2. Active, uncontrolled central nervous system (CNS) leukemia involvement

3. Active serious infection not controlled by oral or intravenous antibiotics.

4. Active secondary malignancy other than skin cancer (e.g., basal cell carcinoma or squamous cell carcinoma) that in the investigator's opinion will shorten survival to less than 1 year.

5. Known hepatitis B or C infection, or known seropositivity for HIV

6. Active Grade III-V cardiac failure as defined by the New York Heart Association Criteria.

7. Patients with a cardiac ejection fraction (as measured by either MUGA or echocardiogram) <40%

8. Prior history of treatment with blinatumomab.

9. Treatment with any investigational antileukemic agents or chemotherapy agents in the last two weeks , unless full recovery from side effects has occurred or patient has rapidly progressive disease judged to be life-threatening by the investigator. .

10. Pregnant and lactating women will not be eligible; women of childbearing potential should have a negative pregnancy test prior to entering on the study and be willing to practice methods of contraception. Women do not have childbearing potential if they have had a hysterectomy or are postmenopausal without menses for 12 months. In addition, men enrolled on this study should understand the risks to any sexual partner of childbearing potential and should practice an effective method of birth control.

4. TREATMENT OF SUBJECTS

4.1 Variations in dose reductions of the individual chemotherapy or the administration of blinatumomab or supportive care dose schedules other than those suggested below are allowed in the best interest of patients. Such patients should be discussed with the PI. Dose escalations of chemotherapy above those outlined in the protocol; however, are not allowed. Variations in infusion times due to minor differences in IV bag overfill/underfill and institutional procedure on flushing chemotherapy lines will not result in protocol deviation.

4.2 **Treatment Overview (Figure 1)** – Blinatumomab will be given in combination with mini-hyper-CVD. Each cycle of induction and consolidation is 42 days (+/- 7 days). Mini-hyper-CVD

chemotherapy regimen consists of a total of 8 cycles with mini-hyper-CVD alternating with high-dose methotrexate and cytarabine administered approximately every 21 days (+/- 7 days to allow for recovery from myelosuppression or infection). A total of 4 cycles of blinatumomab are planned in induction/consolidation and 2 additional cycles during maintenance. Blinatumomab is given by continuous infusion for 4 weeks, followed by a 2 week treatment-free period. Patients will then receive 2 years of maintenance therapy with POMP (6-mercaptopurine [6-MP], vincristine, methotrexate and prednisone) and blinatumomab single agent given in cycles 6 and 12 of the first year and vincristine and prednisone alone for the second year. Patients achieving remission will be referred to ASCT at the discretion of the treating physician, based on donor availability and suitability for ASCT.

4.3 **Cycle 1** – Eligible patients will receive mini-hyper-CVD cycle 1 phase A beginning on day -3 (day minus 3). After completion of mini-hyper-CVD, patients will receive the first cycle of blinatumomab (starting on "day 1"). Blinatumomab will be given as a continuous intravenous infusion over 4 weeks followed by a treatment-free period of 2 weeks, defined as one 42 day treatment cycle. Patients will remain hospitalized up through day 6 of blinatumomab. Hospital stay may be prolonged in patients with infections or other issues requiring inpatient stay. Subsequent blinatumomab will be given in the outpatient setting via an ambulatory infusion pump.

Mini-hyper-CVD cycle 1 phase B will be given on day 21 (or later as count recovery allows). Treatment will be given in the inpatient setting, and patients will continue to receive blinatumomab concomitantly with mini-hyper-CVD. Patients can continue outpatient treatment with blinatumomab once mini-hyper-CVD has completed.

4.4 **Cycle 2-4** – Mini-hyper-CVD cycle 2 phase A and blinatumomab will begin on day 1. Blinatumomab will be given as a continuous intravenous infusion over 4 weeks followed by a treatment-free period of 2 weeks, defined as one 42 day treatment cycle. Patients will remain hospitalized until the completion of both cycle 2 phase A mini-hyper-CVD and at least 2 days of blinatumomab. Hospital stay may be prolonged in patients with infections or other issues requiring inpatient stay. Subsequent blinatumomab will be given in the outpatient setting via an ambulatory infusion pump.

Mini-hyper-CVD cycle 2 phase B will be given on day 21 (or later as count recovery allows). Treatment will be given in the inpatient setting, and patients will continue to receive blinatumomab concomitantly with mini-hyper-CVD. Patients can continue outpatient treatment with blinatumomab once mini-hyper-CVD has completed.

4.5 **Maintenance phase -** At the completion of the 4 cycles of mini-hyper-CVD plus blinatumomab, patients who did not undergo ASCT will receive up to 2 years of maintenance. Patients may be moved from the intensive chemotherapy phase to the maintenance phase with POMP prior to completion of 4 cycles due to frequent infectious complications or if the patient is intolerant to the chemotherapy after discussion with the PI. If an early transition to maintenance phase is planned, it is recommended that patients receive at least 1 complete cycle of mini-hyper-CVD plus blinatumomab. The reason for early transition to the maintenance phase will be documented in the medical record.

4.6 **Mini-hyper-CVD Regimen**

1. General Considerations

- The mini-hyper-CVD (A phase) will alternate with methotrexate and cytarabine (B phase) administered every 21 days or later after the last mini-hyper-CVD course (A phase), or earlier if count recovery allows.
- Anti-emetic therapy with each course of chemotherapy as needed.
- Filgrastim product will be administered with each course after the completion of chemotherapy. The choice of Filgrastim product will be determined by the institutional formulary and patient's insurance.
- Next course/phases may be started when granulocytes > 1.0 x 10^9/L and platelets ≥ 50 x 10^9/L. Courses/phases may be started with dose reductions prior to full platelet recovery, if the treatment is delayed (e.g., greater than 28 days from last course/phase).
- Prophylactic antimicrobials may be given with each course until neutrophil recovery to 500/μL or greater (or other antibiotics if being treated for active infection). Suggestions include: Levaquin 500 mg po daily (or other quinolone) or trimethoprim-sulfamethoxazole double strength one tablet p.o. b.i.d. or other appropriate antibacterial agent. Fluconazole 400 mg p.o. daily or other appropriate antifungal agent. Valacyclovir 500 mg p.o. daily, acyclovir 400 mg p.o. b.i.d. or other appropriate antiviral agent.
- Omit azoles day -1 (day before), same day (day of), and day +1 (day after) vincristine
- For patients with CD20 expression (≥20% by flow cytometry), rituximab 375 mg/m^2 may be added at the discretion of the treating physician. Up to 4 doses of rituximab may be given during each cycle (up to 8 total doses).
- All patients recommended to have a baseline chest radiograph before each course of methotrexate and cytarabine; methotrexate dose will be adjusted as indicated below for pleural effusions.

