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Author(s)	Miyatake, Yukiko; Kuribayashi-Shigetomi, Kaori; Ohta, Yusuke; Ikeshita, Shunji; Subagyo, Agus; Sueoka, Kazuhisa; Kakugo, Akira; Amano, Maho; Takahashi, Toshiyuki; Okajima, Takaharu; Kasahara, Masanori
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Visualising the dynamics of live pancreatic microtumours self-organised through cell-in-cell invasion

Yukiko Miyatake^{1,*}, Kaori Kuribayashi-Shigetomi^{2,3,*}, Yusuke Ohta¹, Shunji Ikeshita¹,

Agus Subagyo^{3,4}, Kazuhisa Sueoka³, Akira Kakugo⁵, Maho Amano⁶,

Toshiyuki Takahashi⁷, Takaharu Okajima³, Masanori Kasahara¹

Running title: Dynamics of live pancreatic microtumours

¹ Department of Pathology, Faculty of Medicine and Graduate School of Medicine,

Hokkaido University

² Institute for the Advancement of Higher Education, Hokkaido University

³ Graduate School of Information Science and Technology, Hokkaido University

⁴ Creative Research Institution Sousei, Hokkaido University

⁵ Faculty of Science, Hokkaido University

⁶ Research Development Section, Hokkaido University

⁷Department of Pathology, Hokkaido Gastroenterology Hospital

*Correspondence to:

Yukiko Miyatake, Ph.D

Department of Pathology, Hokkaido University Faculty of Medicine, Kita-15, Nishi-7,

Kita-ku, Sapporo 060-8638, Japan

Phone: +81-11-706-5050; Fax: +81-11-706-7825

E-mail: yukimiya@med.hokudai.ac.jp

Kaori Kuribayashi-Shigetomi, Ph.D

Graduate School of Information Science and Technology, Hokkaido University, Sapporo

Kita-14, Nishi-9, Kita-ku, Sapporo, 060-0814, Japan

Phone: +81-11-706-7591; Fax: +81-11-706-7591

Phone: kaorik@ist.hokudai.ac.jp

SUPPLEMENTARY MOVIE INFORMATION

Supplementary Movie 1. PCI-55 cells that were cultured on the micro/nanoplate. Sequential images were acquired every 15 min.

Supplementary Movie 2. 3D-reconstructed image. PDAC microtumours anchored to multiple microislands showed morphological polarity.

Supplementary Movie 3. PCI-55 cells that were anchored to a microisland through cellcell invasion. Sequential images were acquired every 15 sec and partially every 30 sec.

Supplementary Movie 4. Cell-in-cell structures on the micro/nanoplate are reversible. Sequential images were acquired every 1min.

Supplementary Movie 5. Treatment with 1 μ m nocodazole prevented PCI-55 microtumours from anchoring to the micro/nanoplate. Sequential images were acquired every 5 min.

Supplementary Movie 6. Live- and dead-cell monitoring by Annexin V (green) and EthD-1 (red). Most of the nocodazole-treated microtumours showed anchorage-independent spheroidal morphology, and many live PCI-55 cells were scattered outside of the disrupted microtumours and still survived. Sequential images were acquired every 5 min.

Supplementary Movie 7. Live PCI-55 microtumours actively moving to catch debris in the culture medium. Sequential images were acquired every 15 min.

Supplementary Movie 8. Live PCI-55 microtumours stretching to catch external floating apoptotic cells and dead-cell debris with huge lamellipoedia. Sequential images were acquired every 3 min.

Supplementary Movie 9. Many floating green-fluorescent nanobeads drifted to near anchorage-dependent PCI-55 microtumours. Sequential images were acquired every 1 min.

Supplementary Movie 10. When anchorage-dependent PCI-55 microtumours were harvested and cultured back in a culture dish they stopped elongating and began removing PS from their surfaces. Sequential images were acquired every 5 min.

Supplementary Movie 11. When PCI-55 microtumours were cocultured with KHYG-1, a human natural killer (NK) cell line, they were not attacked by KHYG-1 cells. Sequential images were acquired every 2 min 30 sec.

Supplementary Movie 12. PDAC microtumours that accepted Annexin V+/EthD-1+ PDAC cells accumulated Annexin V on their external surfaces, whereas EthD-1 accumulated inside the cells. Sequential images were acquired every 5 min. **Supplementary Movie 13.** When live PCI-55 microtumours treated with UV produced dead-cell debris, the mature PCI-55 microtumours aggressively devoured this dead-cell debris. Sequential images were acquired every 3 min.