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Stage IV Pancreatic Cancer Patient Treated With FOLFIRINOX Combined With Oral Methioninase: A Highly-Rare Case With Long-term Stable Disease

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Abstract. *Background: Pancreatic cancer is one of the most recalcitrant cancers, and more effective therapy is needed. Pre-clinical studies have shown that patient-derived orthotopic xenograft (PDOX) mouse models of pancreatic cancer are effectively treated with oral recombinant methioninase (o-rMETase). Case Report: A 62-year-old woman diagnosed with stage IV pancreatic cancer was treated with the combination of 5-fluorouracil/leucovorin, irinotecan, and oxaliplatinum (FOLFIRINOX) every two weeks and o-rMETase twice a day as a supplement. The patient was also on a low-methionine diet. Disease progression was monitored by CA19-9 and computed tomography. The patient initially responded to FOLFIRINOX, shown by a great reduction in CA19-9 levels, with tumor shrinkage shown by computed tomography. The patient began taking o-rMETase and went on a low-methionine diet one year after diagnosis which she has maintained without side effects for 7 months. The patient's CA19-9 level and tumor size remain stable 19 months after diagnosis. The patient is alive and has maintained a high performance status. Historical data show that less than 5% of stage IV pancreatic-cancer patients on FOLFIRINOX have stable disease 1.5 years after diagnosis. Conclusion: The combination of o-rMETase and FOLFIRINOX may be synergistic in stage IV pancreatic cancer.*

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Key Words: Pancreatic cancer, stage IV, FOLFIRINOX, oral recombinant methioninase, supplement, combination therapy, stable disease, CA19-9, methionine addiction, Hoffman effect.

Pancreatic cancer is the 3rd leading cause of cancer-related death in the United States and 4th in Japan (1, 2). Pancreatic cancer often progresses without symptoms, and most patients are not indicated for surgery because of tumor invasion of adjacent organs or distant metastases. Chemotherapy for advanced cases currently comprises FOLFIRINOX [5-fluorouracil (5-FU)/leucovorin, irinotecan, and oxaliplatinum] or gemcitabine (GEM) + nab-paclitaxel as first-line standard of care. However, these regimens only lead to 9-11 months median survival after initiation of therapy (3). This is a small improvement of previous standard of care first-line therapy with GEM alone, which resulted in approximately 6 months survival. More effective therapies are urgently needed for pancreatic cancer (3).

Methionine addiction is a fundamental and general hallmark of cancer (4-6) and is termed the Hoffman effect (7). Pre-clinical studies have demonstrated that methionine addiction is a potent target for pancreatic cancer (8). Methionine addiction of cancer is due to a much higher than normal methionine requirement of cancer cells, which appears to be due to excess transmethylation reactions (9-11).

Methionine-restricted cancer cells selectively arrest in late S/G_2 of the cell cycle (12). Therefore, antimetabolites (13, 14) and alkylating agents (15-17) tend to show synergistic effects with methionine restriction. Thus, methionine restriction can be synergistic with chemotherapy in pancreatic cancer, especially FOLFIRINOX, which uses an antimetabolite (5-FU) and an alkylating agent (oxaliplatinum). 5-FU has been shown to have a synergistic effect combined with methionine restriction in gastric cancer in the clinic (18).