2. Hyper-CVD
   - Phase 1A
     - Cyclophosphamide (CTX) 150 mg/m^2 intravenously (IV) over 3 hrs (±1 hour) every 12 hrs x 6 doses days -3, -2, -1
     - MESNA 300 mg/m^2/d IV continuous infusion daily for 24 hrs (± 4 hours), starting approximately 1 hour prior to CTX and completing by approximately 12 hrs after the last dose of CTX.
     - Vincristine 2 mg IV on day 0 (± 3 days) and day 7 (± 3 days). Vincristine is not considered to be myelosuppressive and may be given while patients are receiving concomitant G-CSF; no known adverse effects have been observed with the 2 agents given together.
     - Dexamethasone 20 mg IV or p.o. daily on days -3 to 0)± 3 days) and days 7-10 (± 3 days).
     - Filgrastim product 10 mcg/kg (rounded) subcutaneously daily (or 5 mcg/kg twice daily) until post-nadir granulocytes > 1.0 x 10^9/L. Filgrastim product may be stopped earlier for bone pain or other related toxicity. Minimum time allowed between courses is 14 days. Pegfilgrastim may replace filgrastim product at a dose of 6 mg SQ on day 1 (±3 days).
     - CNS prophylaxis is recommended with methotrexate 12 mg intrathecally (IT) (6 mg via Ommaya reservoir) on day -2 (± 3 days) and cytarabine 100 mg IT on day 7 (± 3 days).
     - For patients with CD20 expression (≥20% by flow cytometry), rituximab 375 mg/m^2 may be added at the discretion of the treating physician. Recommended days of rituximab administration are days -3 (±3 days) and 0 (±3 days) (prior to initiation of blinatumomab).

- Phase 2A to 4A
  - Cyclophosphamide (CTX) 150 mg/m^2 intravenously (IV) over 3 hrs (± 1 hour) approximately every 12 hrs x 6 doses days 1, 2, 3
  - MESNA 300 mg/m^2/d IV continuous infusion daily for 24 hrs (± 4 hours), starting approximately 1 hour prior to CTX and completing by approximately 12 hrs after the last dose of CTX.
  - Vincristine 2 mg IV on day 4 (± 3 days) and day 11 (± 3 days). Vincristine is not considered to be myelosuppressive and may be given while patients are receiving concomitantfilgrastim product; no known adverse effects have been observed with the 2 agents given together.
  - Dexamethasone 20 mg IV or p.o. daily on days 1-4 (± 3 days) and days 11-14 (± 3 days).
  - Filgrastim product10 mcg/kg (rounded) subcutaneously daily (or 5 mcg/kg twice daily) until post-nadir granulocytes > 1.0 x 10^9/L. Filgrastim product may be stopped earlier for bone pain or other related toxicity. Minimum time allowed between courses is 14 days. Pegfilgrastim may replace filgrastim product at a dose of 6 mg SQ on day 5 (+ 3 days).
  - CNS prophylaxis is recommended with methotrexate 12 mg IT (6 mg via Ommaya reservoir) on day 2 (± 3 days) and cytarabine 100 mg IT on day 7 (± 3 days) in Cycle 2A.
  - For patients with CD20 expression (≥20% by flow cytometry), rituximab 375 mg/m2 may be added at the discretion of the treating physician. Recommended days of rituximab administration are days 1 (+3 days) and 8 (+3 days) of Cycle 2A.

3. Methotrexate and cytarabine (cycles 1B to 4B)
   - Methotrexate (MTX) 50 mg/m^2 IV over 2 hrs (+1 hour) followed by 200 mg/m^2 over 22 hrs (+3 days) on day 22. Total duration of administration is approximately 24 hours (2 plus 22 hours) (+3 hours).
   - Cytarabine 0.5 g/m^2 IV over 3 hrs (+1 hour) approximately every 12 hrs for 4 doses on days 23 and 24.
   - Leucovorin rescue 50 mg IV or PO followed by 15 mg IV or PO approximately every 6 hours for up to 8 doses beginning 12 hrs (± 3 hrs) post MTX completion, i.e. approximately 36 hours from start of MTX.
   - Check MTX levels around time 0h, 24h and 48h post completion of MTX unless methotrexate cleared:
     - if > 20 μM at time 0, hold cytarabine and repeat level; if continues to be > 20 μM reduce cytarabine to 0.25 g/m^2 IV over 2 hours every 12 hours for 4 doses on days 23 and 24. Begin leucovorin rescue as described above.
     - if > 1 μM at 24hrs or > 0.1 μM at 48 hours, increase leucovorin rescue to 50 mg IV or PO every 6 hrs until serum methotrexate level is < 0.1 μM. Clearance to levels 0.15 μM or less is acceptable in patients with normal renal function.
     - Leucovorin rescue may be increased further for elevated methotrexate levels or delayed clearance
   - Filgrastim product 10 mcg/kg (rounded) subcutaneously daily (or 5 mcg /kg twice daily) until post-nadir granulocytes ≥ 1.0 x 10^9/L. Filgrastim product may be stopped earlier for bone pain or other related toxicity. Minimum time allowed between courses is 14 days. Pegfilgrastim may replace Filgrastim product at 6 mg SQ on day 25 (+3 days).

