Table S1. Basic hematology of mice with different genotypes

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Hematocrit (%)</th>
<th>RBC (x10⁶/ml)</th>
<th>WBC (x10³/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ku70 +/+ (n=6)</td>
<td>52.2±1.8</td>
<td>2,708±507</td>
<td>8,847±2,087</td>
</tr>
<tr>
<td>Ku70 +/- (n=4)</td>
<td>53.8±0.5</td>
<td>2,805±233</td>
<td>8,733±2,004</td>
</tr>
<tr>
<td>Ku70 -/- (n=6)</td>
<td>53.8±1.8</td>
<td>2,477±191</td>
<td>1,685±638</td>
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</tbody>
</table>
Figure S1. (A). Ku70-/− mice are able to produce myeloid cells. Absolute myeloid (Mac1+) cell numbers were calculated by multiplying total WBCs by the percentage of Mac1+ in PB (n=6). No significant difference was observed.

(B) Ku70-/− mice display reduced BM cellularity. BM cells were isolated from WT and Ku70-/− mice (n=6 each genotype), and mononuclear cells were counted using hemacytometer. Error bars indicate the SD, and significance was determined by a two-tailed t test. asterisk, P<0.01.
Figure S2. BM microenvironment in Ku70-/− mice is able to support both lymphogenesis and myelogenesis. 5x10^6 WT (CD45.1) BM cells were transplanted into WT (CD45.2, n=5) and Ku70-/− (CD45.2, n=3) mice without any conditioning. Sixteen week post transplantation, donor derived reconstitution of both myeloid and lymphoid lineages was analyzed by FACS.
Figure S3. Ku70-/- HSCs are sensitive to 5-FU treatment. BM cells from WT and Ku70-/- mice were transplanted into lethally irradiated WT recipients. 12 weeks post-transplantation, the recipients (n=5 each group) were treated with 5-FU (90mg/Kg) weekly, the arrows indicate treatment time. Survival of the recipients was monitored. The recipients reconstituted with Ku70-/- BM cells were treated twice (the first two doses), the recipients reconstituted with WT BM cells received three doses.
Figure S4. (A). LSK cells were sorted into 96-well plates at 80 cells/well, and cultured in IMDM in the presence of cytokine cocktails containing 20ng/ml IL-3, 50ng/ml SCF, 50ng/ml Flt-3L and 50ng/ml Tpo for 6 days. At the end of culture, cell numbers were counted using hemacytometer, the expansion fold was calculated.

(B) At the end of culture, apoptosis was determined by Annexin V and DAPI staining, and apoptosis fraction (AnnexinV+, DAPI-) was quantitated. Data are representative of three independent experiments. Error bars indicated SD, and significance was determined by a two-tailed t test.
Figure S5. (A). Peripheral blood was collected by retro-obital bleeding method, and white blood cell (WBC) numbers were counted using Coulter. TgBcl2/Ku70-/- mice showed slight but not significant increase in WBC numbers compared to Ku70-/- mice. Error bars indicate the SD, and significance was determined by a two-tailed t test. asterisk, P<0.01.

(B) BM cells were isolated from WT, Ku70-/-, TgBcl2 and TgBcl2/Ku70-/- mice (n=6 each genotype), and mononuclear cells were counted using hemacytometer. Error bars indicate the SD, and significance was determined by a two-tailed t test. asterisk, P<0.01.
Figure S5C. Overexpression of Bcl2 does not correct the SCID phenotypes in Ku70-/− mice. Peripheral blood from age-matched WT, Ku70-/−, TgBcl2, and TgBcl2/Ku70-/− mice (WT, TgBcl2, n=5 each; Ku70-/−, TgBcl2/Ku70-/−, n=3 each) were collected, myeloid and lymphoid cells were analyzed by FACS.
Figure S5. (D). Bcl2 overexpression did not restore the HSC/progenitor pools in Ku70-/- mice. BM cells from age-matched WT, Ku70-/-, TgBcl2, and TgBcl2/Ku70-/- mice (WT, Ku70-/-, n=6 each; TgBcl2, n=5, TgBcl2/Ku70-/-, n=4) were collected and analyzed, and absolute numbers of LSK and SLAM-LSK were calculated. The reduction of LSK and SLAM-LSK cell numbers was not altered by the overexpression of Bcl2. Significance was determined by a two-tailed t test. asterisk, P<0.01.
Figure S6. Ku70-/- HSCs are hypersensitive to radiation. WT and Ku70-/- mice were irradiated with 2 Gy radiation, and 24 hours post radiation, BM cells were isolated and analyzed by FACS for LSK and SLAM-LSK pools. Ku70-/- BM was hypersensitive to radiation, 24 hours after 2Gy radiation, LSK fraction was almost completely diminished.