Official title: Stereotactic body radiation therapy plus pembrolizumab and trametinib versus stereotactic body radiation therapy plus gemcitabine for locally recurrent pancreatic cancer after surgical resection: an open-label, randomized phase 2 trial

NCT number: NCT02704156

Document state: 5th January 2021

1. Participants and eligibility

The following inclusion and exclusion criteria would be employed to preserve high internal validity and reduce risks of SBRT, gemcitabine, immunotherapy or targeted therapy-related adverse effects.

Inclusion criteria:

1. Histologically confirmed pancreatic ductal adenocarcinoma with unequivocal first progression after surgery followed by chemotherapy

2. Without any immunotherapy or targeted therapy

3. A life expectancy of >3 months

4. ECOG of 0 to1

5. Age of 18 years or older

6. Analysis of surgical specimens showed KRAS mutations and positive immunohistochemical staining of PD-L1, including IC1 or TC1, IC2 or TC2, IC3 or TC3.

7. Blood routine examination: Absolute neutrophil count (ANC) $\geq 1.5 \times 10^9$ cells/L, leukocyte count$\geq 3.5 \times 10^9$ cells/L, platelets $\geq 70 \times 10^9$ cells/L, hemoglobin ≥ 8.0 g/dl

8. Liver and kidney function tests: Albumin > 2.5 g/dL, total bilirubin < 3 mg/dL, creatinine < 2.0 mg/dL, AST<2.5 × ULN (Upper Limit of Normal) (0-64U/L), ALT<2.5 × ULN(0-64U/L)

9. INR < 2 (0.9-1.1)

10. Ability of the research subject or authorized legal representative to understand and the willingness to sign a written informed consent document.

Exclusion criteria:

1. Prior immunotherapy or targeted therapy

2. Evidences of metastatic disease confirmed by chest CT or FDG PET-CT

3. Contraindication to receiving immunotherapy, targeted therapy or SBRT

4. ECOG ≥2

5. Age <18 years

6. Analysis of surgical specimens showed KRAS wild type or negative immunohistochemical staining of PD-L1, including IC0 or TC0.

7. Secondary malignancy

8. Abnormal results of blood routine examinations and liver and kidney and coagulation tests

9. Patients with active inflammatory bowel diseases or peptic ulcer

10. Gastrointestinal bleeding or perforation within 6 months

11. Heart failure: NYHA III-IV

12. Respiratory insufficiency

13. Women who are pregnant

14. Participation in another clinical treatment trial while on study

15. Patients in whom fiducial implantation was not possible

16. Inability of the research subject or authorized legal representative to understand and the willingness to sign a written informed consent document.

2. Analysis of KRAS mutations and immunohistochemical staining of PD-L1

All patients' surgical specimens underwent immunohistochemical staining of PD-L1, evaluated by the SP142 immunohistochemistry assay (Ventana Medical Systems, Tucson, AZ). PD-L1 TC expression was scored as the percentage of PD-L1 TC, which was stratified as TC3 ≥ 50% or TC2 ≥ 5% but < 50% or TC1 ≥ 1% but <5%. PD-L1 IC expression was scored as the percentage of tumor area stained positive, which was classified as IC3 ≥ 10% or IC2 ≥ 5% but < 10% or IC1 ≥ 1% but <5%. *KRAS* mutations were analyzed by PCR amplification and direct sequencing of exon 2. K-RAS Mutation Kit (DxS, Manchester, UK) was used in the case of failure of direct sequencing. Additionally, Restriction Length Fragment Polymorphism method was used for further confirmation. All immunochemistry assays and variants were reviewed by a molecular pathologist.

3. Doses of pembrolizumab, trametinib and gemcitabine

In the SBRT plus pembrolizumab and trametinib group, 200mg pembrolizumab was administered intravenously every 3 weeks and 2mg trametinib was given orally once daily. The pembrolizumab dose could be temporarily interrupted due to toxicity. And

pembrolizumab could be resumed if the adverse events had been reduced to grade 1 or 0. The dose of trametinib could be reduce to 1.5mg then 1mg daily to manage toxic effects, while a further reduction below 1mg once daily was not allowed. If one of the drugs were withheld, the other drug could be continued. Treatment continued until disease progression, unacceptable toxicity, or patient or investigator decision to discontinue. Delivery of pembrolizumab and trametinib was initiated one week after SBRT.