- For patients with CD20 expression (≥20% by flow cytometry), rituximab 375 mg/m^2 may be added at the discretion of the treating physician. Recommended days of rituximab administration are days 22 (±3 days) and 29 (±3 days) of Cycles 1B and 2B.
- CNS prophylaxis is recommended with Cytarabine 100 mg IT on day 26 (± 3 days) and Methotrexate 12 mg IT (6 mg via Ommaya reservoir) on day 29 (± 3 Days) in Cycles 1B and 2B.

4.7 Blinatumomab

- General considerations
  - Patients will receive blinatumomab as continuous intravenous infusion over 4 weeks followed by a treatment-free period of 2 weeks, defined as one 42-day treatment cycle. A total of 4 cycles of blinatumomab are planned for induction/consolidation and 2 cycles during maintenance.
  - Blinatumomab will be given in the inpatient setting for the first 6 days of cycle 1 and the first 2 days of cycle 2 as described below.
  - In the outpatient setting, the subject will return to the study site at specific intervals to change the infusion bag. The drug administration should not be interrupted, if possible. In case of infusion interruption, due to any technical or logistic reason, the interruption should be as short as possible and the infusion continued at the earliest time possible. Every interruption longer than 3 hours should be documented. If the interruption is longer than 4 hours, re-start of the infusion should be performed under healthcare supervision. Re-administration of dexamethasone premedication prior to re-initiating blinatumomab is recommended. If possible, the infusion duration before and after an interruption should total 28 days per treatment cycle.
- Dexamethasone premedication
  - Premedication with dexamethasone is intended to prevent CRS events associated with blinatumomab treatment. Within approximately1 hour before the start of treatment in each treatment cycle, mandatory premedication with dexamethasone at 20 mg IV or orally daily is required for the prevention of CRS resulting from blinatumomab. If at least 20mg IV of dexamethasone is given as part of the mini-hyper-CVD regimen within 1 hour of starting blinatumomab, this administration will serve as the requisite steroid premedication and no additional dose will be required.
  - Dexamethasone 20mg IV or orally daily should also be given within approximately 1 hour of any dose escalation (e.g. on day 5 of cycle 1).
- Treatment regimen
  - Cycle 1
    - The initial dose of blinatumomab will be 9 ug/day for the first 4 days of treatment (days 1-4) (±3 days) to mitigate the potential for cytokine release syndrome (CRS) or CNS events associated with introduction of blinatumomab)
    - Blinatumomab will be escalated to 28 ug/day on day 5 through day 28.
    - Patients should remain inpatient for the first 6 days of blinatumomab treatment (at least 48 hours at the higher dose level).
    - Subsequent blinatumomab infusion may be continued as an outpatient.
  - Cycles 2 to 4
    - Blinatumomab will be given at a dose of 28 ug/day on days 1 (±3 days) through day 28 (±3 days).

        - Patient should remain inpatient for the first 48 hours of blinatumomab treatment during cycle 2. Subsequent cycles will be administered as outpatients.
        - Subsequent blinatumomab infusion may be continued as an outpatient.
    - Patients who weigh less than 45kg at the start of each Blinatumomab cycle will have blinatumomab dosing discussed and approved by the principle investigator.

4.8 Intrathecal Treatments

- 8 prophylactic IT chemotherapy treatments are recommended. However, if the patient has had prior IT therapy, number of IT chemotherapy can be reduced if thought by the treating physician to be in the best interest of the patient after discussion with the PI.

4.9 Suggested Standard Dose Reductions/Modifications

- Vincristine 1 mg IV days 1 and 8 (50% reduction) if:
    - Bilirubin > 2 mg/dl and ≤ 3 mg/dl
    - Clinically significant grade 2 peripheral neuropathy persisting greater than 2 weeks.
    - Eliminate vincristine for grade 3-4 peripheral neuropathy, including grade 3-4 ileus suspected to be related to vincristine, or bilirubin > 3 mg/dL.
- Methotrexate:
    - Consider reduction by 25%-50% for grade 3 or worse mucositis with previous methotrexate course.
    - Reduce by 50% for calculated creatinine clearance 10-50 ml/min, if < 10 ml/min, hold methotrexate.
    - Reduce by 25% to 75% for delayed excretion and/or nephrotoxicity with previous methotrexate course.
    - Reduce by 50% for pleural effusion or ascites (drain effusion if possible). All patients should have a chest radiograph prior to each course of methotrexate and cytarabine.
- Blinatumomab
    - Table 1 outlines the recommended dose adjustments according to type of adverse event and grade.
        - Common Terminology Criteria for Adverse Events (CTCAE) grade 4 adverse events at least possibly related to blinatumomab will require permanent discontinuation of blinatumomab.
        - For subjects who experience a CTCAE grade 3 CNS adverse event, a contrast-enhanced magnetic resonance imaging of the head should be performed prior to re-initiation of blinatumomab.
        - For all other CTCAE grade 3 events and clinically significant laboratory value changes, investigator assessment should be used to determine the risk/benefit to continue blinatumomab therapy with or without dose reduction or discontinue therapy.
        - Patients who have been dose-reduced will have an option to receive the higher dose level once the adverse event has resolved to grade 1 or less. Restart of the infusion should be performed in the hospital. Before blinatumomab is re-started, premedication with dexamethasone 20 mg IV is required.
        - In addition to the events described in Table 2, the dose may be temporarily or permanently reduced to 9 µg/day if, by investigator´s judgment, it is necessary for safety reasons. After at least 7 days of dosing at 9 µg/day, the dose may

be increased to 28 µg/day or treatment may be continued at the dose of 9 µg/day after consultation with the PI.

