Summary attachment for the clinical protocol:

T-cell Therapy in Combination with Nivolumab, Relatlimab and Ipilimumab for Patients with Advanced Ovarian-, Fallopian Tube- and Primary Peritoneal Cancer

Primary completion date: 02.07.2024 (premature closure of the trial)

EudraCT no: 2020-002738-34

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Sponsor protocol code: GY2028

Clinicaltrials.gov NCT no.: NCT04611126

The trial was a clinical phaseI/II trial. It was closed the 2nd of July 2024 and data will be reported at www.clinicaltrials.gov. In addition, study results are expected to be published within the present year.

Short trial description:

In this trial treatment with tumor infiltrating lymphocyte (TIL) therapy was combined with treatment with two immune checkpoint inhibitors (PD1/LAG3 blockade) in platin-resistant ovarian cancer (OC) patients.

Study design

Included patients had platinum-resistant OC, an Eastern Cooperative Oncology Group (ECOG) performance status of 0-1, acceptable organ function and a metastasis of at least 1 cm^2 available for surgical resection. After inclusion patients underwent tumor resection and the TILs were grown *ex vivo*. Three to four weeks post tumor resection the patient was admitted for one week of lymphodepleting chemotherapy with cyclophosphamide (60 mg/kg for 2 days) followed by fludarabine phosphate (25 mg/m^2 for 5 days). Two days before TIL administration the first dose of nivolumab (BMS) 240 mg and relatlimab (BMS) 80 mg were administered. Nivolumab/relatlimab infusion was repeated every 14th day for a maximum of 4 cycles. The first evaluation of response was performed 6 weeks post TIL therapy.
Initially, inclusion of 18 patients in three steps (6+6+6) was planned with the addition of a CTLA-4 inhibitor (ipilimumab) in step two. However, due to slow recruitment the trial was closed prematurely.

Endpoints

The primary endpoints in the study were to access the safety and feasibility of the treatment. Secondary objectives included clinical efficacy according to RECIST 1.1 and characterization of infusion products and antitumor immune responses (immune monitoring).

Subjects

13 patients were screened and 6 subjects, with late-stage platinum resistant OC, were included in the trial. One patient was excluded before treatment due to TIL expansion failure. Thus, 5 patients received treatment with TILs and checkpoint inhibitors. Patient recruitment was slow mainly due to low referral of patients fulfilling inclusion criteria and the trial was closed prematurely for this reason.

Results

In total 5 patients were treated with ex vivo expanded tumor infiltrating lymphocytes in the period from June 2021 to December 2023. The patients ovarian cancer differed histologically, including one patient with undifferentiated carcinoma, two patients with low grade adenocarcinoma and two patients with high grade adenocarcinoma. The disease burden at baseline was high in all included patients. The median age was 62 years (range 45-70), and the median number of prior treatment lines was 3 (range 3-4).

Safety and feasibility

Tumor harvest and *ex vivo* expansion of TILs was successful in 5/6 included patients. Further, treatment with ICI was feasible with a median number of PD1/LAG3 infusions of three (range 2-4). Reasons for discontinuation of check-point inhibitors were hyperthyroidism and clinical deterioration/progression of the malignant disease.
Combined treatment with TIL and ICI was feasible, but the overall feasibility of the trial was challenged by the slow recruitment.

The treatment was well tolerated. All treatment related adverse events are shown in *Table 1.* Adverse events related to the chemotherapy were expected and included, in all patients, neutropenia, lymphopenia, thrombocytopenia, anemia, nausea and hyponatremia. Also, most patients (80%) experienced chemotherapy related worsening of PS and fatigue. The frequency of these AEs is comparable with other TIL trials.
The most frequently reported adverse events related to the TIL infusion were fever (80%) and fatigue (60%) which is also expected and comparable with previous trials.
Also, the treatment with checkpoint inhibitors was well tolerated. Adverse event included hyperthyroidism (40%), maculopapular rash (40%), dry mouth (20%), dyspnea (20%), fever (20%), pain (20%) and troponin increase (20%).
Only one grade ≥ 3 adverse event was registered in relation to TIL therapy and/or checkpoint inhibitors. This AE (grade 3 maculopapular rash) resolved spontaneously without the need for medical interventions.
Serious adverse events are shown in *Table 2*. Patients were in a fragile state and the post treatment complication rate was higher than expected. Severe complications included one case of life-threatening cardiac effusion due to disease progression and one case of severe CMV reactivation following chemotherapy. Finally, two patients developed fatal bowel obstruction which was attributed to the underlying disease.
Three serious adverse events could be categorized as serious adverse reactions (SAR) (Table 2). No SUSARs were reported.

