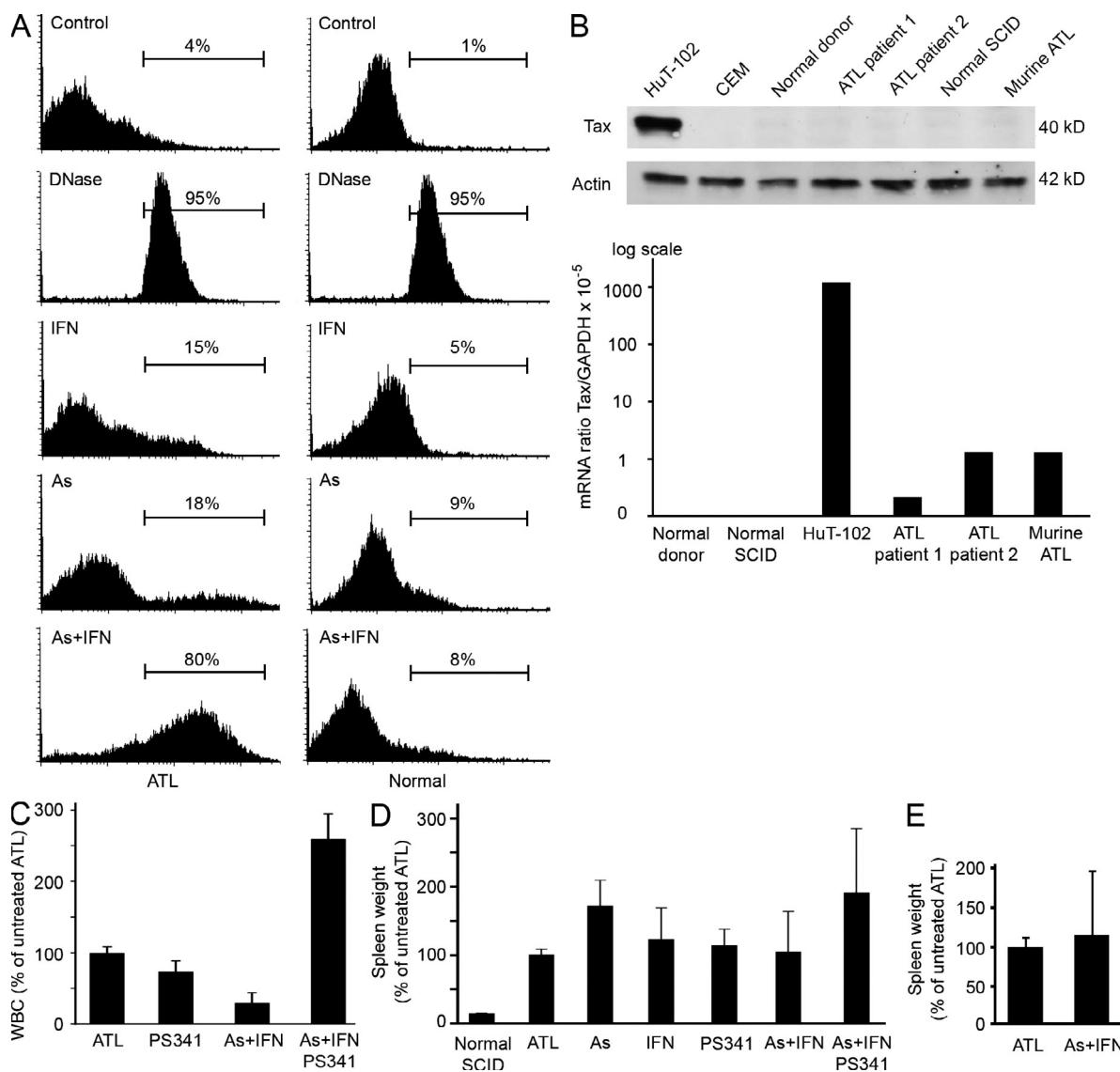
