Abstract
c-Myb is a critical transcription factor required for hematopoietic cell lineage fate selection, proliferation, and maturation. It is a well validated therapeutic target for human leukemias, and for solid tumors, in particular breast and colon cancer, as well. Previous studies conducted in our lab, and others, suggest that inhibition of c-Myb with antisense oligonucleotides can inhibit leukemia cell growth in vitro, and in mouse models. These studies have led us to believe that small molecule inhibitors of c-Myb would be effective anti-leukemic agents, and likely useful for other tumors as well. Further, since small molecule inhibitors would not be plagued by the delivery problems which bedevil nucleic acid drugs, we hypothesize that they would be much more efficient. Herein, we propose to develop a cell-based assay for screening a small molecule compound library for the presence of lead c-Myb inhibitors that might be further modified for efficiency, specificity, and safety as needed. We have already established a c-Myb reporter cell line in which the well characterized c-myb-responsive mim-1 promoter is used to drive the expression of firefly luciferase, while the Myb-independent SV40-promoter is used to control the expression of Renilla luciferase. A c-Myb specific siRNA, which decreased c-Myb mRNA by 80%, also led to 90% reduction of luciferase levels, but only 16% reduction of Renilla activity within 48 hours after transfection, indicating the specificity of the mim-1 reporter cell line. We also found that the histone deacetylase inhibitor trichostatin A (TSA) can potently reduce luciferase level, but not Renilla activity, by inhibiting c-Myb expression. The c-Myb reporter cell line can therefore be used to screen for compounds with c-Myb inhibition activity using TSA as a positive control.