PLEASE NOTE: *This trial has been registered retrospectively.*

Trial Description

Title

Isolation of primary cells from differentiated thyroid carcinomas to build 2D/3D cell cultures and human tissue

Trial Acronym

Refire-1

URL of the trial

[---]*

Brief Summary in Lay Language

Scientific studies in general are needed to improve our understanding of the pathogenesis and diagnosis as well as to develop new treatment approaches based on it.

In Germany, around 6,500 people suffer from thyroid cancer each year (as of 2012). There are different types of thyroid cancer, which emanates from different cells of the thyroid gland. Papillary carcinoma is the most common malignant thyroid tumor followed by follicular carcinoma. Both are also referred to as differentiated thyroid tumors. The cure prospects are good for differentiated thyroid tumors if recognized early. Over 90% of localized differentiated thyroid carcinomas are operated and treated with a radioiodine therapy. Even with distant metastatic differentiated thyroid carcinomas, a complete remission and a 10-year survival probability of 90% can be achieved by radioiodine therapy with good iodine uptake. Without or with insufficient radioiodine uptake, this probability is only 10%. The therapeutic options for radioiodine-negative thyroid carcinoma have been improved over the last years, but there is still a need for individualized and gentle treatment options.

Malignant tumors are very different in different patients and may have multiple gene mutations (sudden changes in genetic information in the nucleus), which sometimes complicates a standardized tumor treatment. In order to better examine gene mutations and to better predict the effectiveness of different drugs in the individual case, tumor models have been developed in the past from patient-derived cells. In this way, numerous patients with lung cancer or colon cancer are analyzed in more detail, resulting in an improvement in treatment options. In comparison, research on thyroid tumors is still insufficient.

The primary goal of the study described here is to construct two- and three-dimensional tumor models from human cells. For this purpose, tissue samples from patients with differentiated thyroid carcinomas or with clinically high-grade suspicion of a differentiated thyroid carcinoma during a planned operation is taken. Part of the tissue sample is cultured, grown and examined in a special nutrient solution. Based on the tumor models, the mode of action of various drugs

or their influence on the cell division and vitality and regulation of the sodium-iodine-symporter expression and function of differentiated thyroid carcinoma cells will be investigated. The results obtained will be correlated with the clinical course as well as with the molecular properties of the tumors and the histological findings.

Brief Summary in Scientific Language

Thyroid cancer affects about 6,500 people a year in Germany (as of 2012) [Robert Koch-Institut 2015]. Differentiated thyroid carcinomas represent the most common type of thyroid carcinoma (95%) [Willhauck et al. 2011]. They are (simplified) divided into papillary (PTC), follicular (FTC) and poorly differentiated thyroid carcinomas (PDTC). Histologically, they are distinguished from the rarer undifferentiated (anaplastic) carcinomas as well as the medullary carcinomas [Ceresini et al. 2012].
Over 90% of differentiated thyroid carcinomas are (radio-) iodine-storing carcinomas and can be treated successfully by thyroidectomy, often combined with radioiodine (RI) therapy [Paschke et al. 2015]. In RI therapy, patients receive radioactive iodine-131, which post-operatively removes remaining thyroid tissue and can be used to treat residual tumor manifestations and detect them with high sensitivity and specificity. Even with distant metastatic differentiated thyroid carcinomas, a good iodine uptake can achieve a full remission and a 10-year survival rate of 90%. In the case of loss of RI storage capacity or insufficient RI storage of the tumor manifestations, the treatment effects that can be achieved with RI are (clearly) limited and the 10-year survival probability decreases (in the case of clearly RI-negative tumors) to 10% [Paschke et al. 2015]. This is observed in the course of the disease in about two thirds of patients with distant metastases [Durante et al. 2006].

The growing understanding of the complex pathogenesis and the underlying intracellular signaling pathways in malignant and normal cells allows for a progressive breakdown of tumor-causing events and the identification of molecular targets. One of the central signaling pathways is the RAS-RAF-MEK-ERK signaling pathway [Wang et al. 2007]. Activation of this pathway leads to cell growth and proliferation. Differentiated thyroid carcinomas often have genetic changes in the signal kinases (including point mutations in BRAF and RAS, RET / PTC translocation), which lead to a constitutive activation of the downstream signaling cascade and thus to tumorigenesis [Fallahi et al. 2015, Nikiforov 2008, Ricarte-Filho et al. 2009]. In addition, an influence of these mutations on the loss of RI uptake capacity has been described [Liu et al. 2007, Yang et al. 2014]. Interestingly, the mutations rarely occur simultaneously, which further complicates standardized tumor treatment [Fassnacht et al. 2009].
Tyrosine kinase inhibitors such as the multikinase inhibitors sorafenib and levantinib are directly involved in this signaling pathway and inhibit several kinases. By using these substances, a significant extension of progression-free but not overall survival was achieved. At the same time, the drugs often cause distressing and, more rarely, serious side effects, which often severely impair patients' quality of life [Brose et al. 2014, Paschke et al. 2015, Schlumberger et al 2015]. A balance between the benefit-risk ratio must therefore always be carried out individually for each patient and alternative therapeutic measures (e.g. surgical or radiotherapeutic) must be ruled out in advance [Kreissl et al. 2015].
Another promising but hitherto unsuccessful therapeutic approach is the reinduction of RI storage by increasing the sodium iodide-symporter (NIS) expression rate, also called redifferentiation (e.g. with dabrafenib and selumetinib). In clinical studies, these substances were able to restore iodid uptake in some of the patients and cause partial remission of the tumors [Ho et al. 2013, Rothenberg et al. 2015].
In general, the therapeutic options for RI-refractory thyroid carcinomas have