In the SBRT plus gemcitabine group, patients received intravenous gemcitabine ($1000mg/m^2$) on day 1 and 8 of each 21-day cycle for eight cycles in the absence of disease progression. Dose modifications depended on the grade of adverse effects. For grade 3 or more toxicity, treatment was delayed until adverse effects resolved to grade 2 or less, and gemcitabine could be reduced to $750mg/m^2$. Similarly, treatment also continued until disease progression, unacceptable toxicity, patient refusal to participate in the study, investigator decision to discontinue or intercurrent illness that prevented further administration of treatment. Chemotherapy was also initiated one week after SBRT.

4. CT simulation for treatment planning of SBRT

Each patient should fast for at least eight hours before the simulation. Vacuum bags are customized with patients in the supine position, according to the patient's body shape for immobilization during Cyberknife. SBRT is delivered *via* Cyberknife, an image-guided frameless stereotactic robotic radiosurgery system (Accuray Corporation,

Sunnyvale CA), that consists of a linear accelerator mounted on a robot arm with six degrees of freedom. In this system, the confluence of a large number of non-isocentric pencil beams permits the treatment of irregularly shaped target volumes with rapid dose falloffs. Cyberknife tracking system automatically compensates for the alignment offset and patient motions by adjusting the treatment isocenter. In addition, a CT based treatment planning system is used at our institution. Then, plain CT and an enhanced pancreatic parenchymal CT are performed for radiation treatment planning and target delineations. CT images are acquired under breath hold (preferably end-expiratory). Pretreatment diagnostic imaging would be co-registered to the simulation CT in cases where the patient is unable to tolerate intravenous contrast. The scan range includes the whole pancreas, at least 10 cm above and below the tumor. Spiral CT is performed with a slice thickness of 1.5 mm, and images are reconstructed in slices of 1.5 mm at most. Intravenous contrast enhancement is performed with an injection of 80-100 ml of iodixanol, a flow rate of 2.5 ml/sec, and a delay of 45-55 seconds; as acquired for the pancreatic parenchymal phase.

5. Registration and tracking of SBRT

The co-registrations of biphasic CT images are based on fiducials and anatomical (spinal) fusion. Before CT simulation, fiducials should be implanted using endoscopic ultrasound or CT guidance. This is pivotal for treatment planning and delivery. CT simulation will be performed 7-10 days after fiducial placement. This time interval is required to avoid early fiducial marker displacement or migration. In order to improve

the accuracy of the treatment planning, the recommended number of implanted fiducials is preferably close to 3-5. As a result, given that fiducials could simulate the spatial location and displacement of the tumor, which is attributable to respiration, motion tracking should be performed by means of the correlation with these seeds; and fiducial markers render the Synchrony system equipped in Cyberknife feasible. This allows for respiratory motion tracking during irradiation. Nevertheless, patients with high risk of bleeding, abdominal infection, pancreatitis or pancreatic fistula are contradictory to several fiducial implants. Hence, one fiducial plus X-sight spine and Synchrony Tracking technique would be alternatively used. Before treatment, direct digital radiography images of the spine would be applied to detect 6-D errors; and this would be subsequently corrected for X-sight spine tracking on the patient's positioning. This would enable fiducial tracking during treatment.