- Table 2 outlines the recommendations for dexamethasone premedication and management of CRS and CNS events.
- Other modifications of drug schedules may be implemented if judged to be in the best clinical interest of the patient after discussion with PI or at the discretion of the treating physician. This includes delays in chemotherapy cycles because of persistent myelosuppression, other side effects, patient request, or other reasons.
- Dose reductions exceeding those above or in other agents, e.g., leucovorin, antibiotics, antiemetics, etc., are allowed after discussion with the PI

Table 1 – Dose Adjustments of Blinatumomab

Toxicity	Grade	Action
Cytokine release syndrome (CRS)	Grade 3	Withhold blinatumomab until no more than Grade 1 (mild), then restart blinatumomab at 9 mcg/day. Escalate to 28 mcg/day after 7 days if the toxicity does not recur. If the toxicity occurred at 9 mcg/day, or if the toxicity takes more than 7 days to resolve, discontinue blinatumomab permanently
	Grade 4	Discontinue blinatumomab permanently
Neurological toxicity	Seizure	Discontinue blinatumomab permanently if more than one seizure occurs
	Grade 3	Withhold blinatumomab until no more than Grade 1 (mild) and for at least 3 days, then restart blinatumomab at 9 mcg/day. Escalate to 28 mcg/day after 7 days if the toxicity does not recur. If the toxicity occurred at 9 mcg/day, or if the toxicity takes more than 7 days to resolve, discontinue blinatumomab permanently
	Grade 4	Discontinue blinatumomab permanently
Other Clinically Relevant Adverse Reactions	Grade 3	Withhold blinatumomab until no more than Grade 1 (mild), then restart blinatumomab at 9 mcg/day. Escalate to 28 mcg/day after 7 days if the toxicity does not recur. If the toxicity takes more than 14 days to resolve, discontinue blinatumomab permanently. If toxicity is solely attributable to laboratory parameter, consider interruption at investigator discretion.
	Grade 4	Discontinue blinatumomab permanently

Table 2 - Recommendations for Dexamethasone Premedication

Treatment Phase	Target Patient	Dexamethasone Dose
Pre-dose dexamethasone before each blinatumomab treatment	All patients (before each cycle and dose increase)	Dexamethasone 20 mg IV within 1 hour before start of treatment in each treatment cycle, and within 1 hours before dose increase
Infusion interruption/dose modification due to adverse event	Patients who interrupt treatment > 4 hours	Dexamethasone 20 mg IV within 1 hourbefore re-start of treatment
In case of signs of CRS	Patients with signs of CRS	Dexamethasone orally or IV at a dose of up to 24mg/day for up to 3 days. The dose should then be reduced step-wise over 4 days
Infusion interruption/dose modification due to CNS events	Patients with CNS-related AE	Dexamethasone orally or IV at a dose of up to 24mg/day for up to 3 days. The dose should then be reduced step-wise over 4 days

4.10 Maintenance

Following 4 cycles of mini-hyper-CVD plus blinatumomab, maintenance chemotherapy with POMP (6-MP, vincristine, methotrexate and prednisone) for 12 cycles, followed by vincristine and prednisone alone for another 12 cycles will be given (for total of 2 years of maintenance therapy). Cycle 6 and 12 of POMP will be replaced with a full blinatumomab cycle. Blinatumomab will be administered at the dose of 28 µg/day for 28 days (42 day, cycle). Administration of Blinatumomab will be performed in outpatient unless contra-indicated. When patients transition from POMP to vincristine and prednisone alone, they should continue the dose of vincristine and prednisone they were receiving during the POMP phase.

Maintenance chemotherapy with 6-mercaptopurine (6-MP), methotrexate (MTX), vincristine, and prednisone (POMP) for approximately 12 cycles:

- 6-MP 50 mg PO twice times daily (BID)
- MTX 10mg/m^2 (rounded) PO weekly
- Vincristine 2 mg IV on day 1 approximately every 28 days
- Prednisone 50 mg PO daily days 1 to 5 approximately every 28 days, starting with vincristine (if given).

Patients will be started at dose level 0. Suggested maintenance chemotherapy dose adjustments are as below:

Table 3. Suggested Maintenance Chemotherapy Dose Adjustments

Dose Level	Vincristine	Prednisone	6-MP	Methotrexate
0	2 mg	50 mg	50 mg bid	10 mg/m^2 weekly
-1	1 mg	40 mg	50 mg daily	7.5 mg/m^2 weekly
-2	--	30 mg	--	5 mg/m^2 weekly
-3	--	--	--	--

1. Prednisone
   - Dose should remain at 50 mg unless steroid myopathy or other significant toxicity occurs.
   - Prednisone not required to be dose-adjusted for hyperglycemia.
   - Further reductions beyond what is shown in the table above may be allowed if recommended by the treating physician and after discussion with the PI.

2. Vincristine
   - Decrease by one dose level for grade 2 peripheral neuropathy persisting longer than 2 weeks and bilirubin >2 mg/dl and ≤ 3 mg/dl
   - Discontinue for grade 3 or greater peripheral neuropathy persisting longer than 2 weeks, including grade 3-4 ileus suspected to be related to vincristine, or bilirubin > 3 mg/dL.

3. 6-MP and methotrexate
   - For Grade 3 myelosuppression or worse, or for Grade 3 or worse non-myelosuppression complications, dose should be reduced.
   - May interrupt treatment and resume at -1 level when toxicity resolved to Grade 1 or better.

- Other dose modifications may be implemented if judged to be in the best clinical interest of the patient after discussion with principal investigator or at the discretion of the treating

physician. This includes delays in chemotherapy cycles because of persistent myelosuppression, other side effects, patient request, intrathecal chemotherapy schedule or other reasons.

5. CONCOMITANT MEDICATIONS

Medications that inhibit platelet function (i.e.,aspirin, dipyridamole, epoprostenol, eptifibatide, clopidogrel, cilostazol, abciximab, ticlopidine, and any non-steroidal anti-inflammatory drug) or Anticoagulants (warfarin, heparin/low molecular weight heparin [e.g., danaparoid, dalteparin, tinzaparin, enoxaparin]) should be avoided as much as possible during induction and consolidation due to expected thrombocytopenia.

Exceptions include low-dose warfarin for prophylaxis to prevent catheter thrombosis, and for heparin-flushes for IV lines. If patients develop deep vein thrombosis during the course of therapy or are receiving anticoagulation for indications such as recent thrombosis or artificial heart valves these drugs may be continued with close monitoring of the patients.

Prophylactic antibiotics, antifungals, and antiviral agents as discussed above are recommended; however, the use of these or other drugs will be left to the treating physician's discretion. Antifungal prophylaxis with azoles should be avoided on Day -1, 0, and +1 of vincristine.