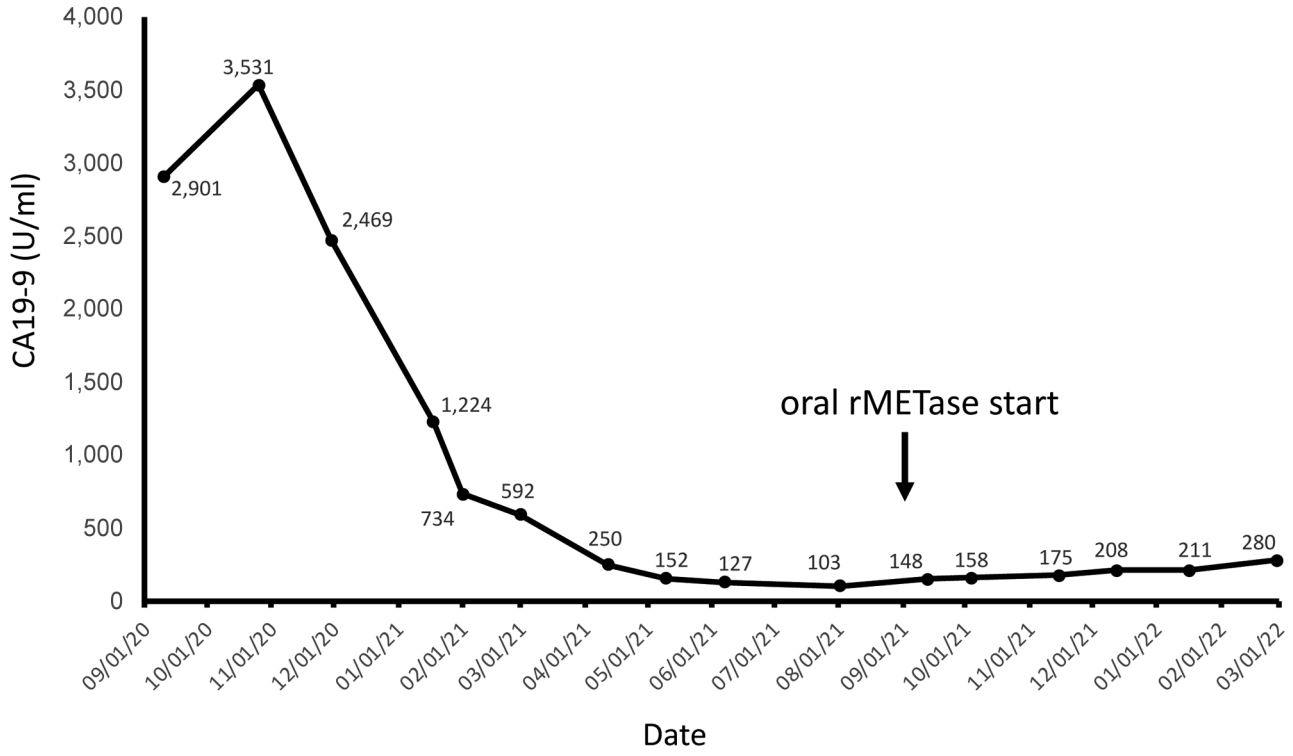

Figure 1. *Time course of patient's CA19-9.*

Our laboratory has developed recombinant methioninase (rMETase) to target methionine restriction (19). Recently we have demonstrated that rMETase can be administered orally effectively in the clinic, where it is effective as a supplement (20-22).

In the present case report, a 62-year-old female with pancreatic cancer treated with FOLFIRINOX and oral rMETase, as a supplement, currently has stable disease 1.5 years after diagnosis as Stage IV, a highly-rare case.

Materials and Methods

rMETase production and formulation. rMETase was produced by fermentation of recombinant *E. coli* transfected with the methioninase gene from *Pseudomonas putida*. Methioninase was purified using a heat step at 60 degrees, polyethylene glycol precipitation, column chromatography with diethylaminoethyl (DEAE)-Sepharose FF, with high yield (23, 24).

Methionine restriction. The patient had methionine restriction with a low-methionine diet according to the Nutritional Oncology Research Institute (NORI) protocol (25) and received oral recombinant methioninase (o-rMETase) twice a day at a dose of 250 units as a supplement.