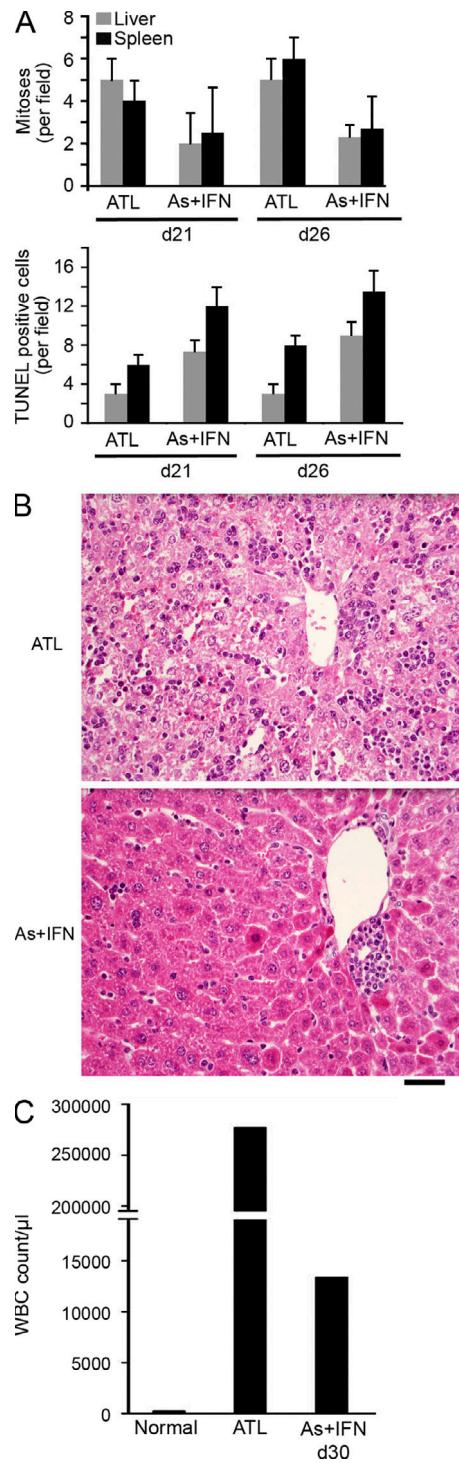


