ISP-15-2  SIRT1 increases proliferation, chemo-resistance and invasiveness of ovarian carcinoma cells

Shinshu University
David Mvunta, Tsutomu Miyamoto, Ryoichi Asaka, Hisanori Kobar, Yasushi Yamada, Hirofumi Ando, Shotaro Higuchi, Koichi Ida, Hiroyasu Kashima, Tanri Shiozawa

[Objective] Sirtuin 1 (SIRT1), initially identified as a longevity gene, is considered to protect cells against age–related diseases including cancer. In contrast, several previous studies suggested oncogenic roles of SIRT1. We previously reported that the overexpression of SIRT1 was an independent poor prognostic factor for ovarian carcinoma (OvCa). The present study aimed to clarify the functional roles of SIRT1 in OvCa. [Methods] The OvCa cell lines (ES2, TOV21G and RMG1) were used to examine the functions of SIRT1 on cell proliferation and chemo-sensitivity (WST-1 assay), apoptosis (Annexin V assay), migration and invasion (invasion assay). The effect of SIRT1 on tumor growth was examined using OvCa xenograft in nude mice. [Results] Knock-down of SIRT1 by siRNA/shRNA decreases cell proliferation (TOV21G and RMG1, P<0.05), cisplatin–resistance (ES2, P<0.05). Overexpression of SIRT1 by cDNA facilitated the chemo–resistance against cisplatin (P<0.05) and paclitaxel (P<0.05), invasion (P<0.05), and reduced apoptosis (P<0.05) in ES2 cells. These effects in ES 2 cells were canceled out by the addition of selective SIRT1 inhibitor, EX527. SIRT1 over–expression resulted in increased tumorigenicity in vivo. [Conclusion] SIRT1 may be involved in the acquisition of aggressiveness and chemo–resistance in OvCa, hence indicating a novel candidate of therapeutic target.

ISP-15-3  Cancer cell and oncogene alter subset populations of T and dendritic cells in the tumor microenvironment of disseminated ovarian cancer model

The University of Tokyo

[Objective] Tumor microenvironment (TME) is modulated by cancer. Little is known about how oncogenes affect adaptive immunity in the TME. We here demonstrate how immune cells in the TME are modulated by cancer dissemination and K-ras. [Methods] Murine ovarian cancer cell line, ID8, or Kras–transduced ID8 (ID8–Kras) was injected into the peritoneal cavity and ascites production was monitored. Subset populations of T cells and dendritic cells (DC) in spleen or ascites from both mice were examined for CD8/CD4, CD11c/mPDCA, regulatory T-cell (Treg)/CD4 using flow cytometry. [Results] Ascites production was accelerated in ID8–Kras mice compared with ID8. CD8/CD4 ratio in ascsites was increased in either ID8 or ID8–Kras mice compared with control, however there was no difference in the ratio between ID8 and ID8–Kras mice. CD11c+mPDCA DC subset (conventional DC) was increased in ascites of ID8 mice. However, increase of the DC subset was cancelled in the ID8–Kras mice. Neither ID8 nor ID8–Kras injection altered Treg proportion in ascites. There was no difference in population of T cells and DCs in the splenocytes. [Conclusion] CD11c+mPDCA DC subset in the TME was increased with ID8 cancer dissemination, followed by the increased CD8–dominant immune response. K-ras–positive cancer cell may inhibit conventional DC migration followed by adaptive immune response to tumor.

ISP-15-4  LATS1 phosphorylation at serine 909 by Gα13 is involved in YAP activation in ovarian cancer cells

Kyushu University
Hiroshi Yagi, Tatsuhiro Ohgami, Ichiro Onoyama, Akimasa Ichinoe, Kenzo Sonoda, Kiyoko Kato

[Objective] G protein–coupled receptors (GPCRs) and their ligands have been implicated in the progression of human cancers. In the previous study, we demonstrated that the activation of heterotrimeric G protein, Gα13/15, promotes cell proliferation through YAP activation in ovarian cancer cells. We herein evaluated the underlying mechanisms by which Gα13/15 activate YAP. [Methods] To examine the signaling pathway exclusively regulated by Gα13, we employed a synthetic biology approach using a GPCR activated solely by artificial ligands (RASSLs). The activation of LATS1 and YAP regulated by Gα13 was evaluated by Western blot. The expression of LATS1 and two YAP–targeted genes, CTGF and CYR61, was examined by real time PCR. [Results] Gα13 induced YAP dephosphorylation and augmented the gene expression of CTGF and CYR61 in ovarian cancer cells. LATS1 phosphorylation at threonine 1079 decreased, conversely, serine 909 phosphorylation significantly increased. Interestingly enough, LATS1 protein level decreased markedly, though LATS1 mRNA level did not change. In the presence of proteasome inhibitor, MG132, Gα13–mediated LATS1 protein degradation was suppressed. [Conclusion] Proteasome–dependent LATS1 degradation after serine 909 phosphorylation induced by Gα13 might contribute to YAP activation in ovarian cancer cells.