Case Report

A 62-year-old female was diagnosed with pancreatic cancer on September 10, 2020, upon examination for back pain. Computed tomography (CT) showed a 30×30 mm low-density tumor in the pancreatic tail. The tumor marker CA19-9 was extremely high (2,901 U/ml). Regarding pathological findings, endoscopic ultrasound-guided fine-needle aspiration (EUS-FNA) was performed and showed ductal adenocarcinoma. After a central venous port was installed, the patient started chemotherapy with FOLFIRINIOX (5-FU 2,400 mg/m^2 continuous injection and 400 mg/m^2 bolus injection; irinotecan 150 mg/m^2 intravenous injection; oxaliplatinum 85 mg/m^2 intravenous injection, every two weeks). FOLFIRINOX therapy was effective: CA19-9 gradually decreased from 2,901 U/ml to 103 U/ml, and the pancreatic tumor shrunk over time (Figure 1 and Figure 2). Twelve months after the start of FOLFIRINOX treatment, the patient started methionine restriction with o-rMETase and a low-methionine diet from September 1, 2021. The patient developed an allergy to oxaliplatinum at the 15th and 21st course of FOLFIRINOX therapy. Therefore, FOLFIRI therapy started, and has continued since November 15, 2021. She was also negative for any BRCA mutations. With FOLFIRI chemotherapy and methioninase, the patient's low CA19-9 level has remained stable (Figure 1) and tumor size has continued to shrink (Figure 2). The patient had no side effects on FOLFIRINOX, or FOLFIRI, and o-rMETase and maintains a high performance status.