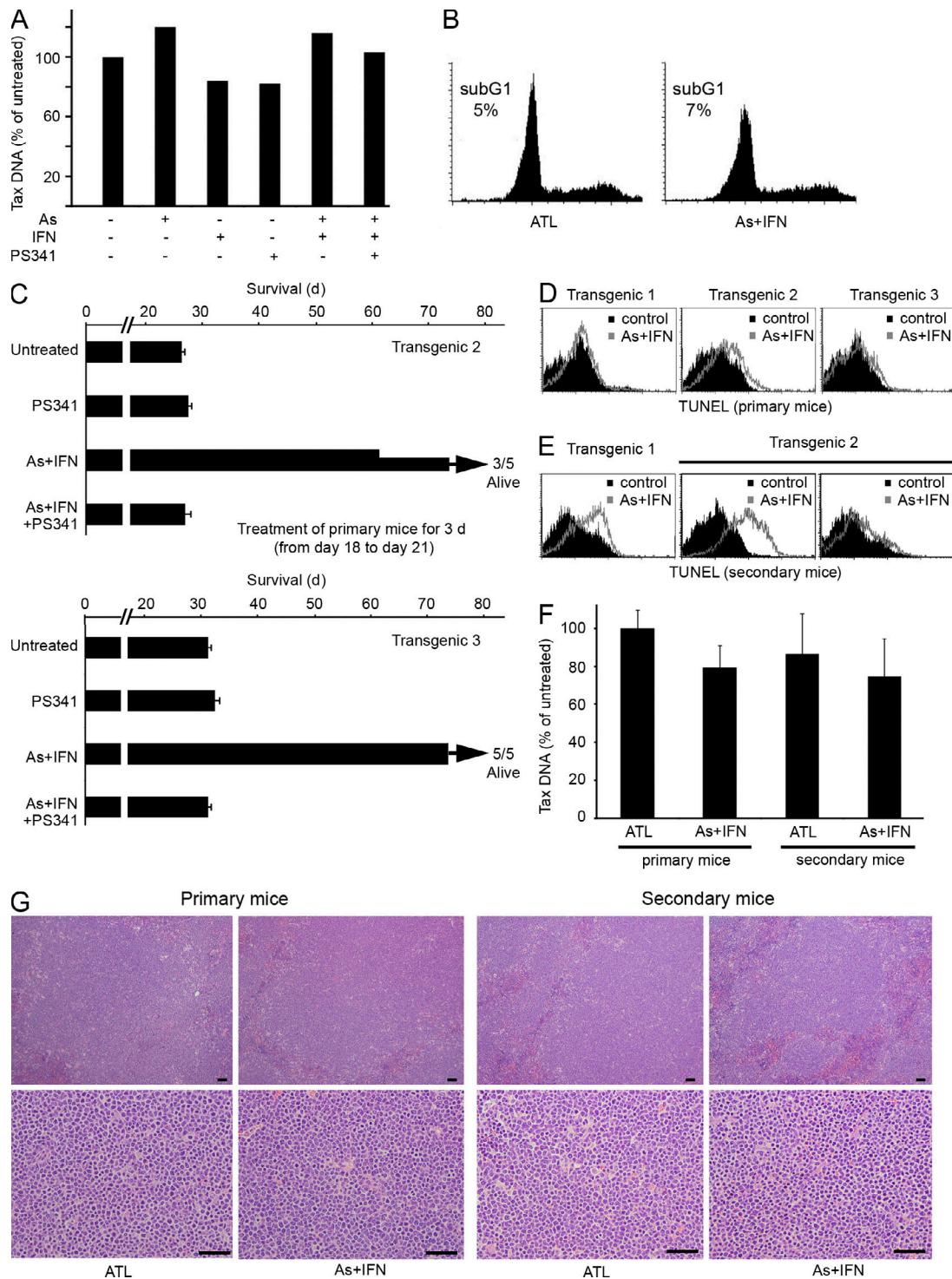
## SUPPLEMENTAL MATERIAL

El Hajj et al., <http://www.jem.org/cgi/content/full/jem.20101095/DC1>

**Figure S1.** Effect of short therapy with IFN- $\alpha$  and  $\text{As}_2\text{O}_3$ . (A) TUNEL staining of ATL cells (left) or healthy lymphocytes (right) after overnight ex vivo treatment with DNase, 1,000 U  $\text{ml}^{-1}$  IFN- $\alpha$ , and/or  $10^{-6}$  M  $\text{As}_2\text{O}_3$  (As). Control indicates untreated cells. (B, top) Western blot analysis of Tax and actin protein expression in human HuT-102 cells, human HTLV-I-negative CEM T cells, PBMCs from a normal donor, circulating leukemia cells from two patients with acute ATL, PBMCs from a normal SCID mouse, and murine ATL cells. (bottom) Tax messenger RNA (mRNA) expression in PBMCs from a normal donor, a normal SCID mouse, HuT-102 cells, primary human ATL cells, and murine Tax transgenic ATL cells. (C) Effect at day 18 after inoculation of 3-d IFN- $\alpha$  +  $\text{As}_2\text{O}_3$  treatment on circulating leukemia cell numbers ( $n = 6$  for each condition). ATL denotes untreated animals, and PS-341 denotes the proteasome inhibitor. Graph represents pooled data from three different transgenic mice. (D) Effect at day 18 after inoculation of 3-d IFN- $\alpha$  +  $\text{As}_2\text{O}_3$  treatment on the spleen weight ( $n = 6$  for each condition). (E) Effect at day 12 after inoculation of a 6-d IFN- $\alpha$  +  $\text{As}_2\text{O}_3$  treatment on the spleen weight ( $n = 5$  for each condition). Graph represents pooled data from three different transgenic mice. (C–E) Error bars show mean  $\pm$  SD.