6. STUDY PROCEDURES

6.1. PRETREATMENT EVALUATION – within 28 days prior treatment dosing

1. History and physical examination and vital signs, documentation of disease.
2. Concomitant medication and AE assessment
3. Performance status assessment
4. CBC, platelet count, differential, total bilirubin, creatinine, ALT OR AST.
5. Bone marrow aspirate and cytogenetics.
6. EKG
7. Pregnancy test in patients of appropriate age and menopausal state
8. Echocardiogram or MUGA scan to assess cardiac function
9. A chest radiograph should be performed at baseline
10. Flow cytometry for immune correlates on peripheral blood or bone marrow.

6.2 EVALUATION DURING STUDY

1. At least weekly CBC, platelet count and differential for courses 1-4, then every 4-6 weeks during maintenance.
2. Total bilirubin, ALT OR AST and creatinine weekly for courses 1-4, then every 4-6 weeks during maintenance.
3. Bone marrow aspiration on day 21 ($\pm$ 7 days) and day 42 ($\pm$ 7 days) of cycle 1 (or at the time of hematologic recovery). Bone marrow aspiration should also be performed at the end of cycle 2 (day 42 $\pm$ 7 days), for patients who remain positive for minimal disease after cycle 1, and at the end of cycle 4 (day 42 $\pm$ 7 days) prior to beginning maintenance therapy. When possible, add cytogenetics, flow minimal residual disease assessment. Additional bone marrow assessment during maintenance as clinically indicated.

4. Flow cytometry of the peripheral blood will be run on days 1 (±2 days) and 14 (±2 days) of each cycle of blinatumomab, as well as on day 28 (±2 days) of the last cycle (see Appendix A) (If omitted and after discussion with the PI this will not constitute a deviation on the study).
5. Evaluate for toxicity assessment
6. Chest X-ray before each course of methotrexate and cytarabine (B cycles)
7. At each study visit concomitant medication and AE assessment will be recorded.
8. ECHO or MUGA scan at end of cycle 2B

Table 4. Schedule of Study Procedures

	Mini-hyper-CVD plus blinatumomab									**Maintenance**	**Follow-up****
A: Hyper-CVD B: MTX/Cytarabine	**Pre-Rx**	**1A**	**1B**	**2A**	**2B**	**3A**	**3B**	**4A**	**4B**		**30 Days after last dose of study drugs**
Informed consent	X										
EKG	X										
History + physical	X										
Vital signs	X										
Performance Status	X										
CBC/Diff/Platelet	X	X	X	X	X	X	X	X	X	Every 4-6 weeks	
SGPT or SGOT, bili, creat	X	X	X	X	X	X	X	X	X	Every 4-6 weeks	
Marrow, cytogenetic	X	X	X		X				X	As indicated	
MRD assessment by FCM		X	X		X				X	Every 3-6 months	
Flow cytometry for immune correlates[a]	X	X	X	X	X	X	X	X	X	X	
Pregnancy test (If indicated)	X										
Echocardiogram or MUGA	X				X						
Chest X-ray	X		X		X		X		X		
Concomitant medication and AE assessment	X	X	X	X	X	X	X	X	X		X
Toxicity assessment		X	X	X	X	X	X	X	X		X

[a] Flow cytometry will be run on days 1 and 14 of each cycle of blinatumomab (and day 28 for the last cycle). Flow will be performed on peripheral blood specimens or on BM specimens (when BM assessment is otherwise mandated per protocol).

** Patients will be followed every 3 months (±1 month) for 1 year and then every 6 months (±3 months) for survival via brief phone call, even after being taken off-protocol.

6.3 Follow-up

Thirty days after last dose of the study drugs concomitant medication and AE assessment will be recorded. This may be done over the phone with a member of the study staff. The phone call should last about 10 minutes.

6.4 Long-term Follow-up

Patients will be followed periodically every 3 months (±1 month) for 1 year and then every 6 months (±3 months) for survival via brief phone call, even after being taken off-protocol.

7. EFFICACY AND SAFETY ASSESSMENTS

7.1. CRITERIA FOR RESPONSE

1. Complete Remission (CR): Normalization of the peripheral blood and bone marrow with 5% or less blasts in normocellular or hypercellular marrow with a granulocyte count of 1 x 10^9/L or above, and platelet count of 100 x 10^9/L. Complete resolution of all sites of extramedullary disease is required for CR.
2. Complete remission without recovery of counts (CRi): Peripheral blood and marrow results as for CR, but with incomplete recover of counts (platelets < 100 x 10^9/L; neutrophils < 1 x 10^9/L).
3. Partial Response (PR): As above for CR except for the presence of 6-25% marrow blasts.
4. MRD negativity: Absence of detectable leukemia using multiparameter flow cytometry with a sensitivity of ≤0.01%
5. Relapse and resistant disease will be defined based on morphological assessment of bone marrow and peripheral blood.

7.2. EVALUATION OF TOXICITY

Toxicities will be graded according to the NCI Common Toxicity Criteria for Adverse Event Reporting Version 4.0. The toxicity of the regimen will be monitored continuously during the course of the study.

7.3. CRITERIA FOR REMOVAL FROM THE STUDY

1. Adverse events that are not manageable with dose adjustments and/or optimal medical management, or that, in the opinion of the investigator, pose an unacceptable risk for the patient
2. Non-compliance by the patient with protocol requirements
3. Failure to achieve CR, CRi or PR after 1 cycle, unless the patient is thought to have derived clinical benefit, after discussion with the PI
4. Disease progression.
5. Patient death
6. Patient decision (e.g. withdrawal of consent)
7. Investigator decision, if it is deemed in the best interest of the patient

Patients who discontinue the study treatments will have a follow-up visit 30 days (± 10 days) after the last dose of the study drugs. For patients who cannot come for the clinic visit, a phone call to assess for any side-effects will be done.

7.4 DEFINITION OF STUDY END-POINTS

1. Relapse-free survival is the time from documented complete response until relapse or death.

2. **Event-free survival** is the time from the first day of treatment until any failure (resistant disease, relapse, or death).