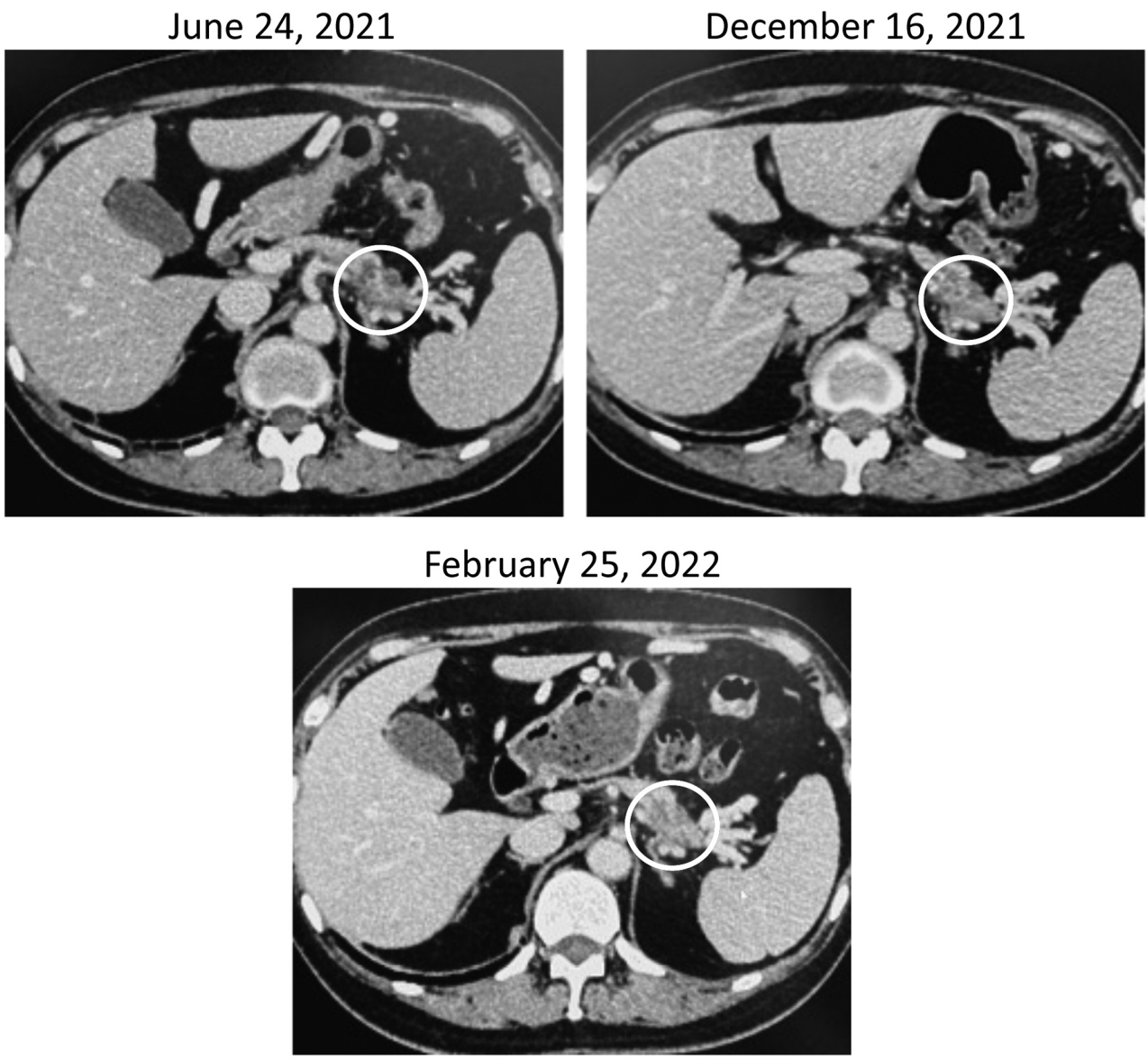

Figure 2. *Computed tomography findings of pancreatic cancer before and after o-rMETase was started on September 1, 2021.*

Discussion

The median progression-free survival (PFS) of the FOLFIRINOX therapy reported by a clinical trial is 6.4 months and the 1.5-year PFS rate is under 5% (3).

Methionine addiction is a fundamental and general hallmark of cancer cells (4-6, 26-28). Even though cancer cells can synthesize methionine from homocysteine at normal or higher rates, they require a large amount of exogenous methionine due to excess transmethylation reactions (9-11, 29).

Eight cancer patients participated in a Phase I study of dietary methionine restriction. Of these patients, prostate cancer and renal cell cancer patients showed stable disease or partial response, with no side effects (30). Methionine-restricted cancer cells selectively arrest in late S/G_2 of the cell cycle (12); therefore, the combination of cell-cycle-specific chemotherapy with methionine restriction has shown clinical efficacy. In Phase I and Phase II trials, a methionine-free diet with cystemustine showed partial responses and stable disease in melanoma and glioma patients (31, 32). A methionine-free diet and FOLFOX also showed effectiveness for metastatic colorectal cancer (33). Methionine-free total parenteral nutrition (TPN) with 5-FU for gastric cancer resulted in significantly better efficacy than TPN containing methionine and 5-FU (18).

In a pilot Phase I trial, intravenous injection of rMETase rapidly decreased methionine to undetectable levels and showed no side effects after administration over a 24 hour period (34,35). Subsequently, long-term studies of intravenously-administered rMETase showed rapid methionine depletion in macaque monkeys but anaphylaxis occurred unless the enzyme was PEGylated (36, 37). A breakthrough for rMETase occurred in 2018 when we reported that oral rMETase (o-rMETase) was effective in a pancreatic cancer cell line and patient-derived tumors in mice (38-41) o-rMETase could overcome gemcitabine resistance in a pancreatic-cancer cell line (39). Subsequently, we reported the efficacy of oral rMETase in breast cancer, sarcoma, and prostate cancer in PDOX models (20-22, 42-45) and clinically as a supplement in prostate cancer patients (20-22).

In the present pancreatic-cancer case, methionine restriction with o-rMETase and a low-methionine diet showed apparent synergistic efficacy in combination with FOLFIRINOX and FOLFIRI. The patient had stable disease and remains alive with a high-performance status 19 months after diagnosis. In pancreatic cancer patients on FOLFIRINOX alone, less than 5% of the cancer patients had stable disease 1.5 years after diagnosis. Future studies will include more individual case studies with o-rMETase as supplement and subsequent clinical trials. Methionine addiction is the fundamental basis of malignancy and is targeted by methionine restriction (46,47).

Conflicts of Interest

The Authors declare no competing interests in relation to this work.

Authors' Contributions

YK, TT, and RMH wrote the paper. QH produced methioninase. JY, NM, and YA revised the manuscript. CH provided the patient data.

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