

## Appendix C

SCIENTIFIC ABSTRACT

A replication incompetent, recombination incompetent retroviral vector will be used to mark autologous peripheral blood and marrow cells removed and stored at the time of cytogenetic remission or re-induction of chronic phase in Philadelphia chromosome positive CML patients who have developed blast crisis or accelerated phase and have been reinduced into chronic phase following Daunomycin, high-dose Ara-C, and GM-CSF therapy. We estimate that between  $1.2 \times 10^4$  and  $1.2 \times 10^6$  leukemic marrow blast cells and  $4.2 \times 10^4$  and  $4.2 \times 10^6$  leukemic peripheral blood blastic CML cells will be marked with NEO in the autologous cells used for transplant. It is not known how many CML blastic leukemia cells are present in the systemic circulation following induction of the chronic phase or a cytogenetic remission by Daunomycin, high-dose Ara-C, and GM-CSF. The number must be low judging from the duration of remissions which may last greater than one year following autologous transplant. We will look for the number of NEO-marked cells using a methylcellulose late progenitor colony culture system and a PCR assay for the NEO gene used previously by Cornetta, et al (Human Gene Therapy 1:15-30, 1990), and Dr. Malcolm Brenner. In the CML cells under analysis, the percent of these NEO-marked cells which are leukemic can be determined by a PCR assay for the bcr-abl mRNA positive CML cells. We will also use PCR to determine the number of different vector integration sites and characterize the relapsed cells with respect to the presence of polyclonal or clonal vector integration sites. These studies will clarify if relapse arises from the leukemic CML blast cells present in the autologous cells infused after TBI, VP-16, and cytoxan (if polyclonal CML NEO-marked blastic cells appear at the time of relapse), or if residual systemic disease contributes to relapse (if none of the CML leukemic blasts at the time of relapse contain the NEO gene).