3. Overall response rate is defined as the percentage of patients achieving CR or CRi

4. Overall survival is defined as the time from the first day of treatment to time of death from any cause.

8. REPORTING REQUIREMENTS

It is the responsibility of the PI and the research teams to ensure serious adverse events are reported according to the Code of Federal Regulations, Good Clinical Practices, the protocol guidelines, and Institutional Review Board (IRB) policy.

An adverse event is the appearance or worsening of any undesirable sign, symptom, or medical condition occurring after starting the study drug even if the event is not considered to be related to study drug. Medical conditions/diseases present before starting study drug are only considered adverse events if they worsen after starting study drug. Abnormal laboratory values or test results constitute adverse events only if they induce clinical signs or symptoms, are considered clinically significant, or require therapy.

The investigator (or physician designee) is responsible for verifying and providing source documentation for all adverse events and assigning the attribution for all adverse events for subjects enrolled.

Adverse event reporting will be as per the NCI criteria and the MDACC Leukemia Specific Adverse Event Recording and Reporting Guidelines (Appendix C).

The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0 will be utilized for adverse event reporting. (http://ctep.cancer.gov/reporting/ctc.html).

Investigator Communication with Amgen:

All suspected unexpected serious adverse reactions (SUSARs) related or possibly related to blinatumomab and their follow-up reports must be reported to Amgen within 24 hours of submission to the regulatory agency, IRB or IEC. A copy of any safety report involving an Amgen drug (e.g. blinatumomab) submitted to the regulatory agency, IRB or IEC, must be faxed to Amgen, within 24 hours of such submission.

The Investigator must report all pregnancies and pregnancies occurring in the partner of a patient participating in the study or potential infant exposure through lactation within 10 calendar days of sponsor's awareness to Amgen.

9. OUTSIDE PHYSICIAN PARTICIPATION DURING TREATMENT

a. MDACC Physician communication with the outside physician is required prior to the patient returning to the local physician. This will be documented in the patient record.

b. A letter to the local physician outlining the patient's participation in a clinical trial will request local physician agreement to supervise the patient's care

c. Protocol required evaluations outside MDACC will be documented by telephone, fax or e-mail. Fax and/or e-mail will be dated and signed by the MDACC physician, indicating that they have reviewed it.

d. Changes in drug dose and/or schedule must be discussed with and approved by the MDACC physician investigator, or their representative prior to initiation, and will be documented in the patient record.

e. Documentation to be provided by the local physician will include drug administration records, progress notes, reports of protocol required laboratory and diagnostic studies and documentation of any hospitalizations.

f. The home physician will be requested to report to the MDACC physician investigator all life threatening events within 24 hours of documented occurrence.

g. Patients should receive all induction and consolidation at MDACC but may receive maintenance therapy by an outside physician.

h. Patients will return to MDACC every 3-6 months during maintenance for evaluation.

10. STATISTICAL METHODOLOGY

This is a single-arm phase II study of blinatumomab plus mini-hyper-CVD chemotherapy in patients with relapsed or refractory ALL. Patients will receive blinatumomab plus mini-hyperCVD for up to 8 cycles (4 cycles of Blinatumomab), plus 2 years of maintenance therapy. The primary endpoint of this study is the event free survival (EFS) where events defined as no response, loss of response, or death.

Up to 44 patients will be enrolled in the study with an estimated accrual rate of 2 patients per month. The additional follow up will be 12 months after the last patient enrolls in the study. The primary objective is to evaluate event-free survival (EFS). The study will monitor futility and toxicity.

Efficacy endpoint

Historically, the 6-month EFS rate is 30% (with standard chemotherapy). That corresponds to median EFS of about 3.5 month. It is expected that the 6-month EFS rate will increase by 20% under this study treatment. That is to say, the target 6-month EFS rate is 35%. That corresponds to median EFS of about 4 month.

It is expected that the median EFS time will be 3.5 month or higher under the study treatment. If the trial continues to maximum accrual of 44 patients and maintains sufficient follow-up to observe 25 events (no response, loss of response, or death) with a median EFS of 3.5 month, then a 95% credible interval for median EFS would extend from 2.4 to 5.2 months.

Futility monitoring

Futility will be monitored for the total of 44 patients. We will monitor EFS using the methods of Thall et al. (2005), and we will stop enrolling patients to the study if we have reason to believe that the median EFS is less than 3.5 months. The trial will be stopped if Pr(lamda(E) > lamda(S) | data from the trial) < 0.08, where lamda(S) is the median EFS for the treatment with an inverse gamma distribution IG(14.25, 46.38), which corresponds to a mean of 3.5 months and a standard deviation of 1 month. The prior for lamda(E) is assumed to be IG(2.12, 3.93), corresponding to a mean of 3.5 months and a larger standard deviation of 10. Futility monitoring will start once 5 patients have been enrolled. Assuming an accrual rate of 2 per month and an additional follow-up of 12 months, the operating characteristics for the futility stopping rule under various true scenarios are summarized in the following table 10.1, with results based on 5000 simulations.

Table 10.1: Operating characteristics for futility monitoring

Scenario Name	True Median TTE (Months)	Pr(Stop Early)	Mean # Patients	Trial Duration (Months)
scenario 1	5.00	0.03	42.84	33.54
Scenario 2	3.90	0.10	40.64	32.42
Scenario 3	3.50	0.17	38.70	31.48
Scenario 4	3.00	0.32	34.49	29.44
Scenario 5	2.50	0.63	26.81	25.73

Toxicity monitoring

Toxicity monitoring will be conducted in cohort size of 4, starting from the 1st patient. Denote the probability of toxicity by p(T), where toxicity is defined as any grade 3-4 clinically relevant non-hematologic toxicity or a serious adverse event that is felt to be drug related (Common Terminology Criteria for Adverse Events CTCAE version 4.0). We assume as a priori, p(T) ~ beta (0.6, 1.4). The trial will be stopped if Pr(p(T)> 0.30 | data) >0.90. That is, we will stop the trial for new patient enrollment if at any time during the study we determine that there is more than 90% chance that the toxicity rate is more than 30%. Stopping boundaries corresponding to this toxicity monitoring rule are shown in Table 10. 2 below. The operating characteristics for toxicity monitoring are summarized in Table 10. 3.