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Treatment-related adverse event		
	Any grade, n(%)	Grade ≥ 3, n(%)
Chemotherapy		
Anemia	5 (100%)	1 (20%)
Hyponatriemia	5 (100%)	1 (20%)
Lymphopenia	5 (100%)	5 (100%)
Nausea	5 (100%)	0
Neutropenia	5 (100%)	5 (100%)
Trombocytopenia	5 (100%)	4 (80%)
Decrease in PS	4 (80%)	1 (20%)
Fatigue	4 (80%)	1 (20%)
Infections	3 (60%)	2 (40%)
Vomiting	3 (60%)	0
Diarrhea	2 (40%)	0
Obstipation	2 (40%)	0
Dry mouth	1 (20%)	0
Dyspnoe	1 (20%)	0
Elevated ALAT/ASAT	1 (20%)	0
Increased p-ferritin	1 (20%)	1 (20%)
Oral candida	1 (20%)	0
TIL product		
Fever	4 (80%)	0
Fatigue	3 (60%)	0
Decrease in PS	2 (40%)	0
Elevated ALAT/ASAT	1 (20%)	0
Maculopapular rash	1 (20%)	1 (20%)
Checkpoint inhibitors		
Hyperthyroidism	2 (40%)	0
Maculopapular rash	2 (40%)	1 (20%)
Dry mouth	1 (20%)	0
Dyspnoe	1 (20%)	0
Fever	1 (20%)	0
Pain	1 (20%)	0
Troponine increase	1 (20%)	0

Table 1: All treatment-related adverse events registered during the trial.

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Serious adverse events

PATIENT ID	COMMENT/DESCRIPTION	DATE OF ONSET	SUSPECTED DRUG	TYPE OF SAE
01	Infection	2021-07-23	None	SAE
01	Ileus	2021-08-05	None	SAE
03	Atrial fibrillation	2022-10-14	None	SAE
05	Pulmonary Embolism	2023-10-05	None	SAE
05	Vomiting and fever	2023-10-16	None	SAE
05	Nausea, vomiting, fever	2023-10-23	None	SAE
05	Colon Ileus	2023-10-23	None	SAE
05	Acute Kidney Failure	2023-10-27	None	SAE
05	Increased Troponin I	2023-09-28	None	SAE
06	Pericardial effusion	2024-01-19	None	SAE
02	Hyperthyroidsm	2022-01-04	Checkpoint inhibitors	SAR
05	Bacterial infection during neutropenia	2023-08-25	Chemotherapy	SAR
06	CMV reactivation	2023-12-15	Chemotherapy	SAR

Table 2: All registered serious adverse events (SAEs) and their relation to treatment. SAR= Serious adverse reaction.

Clinical efficacy: Four out of five patients had a reduction in tumor mass following treatment. The median progression free survival (mPFS) was 92 days. Two patients (02 and 03) had stable disease for 5 and 9 months, respectively.
Further, two patients (01 and 05) developed a partial response with > 30% reduction in tumor mass, but unfortunately, both responding patients died from bowel obstruction before confirmation of the response (five- and ten-weeks post treatment, respectively).

Immune monitoring: The composition of infusion products was analyzed using flowcytometry. The infused cells were almost exclusively T-cells (98.5%), predominantly effector memory cells which is comparable with previous trials.
In the two patients with low grade adenocarcinoma, the infusion product consisted almost exclusively of CD4+ T cells. In the remaining patients the fraction of CD8+ T cells were relatively high. The median expression of PD1 and LAG3 on CD8+ T cells were 14% and 34%, respectively, which was lower than reported in our previous trials.
Tumor reactivity of the REP-TILs was tested *in vitro* using autologous tumor cells (tumor cell culture or tumor digest). Tumor reactivity of the CD8+ populations was found in patient 01 and 06.

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Discussion
This clinical trial finds TIL therapy in combination with PD1/LAG3 inhibitors to be safe and feasible in patients with platinum-resistant OC. The feasibility was, however, challenged by the slow recruitment and the fragile patient population. Indications of clinical efficacy was seen in two patients, but we were not able to confirm the responses due to the early death of both patients. Reactivity of the infused cells were found in 2/5 (40%) patients.
Low grade adenocarcinoma is a subtype of ovarian cancer known to have very limited T cell infiltration and poor response to immunotherapy. This might explain the composition of the infusion product (mainly CD4+ T cells) as well as the lack of clinical response in the two patients (02 and 03) with low grade adenocarcinoma.
The expression of LAG3 and PD1 was lower than reported in our previous trials. However, the median expression of LAG3 of 34% and PD1 of 14% on CD8+ T cells was high enough to provide a rationale for the addition of and PD-1 inhibitor (nivolumab) and a LAG3 inhibitor (relatlimab).

Perspectives
The study results suggest a clinical benefit from the combination of TIL therapy and checkpoint inhibitors (PD1/LAG3) in platinum-resistant OC. It is, however, evident that clinical deterioration in the waiting time for TIL therapy is a risk for patients with advanced disease. Also, complications related to the malignant disease constitute a major challenge in late-stage OC. This provides a rationale for moving TIL therapy to earlier treatment lines in OC patients. Finally, histological heterogeneity should be considered in future TIL trials with OC.

Kind regards

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