**Figure S2.** Effect of prolonged therapy with IFN- $\alpha$  and As<sub>2</sub>O<sub>3</sub>. (A) Mitoses or TUNEL-positive cell counts per field at 600 magnification in the spleen and liver at different treatment time points. While indicative, the observed differences did not reach statistical significance. Error bars show mean  $\pm$  SD. (B) Effect of IFN- $\alpha$  + As<sub>2</sub>O<sub>3</sub> (As) treatment on intraparenchymal micrometastases in the liver (day 26). Bar, 50  $\mu$ m. (C) WBC counts at day 30 of IFN- $\alpha$  + As<sub>2</sub>O<sub>3</sub> therapy ( $n = 3$  for each condition).



**Figure S3. Loss of leukemia-initiating activity accounts for the therapeutic effect of the IFN- $\alpha$  +  $\text{As}_2\text{O}_3$  combination.** (A) Quantitative PCR analysis of *Tax* DNA in spleen ATL cells obtained 18 d after inoculation from primary mice treated as indicated for 3 d. (B) Effect of 3-d IFN- $\alpha$  +  $\text{As}_2\text{O}_3$  (As) treatment on the cell cycle 18 d after inoculation. (C) Effect of 3-d treatment of primary mice from two additional founders on the survival of secondary recipient mice of  $10^6$  spleen ATL cells ( $n = 5$  for each condition). (D) TUNEL analysis of splenocytes of primary mice after 3-d treatment with IFN- $\alpha$  +  $\text{As}_2\text{O}_3$ . (E) TUNEL analysis of splenocytes at the time of death from leukemia of secondary recipients of  $10^6$  ATL cells from primary mice. Primary mice were either untreated (control) or had received 3-d IFN- $\alpha$  +  $\text{As}_2\text{O}_3$  with or without PS-341. Secondary mice were not treated. (F) Quantitative PCR analysis of *Tax* DNA in spleen ATL cells from primary mice treated as indicated for 3 d or from secondary recipients of ATL cells from untreated or IFN- $\alpha$ /As $_2$ O $_3$ -treated primary mice ( $n = 3$  for each condition). (C and F) Error bars show mean  $\pm$  SD. (G) Histological analysis of the spleen of primary and secondary mice as described in F at low (top) and high (bottom) magnification. Bars, 100  $\mu$ m.

**Table S1.** Follow up of ATL patients in complete remission after stopping treatment

Diagnosis	Treatment	Follow up after stopping treatment
Chronic ATL	IFN- $\alpha$ + zidovudine	Progression after 1 mo
Chronic ATL	IFN- $\alpha$ + zidovudine	Progression after 3 mo
Acute ATL	IFN- $\alpha$ + zidovudine	Progression after 3 mo
Lymphoma	IFN- $\alpha$ + zidovudine	Progression after 5 mo
Chronic ATL	IFN- $\alpha$ + zidovudine	Progression after 11 mo
Chronic ATL	As + IFN- $\alpha$ + zidovudine	Progression after 1 mo
Chronic ATL	As + IFN- $\alpha$ + zidovudine	Progression after 5 mo
Chronic ATL	As + IFN- $\alpha$ + zidovudine	Progression after 6 mo
Chronic ATL	As + IFN- $\alpha$ + zidovudine	CCR after 7-mo follow up
Chronic ATL	As + IFN- $\alpha$ + zidovudine	CCR after 7-mo follow up
Chronic ATL	As + IFN- $\alpha$ + zidovudine	CCR after 18-mo follow up

CCR, continuous complete remission.