Table10. 2. Toxicity stopping boundaries in cohort size of 4.

Number of patients	Stop the trial if there are this many patients having toxicities
4	3-4
8	5-8
12	6-12
16	8-16
20	9-20
24	11-24
28	12-28
32	14-32
36	15-36
40	16-40

Table 10.3. Operating characteristics for toxicity monitoring in cohort size of 4 (max sample size = 44).

True toxicity rate	Prob(stop the trial early)	Average sample size
0.10	0.004	43.83
0.2	0.05	42.25
0.25	0.13	39.99
0.3	0.27	36.14
0.35	0.48	30.73
0.4	0.69	24.57
0.5	0.95	14.20
0.6	1.00	8.76

Multc Lean Desktop (version 2.1) was used to generate the toxicity stopping boundaries and OneArmTTEsimulator Version 3.0.6 were used for EFS futility simulation.

Futility monitoring in CTC website

The Department of Biostatistics will provide and maintain a website ("Clinical Trial Conduct": https://biostatistics.mdanderson.org/ClinicalTrialConduct/) for futility monitoring on this study. The Clinical Trial Conduct website resides on a secure server, and access is gained through usernames and passwords provided to personnel responsible for enrolling patients and updating patient data. The website is accessed through a browser using secure socket layer (SSL) technology. Personnel responsible for enrolling patients on trials, which includes the principal investigator(s), research nurse(s), and data coordinator(s), will be trained by members of the Department of Biostatistics in the use of the trial website; the importance of timely updating of follow-up times and recording of events will be emphasized in training.

Analysis method

Data analysis will be performed using SAS or SPLUS, as appropriate. The median EFS time will be estimated by Bayesian posterior estimates, along with the 95% credible intervals. Toxicity will be reported by type, frequency and severity. Highest toxicity grades per patient per course will be tabulated for selected adverse events and laboratory measurements. Demographic and laboratory results, i.e., MRD, will be summarized using descriptive statistics, including means with standard deviations, or medians with ranges, histograms and box-plot. Overall response rate will be estimated along with 95% confidence interval. Overall response rate will be compared between subgroups (e.g. different patient characteristics) by Fisher's exact test and minimal residual disease difference will be assessed by Wilcoxon rank test, respectively. The overall survival (OS) is defined as the time from treatment start till death or last follow-up. The duration of response (DOR) is defined as the number of days from the date of initial response (PR or better) to the date of first documented disease progression/relapse or death, whichever occurs first. EFS, OS, and DOR will be estimated using the Kaplan-Meier method, and log rank test will be used to compare the differences in the time-to-event variables between subgroups (e.g. different patient characteristics).

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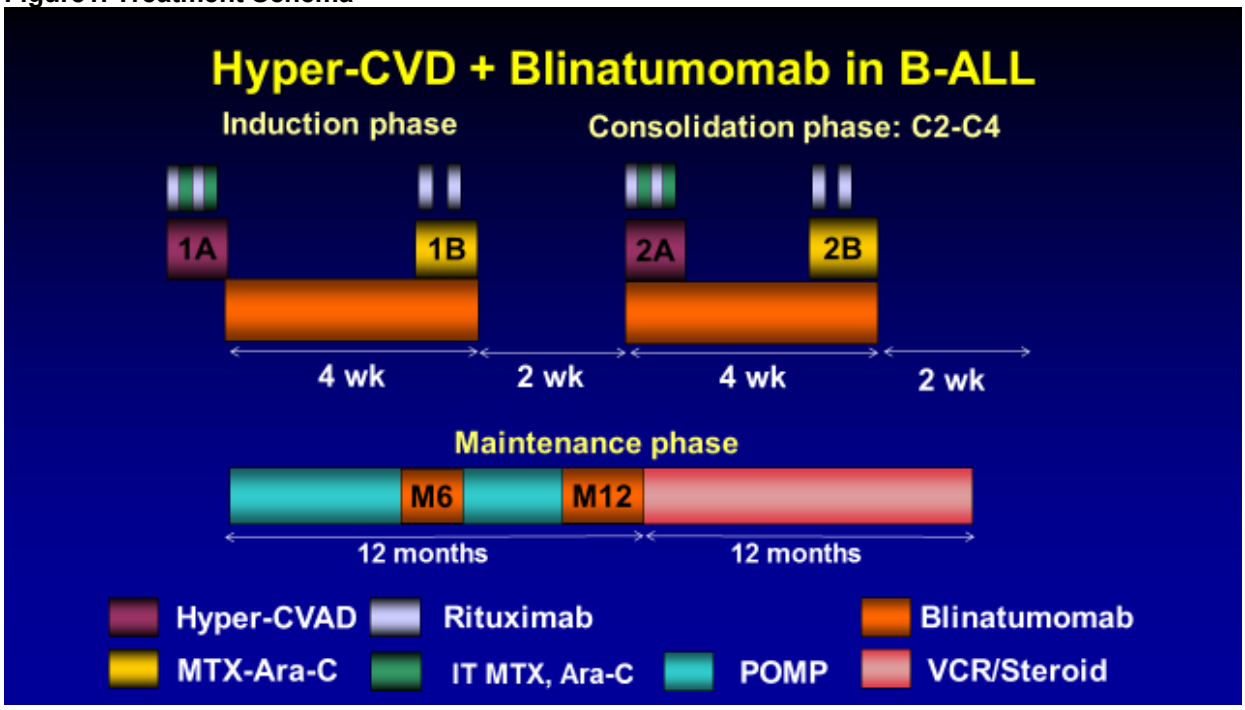
Figure1. Treatment Schema



Appendix A

Immune correlatives- A study of blinatumomab in combination with mini-hyper-CVD. (Katy Rezvani MD, PhD; MD Anderson Cancer Center)

Physiologic control of immunologic responses.
Physiologic mechanisms exist to restrain immune responses, both to maintain self-tolerance and to prevent excessively exuberant pathogen-directed responses, which may be harmful to the host. CTLA4, a negative regulator of immune function, is not expressed on resting T-cells, but is rapidly up-regulated on CD4+ cells after T-cell activation. It competitively binds to CD80/86, to prevent CD28-mediated co-stimulation, which otherwise usually occurs upon engagement of the TCR with antigen presented on HLA class-I molecules. Programmed death-1 (PD1) is also upregulated, although more slowly, after TCR engagement and inhibits TCR-mediated effector function.

