SUPPLEMENTARY METHODS

Mice

Six- to 8-week-old female C57BL/6, nude, and nonobese diabetic severe-combined immunodeficient (NOD/SCID) mice were purchased from Central Lab. Animal Inc. (Seoul, Korea) and used under the protocol IACUC (KUIACUC-2009-126). All animal procedures were performed in accordance with recommendations for the proper use and care of laboratory animals.

DNA constructs

For the generation of the pMSCV/mouseApi5 (mApi5) and pMSCV/human Api5 (hApi5) constructs, the DNA fragments encoding the mApi5 were amplified from cDNAs isolated from TC-1/P3 (A17) cells using a set of primers: 5'-GCAGATCTATGCCGACGGTGGAGGAGC-3' and 5'-GCGAATTCGTATTTCCCCTGAAGGCTC-3'. DNA fragments encoding hApi5 were amplified from cDNAs of CUMC6 tumor cells by PCR using a set of primers: 5'-GCAGATCTATGCCGACAGTAGAGGAGCT-3' and 5'-GCGAATTCCTACTTCCCCTGAAGGTC-3'. The amplified DNAs were cloned into the BglII/EcoRI sites of pMSCV retroviral vector (Clontech, Mountain View, California). For the generation of pMSCV/SCT E7, DNA fragment encoding the immunodominant E7 aa49–57 peptide and flanking AgeI/NheI restriction enzyme sites was made by annealing two single-stranded oligo-nucleotides 5'-CCGGAGAGCCCATTACAATATTGTAACCTTT-3' and 5'-CTAGTCTCGGGTAATGTTATAACATTGGAAA-3'. It was then cloned into pIRES-E6-Kb(26) using AgeI/NheI sites to replace the E6 epitope, generating pIRES-E7-2m-Kb. E7-2m was then amplified with PCR using pIRES-E7-2m-Kb as the template and a set of primers,

5'-AGATCTAGAGCCCATTACAATATTGTAACCTTT-3' and 5'-CTCGAGGGTGGTGGAGGTAGTGGCGGGGCGATGGCTCCGCGCACGCTGC-3'.

Cells

The production and maintenance of TC-1/P0 and TC-1/P3 (A17) cells, expressing endogenous E7 on D^b, has been described previously and used as targets for E7-specific CTL(27). Human tumor cell lines, HeLa, CaSki, MCF-7, MDA231, DU145, PC-3, HepG2, HCT116, A549, A375 (HLA-$A2^+$/MART-1/Melan-A^-)were purchased from American Type Culture Collection (ATCC, USA) and SNU-C4 and SNU-368 cell lines were purchased from Korean Cell Line Bank (KCLB, Seoul, Korea). 526mel (gift from M. Lotze) is a human melanoma cell line (HLA-$A2^+$) which expresses MART-1/Melan-A. The identities of cell lines were confirmed by short tandem repeat (STR) profiling by IDEXX KOREA and used within 6 months of testing.

TC-1/P0/no insert, TC-1/P0/Api5, 293D^b/no insert, 293D^b/hApi5, A375/no insert, A375/hApi5and HeLa/SCT-E7 cell lines were generated using the constructed pMSCV/mAPI5, pMSCV/hAPI5, pMSCV/SCT-E7 or pMSCV/no insert DNAs. The HEK293D^b and HeLa/SCT-E7 cell lines expressing single-chain trimer (SCT) of MHC class I (H-2D^b) linked to an HPV-16 E7 immunodominant CTL epitope (aa 49-57) was generated to evaluate immune resistance against HPV-16 E7-specific murine cytotoxic T lymphocyte (CTL) killing in the human cancer cells(28). A375/no insert, A375/hApi5, and 526mel cells were used as targets for human MART-1-specific T cells (clone KKM), established *de novo* from the peripheral blood of a patient with metastatic melanoma. The MART-1-specific KKM CTL clone was generated using methods developed in the Yee Lab (21, 29) and is specific for the HLA-A2-restricted epitope (M27) of the tumor-associated antigen, MART-1 (Supplementary Fig. 3).

