

SOP No: AHP 68(b)

SUBJECT: NOD-SCID ALL Model
Adoptive Immunotherapy of Leukemia
Cytotoxic T Lymphocytes +/- Antigen Presenting Cells

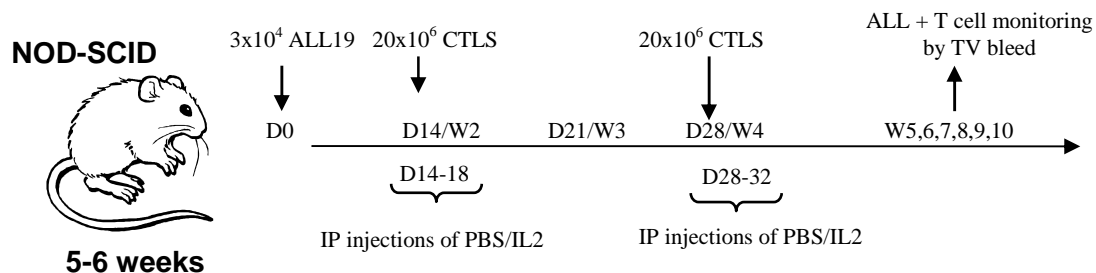
POLICY: This procedure must be performed by an experienced operator

PRECAUTIONS: Gloves, mask, long-sleeve gown, closed in shoes
The clinical score of the mice can increase rapidly in the week preceding sacrifice.

EQUIPMENT: 29g needles
1ml syringes
phosphate buffered saline
Leukaemia-specific CTL's
100,000 IU/mL Human IL2 stock
Antigen Presenting Cells

PROCEDURE:

1. Induce leukemia by injecting – dose is determined by experimental design.
2. Inject 20,000 IU (200 μ L) IL2, IP into each mouse three hours prior to receiving CTLs.
3. Leukemia-specific CTL's are injected I/V in 200 μ l – dose and schedule is determined by experimental design. See diagram below for an example of CTL scheduling
4. Inject 20,000 IU/day I/P for the next four days following CTL injection.
5. Inject 200 μ L APC's I/V or I/P dose and schedule of APC administration is determined by the experimental design. See diagram below for an example of APC scheduling.



Experimental Design:

| Cohort | CTLs | APC | APC: dose | APC: route of admin. | Comment |
|---------------|-------------|------------|---------------------------|-------------------------------------|---|
| 1 | ✓ | x | | | Effect of CTLs alone |
| 2 | ✓ | ✓ | 1x10⁶ | IP | Effect of low dose APC given IP |
| 3 | ✓ | ✓ | 1x10⁶ | IV | Effect of low dose APC given IV |
| 4 | ✓ | ✓ | 2 x 10⁶ | IP | Effect of high dose APC given IP |
| 5 | ✓ | ✓ | 2 x 10⁶ | IV | Effect of high dose APC given IV |

RECOMMENDATIONS:

Mice are monitored 2-3 times per week as per the score sheet noting weight, fur texture, posture, activity and presence of a palpable spleen.

Mice are monitored for engraftment of leukaemia into the peripheral blood from at least week 3 post injection of leukaemia. Once human leukaemia cells (HuCD45/19/10+) are detected in the peripheral blood around 3-5% mice are monitored and scored daily.

Human leukaemia (HuCD45/19/10+) cells above 10% of total CD45+ cells (mouse + human) are indicative of a heavy level of leukaemia engraftment in the spleen. The average number of human leukaemia cells harvested from the spleen is 200-300x10⁶ cells.

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REVISED:

REFERENCES