6. Treatment planning and target delineation

After CT simulation, CT images are transferred to the workstation where the target volumes are contoured by an attending radiation oncologist. Gross tumor volume (GTV) is delineated as a radiographically evident gross disease by contrast CT acquired from the portal-venous phase. At the discretion of the physician, clinical target volume (CTV) encompassing areas of the potential subclinical disease spread is also designated. In most cases, the CTV equals GTV. A 2-5 mm expansion margin is included to determine the planning target volume (PTV). When the tumor is adjacent to critical organs, the expansion of CTV should be avoided. Therefore, an individualized treatment plan

would be developed based on tumor geometries and locations. Ninety percent of PTV should be covered by the prescription dose. The prescription isodose line is limited to 70-75%, which would restrict the tumor D_{max}. If dose level violates the constraint of SBRT, the patient would be considered as ineligible for this trial. The prescribed dose of PTV varies from 35-40Gy/5f with a single dose of 7-8Gy. In particular, these doses would be reduced if the tumor is approximately one-third or more of the duodenum or stomach circumference, or if the tumor abuts the bowel in only one area, as determined by the relationship of the tumor to the duodenum in axial, coronal and sagittal planes in CT scans, or the space between the tumor and the bowel wall is <3 mm. Normal tissue constraints are according to the American Association of Physicists in Medicine guidelines in TG-101.

7. Follow-up

Measureable disease was assessed and documented before treatment. CT or MRI and the tumor biomarker (CA19-9) were performed at baseline (2 weeks of beginning therapy) and every 2 months thereafter for evaluations of response until disease progression or treatment discontinuation, which was based on RECIST version 1.1 by blinded independent imaging review. Laboratory evaluations included hematology, blood chemistry, and liver and kidney function were performed on days 1 (before drug administration), 8 and 15 of each treatment cycle. Treatment-related toxicity toxicity was evaluated throughout the whole treatment period and at each one-month follow-up. All adverse effects were assessed according to the National Cancer Institute Common

Toxicity Criteria for Adverse Events (CTCAE) version 4.0. An independent data monitoring committee of our center reviewed the safety and efficacy results regularly and provided recommendations to the investigators.

8. Outcomes

The primary endpoint was overall survival (OS), calculated as the time from the start of treatment to death. Secondary endpoints included 1-year OS, defined as the proportion of patients alive at 1 year; and progression free survival (PFS), calculated as the time from the start of treatment until documentation of any clinical or radiological disease progression or death, whichever occurred first; 1-year PFS, defined as the proportion of patients without disease progressions at 1 year; treatment-related toxicity and quality of life assessed by The European Organization for Research and Treatment of Cancer (EORTC): Quality of Life Questionnaire-Core 30 (QLQ-C30), which was performed at baseline, one, three, six months and twelve months after initiations of treatment. Higher scores in function domains and global health status indicate better quality of life, while higher scores in symptom domains imply worse quality of life.

9. Statistical analysis

We calculated that 170 patients were needed to be enrolled (85 per treatment group) to give 90% power to detect a statistically significant difference between treatment groups with a type I error of 5% (one-sided), assuming a hazard ratio (HR) of 0.77.

All efficacy endpoints were assessed in the intention-to-treat population. The

population included all patients enrolled in the trials regardless of receiving treatment. Safety was analyzed with data from the patient population who received at least one dose of study treatment. Demographic, disease and treatment characteristics were summarized with frequency and percentage for categorical variables, and median and interquartile range (IQR) for continuous variables. Categorical binary variables were compared with Fischer's exact test or χ^2 test (depending on number of observations). Student t-test or Mann-Whitney U test was used for analysis in the case of normally or non-normally distributed continuous covariates. OS and PFS associated 95% CIs were estimated by Kaplan-Meier methods. The log-rank test was used to compare OS and PFS between different treatment groups. Factors with a P-value <0.05 in the univariate Cox regression analysis were entered as candidate variables into the multivariate Cox proportional hazard regression analysis for identification of predictors correlating with OS and PFS. Quality of life was assessed by QLQ-C30. Scores were linearly transformed to a 0 to 100 scale. A clinically relevant change was defined as change in HRQOL scores of ≥10 points [23, 24]. Patients' scores at different time points were compared with paired samples t-test. All P-values were two-sided. P<0.05 were considered as statistically significant. Statistical analyses were performed with IBM SPSS version 22.0 (SPSS Inc., Armonk, NY) and SAS version 9.4 (SAS Institute, Inc., Cary, NC).