***In vitro* and *in vivo* delivery of siRNAs**

Synthetic small interfering RNA (*siRNA*) specific for *Gfp, mApi5, hAPI5, mErk2, mBim, hFGF2, and hPKCδ* were purchased from Invitrogen (Carlsbad, California); Non-specific *Gfp* (green fluorescent protein), 5'-GCAUCAAGGUGAACUUCAA-3' (sense), 5'-UUGAAGUUCACCUUGAUGC-3' (antisense); *mApi5*, 5'-UUACUGUGCUCUUAUAAGGAGG-3' (sense), 5'-CCUCCUUCU UAUAAGAGCACAGUAA-3' (antisense); *hAPI5*, 5'-GGGUUGUUCAGCCAAAUACUU -3' (sense), 5'-AAGUAUUUGGCUGAACAACCC-3' (antisense), *mErk2*, 5'-AUGUCGAACUUGAAUGGUGCUUCGG-3' (sense); 5'-CCGAAGCACCAUUCAAGUUCGACAU-3' (antisense); *mBim*, 5'-GGAGGGUGUUUGCAAAUG-3'(sense); 5'-GGAGGGUGUUUGCAAAUG-3'(antisense), *hFGF2*, 5'-GAGAGAGGAGUUGUGUCUA-3' (sense), 5'-UAGUCACAACUCCUCUCUC-3' (antisense), *hPKCδ*, 5'-CUCAUGGUACUUCCUCUGU-3' (sense), 5'-ACAGAGGAAGUACCAUGAG-3' (antisense). For *in vitro* delivery, 5×10^5 A17, HeLa, A375/API5, or 526mel cells were transfected with 300 pmol of the synthesized siRNAs using Lipofectamine 2000 (Invitrogen, Carlsbad, California) according to the manufacturer's instructions. For systemic *in vivo* delivery of siRNA into tumor cells, we prepared chitosan nanoparticle (CNP) as described previously (30). Briefly, tripolyphosphate (0.25% w/v) and siRNA (1 μg/μl) were added in RGD-Chitosan solution, and the siRNA/RGD-Chitosan-nanoparticle incubated at 4℃ for 40 minutes and collected by centrifugation at 13,000 rpm for 40 minutes at 4℃. The pellet was washed with sterile water.

Western blot Analysis

A total of 5×10^5 cells were used as described previously(35). The primary antibodies against phospho-AKT (Ser473), AKT, phospho-ERK(T202/Y204), ERK, p38 MAPK, phospho-FGF Receptor(Y653/654), FGF Receptor, phospho-PKCδ (Y311), PKCδ, BCL-$_{XL}$, BID, BIM, BAD, phospho-BAD (S136), and XIAP were purchased from Cell Signaling and used at 1:1000 dilution. Antibodies against dual phospho-p38 MAPK (1:1000, Stressgen, Victoria, Canada); BCL-2, BCL-$_{XL}$, BAX, API5 (1:1000, Santa Cruz Biotechnology, Dallas, Texas); BAK (1:1000, BD Biosciences, San Jose, California) and E7 (provided by Dr. Ju-hong Jun, Seoul National University, Korea) were used for Western blotting followed by the appropriate secondary antibodies conjugated with horseradish peroxidase. Immunoreactive bands were visualized by enhanced chemiluminescence (ECL, Elpis Biotech, Daejeon, Korea) reaction.

***In vivo* tumor treatment experiments**

On the day of tumor challenge, tumor cells were harvested by trypsinization, washed once with Opti-MEM®I (Gibco, Grand Island, New York) and resuspended in 1ml of Opti-MEM®I to desired concentration for subcutaneous injection. Mice were challenged s.c. with 1×10^5 TC-1/P3 (A17), TC-1/P0/no insert orTC-1/P0/Api5 cells per mouse in the left leg. Where indicated, CNP containing *GFP* siRNAs or *API5* siRNAs (5 μg/mouse) were injected intravenously at day 7, 10, and 14 following tumor cell inoculation. At day 14 when mice were injected with the last injection of chitosan nanoparticles, one set of mice (5 per group) were treated with 2×10^6/mouse of adoptively transferred E7-specific $CD8^+$ T cells intravenously through the tail vein(36). In a separate group of mice, thermosensitive chitosan hydrogel(37) containing 50 μM PD98059, 100 nM Rottlerin or DMSO was intra-tumorally injected. One day after hydrogel treatment, mice were either adoptively transferred with 2×10^6 E7-specific $CD8^+$ T cells/mouse or intravenously injected with normal saline, as a

control. Tumor volumes from TC-1/P0/Api5 tumor were recorded on a regular basis for up to 25 days following immunization. For the human CTL/tumor *in vivo* assays, NOD-SCID mice were inoculated subcutaneously with 5×10^6 526mel cells/mouse. After 10days, siRNA chitosan nanoparticles targeting either *GFP*, *PKCδ, or API5* (5 μg/mouse) was injected intravenously for three consecutive days. Approaching the thirteenth day, mice received an adoptive transfer of 1×10^7 MART-1-specific CTL by intravenous injection. siRNA chitosan nanoparticles and MART1-specific CTL adoptive transfer was repeated every 7 days along with 5000 unit IL-2(38) and tumor volume was analyzed as bar graphs at day indicated in the figure.