Immune response to malignant cells.
Malignant cells often over-express normal antigens or express "neoantigens" produced due to the numerous somatic mutations which occur within these cells; both can be recognized by the immune system. However, malignant cells successfully evade immune surveillance and killing. A major mechanism used to evade the immune response is the overexpression of immune checkpoint molecules, such as the PD1 ligand PD-L1. Tumor infiltrating lymphocytes express high levels of PD-1, analogous to "exhausted" T-cells seen in chronic viral infection. Numerous studies in solid tumors and hematologic malignancies have demonstrated that checkpoint inhibition, using antibodies directed at CTLA-4, PD-1 or both, is highly effective at breaking immune tolerance of the tumor, resulting in durable and often profound immunologic disease-control.

Preliminary data regarding the role of immune checkpoint molecules in ALL.

To understand mechanisms of resistance with bispecific antibody therapy, we studied samples from patients with B-ALL treated with blinatumumab for relapse after allogeneic stem cell transplantation (unpublished data); a representative FACS plot is presented in **Fig 1**. Prior to therapy with blinatumumab, PD1 was overexpressed on the patient's CD4+ and CD8+ T cells, relative to normal controls, and was further up-regulated during blinatumomab therapy. This was associated with a rapid and significant reduction in T cell effector function, as measured by effector cytokine production and CD107a degranulation (a measure of T cell cytotoxicity).

Based on these data, **we hypothesize that the function of T-cells directed to CD19-positive targets by blinatumomab may be modulated by the expression levels of exhaustion markers on T cells and their respective ligands on blasts.**

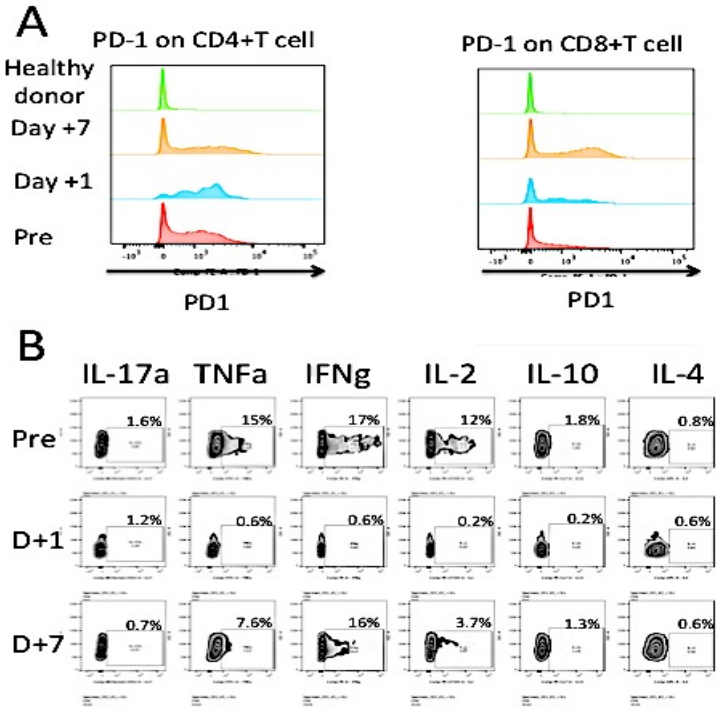


Figure 1. Blinatumomab therapy is associated with significant upregulation in PD1 on CD4+ and CD8+ T cells with a resultant impairment in T cell effector function.

Research plan.

We will use 17 color multiparameter flow cytometry to investigate in detail the immunologic effects of blinatumomab on T cell function and expression of checkpoint molecules, in patients with ALL. We will collect peripheral blood (PB) samples at baseline and on days 1 and 14 of each cycle of blinatumomab (and day 28 for the last cycle)- A total of 4 cycles of blinatumomab are planned in induction/consolidation and 2 additional cycles during maintenance; thus we will analyze samples from 13 timepoints per patient). We will also analyze paired bone marrow (BM), collected at timepoints clinically mandated in the protocol.

To determine how blinatumomab influences the recovery, maturation and function of CD3+ T cells, expression of checkpoint molecules, and the balance between Th1/Th2/Th17 cells and regulatory T cells (Tregs) during therapy, we will perform the following analyses at the timepoints mentioned above: (i) immunophenotypic analysis of naïve, stem-like memory, central and effector memory CD4+ and CD8+ T cells (CD3, CD4, CD8, CD45RA, CD45RO, CCR7, CD62L, CD95, IL-7Rα, IL-2Rβ); Tregs will be defined as CD4+CD25hiCD127loFoxp3+; (ii) measure markers of T cell activation (CD69, CD25, CD137), proliferation (Ki-67); anergy and exhaustion (PD1, CTLA-4, LAG-3, TIM-3, CD160, TIGIT, 2B4, CD57); (iii) Perform T cell functional studies: T cells will be activated via CD3/TCR and CD28 ligation or PMA/inomycin activation and differentiated according to their cytokine profile into Th1 (CD4+IFN-γ+), Th2 (CD4+IL-4+), Th17 (CD4+IL-17+) and Tr1 (CD4+IL-10+). Th1/Th2 ratio will be assessed during blinatumomab therapy.

Numbers and phenotypic characteristics of T cells in PB and BM, at baseline and during therapy, will be correlated with therapeutic response. In addition, we will evaluate CD19 and PD-L1 expression on leukemic blasts using flow cytometry at baseline and at response evaluation, to determine the role of CD19 downregulation or loss and PDL1 upregulation in therapeutic resistance.