improved in recent years, but there is an urgent need for more individualized and gentle treatment options. The prognostic goal is the development of new drugs or the targeted use of existing drugs for a personalized tumor therapy, taking into account the individual tumor heterogeneities.
New substances, medicines and medical devices (e.g. implants) must be tested for their safety for humans before they are released onto the market. Animal experiments are usually carried out for this purpose. However, animal experiments are not only ethically controversial: due to the biodiversity of humans and animals, the results of animal experiments are often not transferable to humans [van der Worp et al. 2010]. Using alternative methods it is possible to avoid unnecessary animal testing. One such alternative method is the production of cell cultures from human cells and the artificial production of human tissue (tissue engineering). These can be used as study models for biomedical examinations instead of animal experiments. As needed, the cell and tissue models are constructed as healthy or diseased cultures (e.g. cancerous tissue) [Amelian et al. 2017, Carvalho et al. 2017]. To build these cultures, human cells from tissue samples are needed.
In this research project, primary cells will be taken from differentiated thyroid carcinomas and metastases and used to construct 2D / 3D cell and tissue cultures. For this purpose, tissue samples from patients with differentiated thyroid carcinomas or with clinically high-grade suspicion of a differentiated thyroid carcinoma will be taken. Part of the tissue sample is cultured, grown and examined in a special nutrient solution. Based on the tumor models, the mode of action of various drugs or their influence on the cell division and vitality and regulation of the sodium-iodine-symporter expression and function of differentiated thyroid carcinoma cells will be investigated. The results obtained will be correlated with the clinical course as well as the molecular properties of the tumors and the histological findings. The benefit of the study is the gaining of new scientific insights into the fundamentals of the molecular biology of the development and therapy of thyroid carcinomas and the resulting possibilities of individualized therapy improvement for future patients.

Organizational Data

- DRKS-ID: **DRKS00014070**
- Date of Registration in DRKS: **2018/09/11**
- Date of Registration in Partner Registry or other Primary Registry: **[---]***
- Investigator Sponsored/Initiated Trial (IST/IIT): **yes**
- Ethics Approval/Approval of the Ethics Committee: **Approved**
- (leading) Ethics Committee Nr.: **41/18 , Ethikkommission der Medizinischen Fakultät der Otto-von-Guericke-Universität Magdeburg**

Secondary IDs

- Universal Trial Number (UTN): **U1111-1211-8628**

Health condition or Problem studied

- ICD10: **C73 - Malignant neoplasm of thyroid gland**

Interventions/Observational Groups

- Arm 1: **A one-armed study design with 2 sub-studies is planned.**

  Sub Study 1:
  Establishment of patient-derived 2D / 3D cell culture and tissue culture.
  N = 10 patients (including at least 3 PTC and 3 FTC) at technical difficulty extension to up to 20 patients. In case of no cell growth in less than 30% (additive) the study will be terminated.
  If a patient is included in the study with suspicion of thyroid cancer, but the suspicion is not confirmed in the histology, he counts as a drop-out and the next patient moves behind.
  Subsequent to the successful completion of sub-study 1, sub-study 2 follows.

  Sub Study 2:
  Examination of various drugs. Sub-Study 2 will include an additional 60 patients.

  Only tissue samples are taken from patients who have a diagnosis of a differentiated thyroid carcinoma or a clinically high-grade suspicion is present on the same and an elective surgery is planned. The tissue samples are separated on the one hand for clinical evaluation and on the other hand for scientific preparation by the pathologist and processed immediately afterwards.

  If both sub-studies are performed, the number of study participants is 80 subjects.

Characteristics

- Study Type: **Non-interventional**
- Study Type Non-Interventional: **Other**
- Allocation: **Single arm study**
- Blinding: **[---]***
- Who is blinded: **[---]***
- Control: **Uncontrolled/Single arm**
- Purpose: **Basic research/physiological study**
- Assignment: **Single (group)**
- Phase: **N/A**
- Off-label use (Zulassungsüberschreitende Anwendung eines Arzneimittels): **N/A**

Primary Outcome

The primary goal of the research project is to isolate primary cells from surgically harvested carcinogenic thyroid tissue or metastases to construct patient-derived 2D / 3D tumor models in vitro. The main focus, but not exclusively, lies on

advanced, known RI-negative or insufficiently RI-storing tumors. The resection material is obtained during clinically indicated elective surgery (e.g. thyroid or tumor resection or histological verification of metastasis).

Secondary Outcome

The secondary objective of the research project is the in-vitro investigation of various drugs and their influence on cell division & -vitality and regulation of NIS expression and function (using RI-uptake experiments) of differentiated thyroid carcinomas using the patient-derived 2D / 3D tumor models. The results are correlated with the clinical course as well as with the molecular pathological properties of the tumors (e.g. BRAF mutation) and histological findings (classification according to WHO).

planned pharmaceuticals:

- Doxorubicin,
- Sorafenib,
- Lenvantinib,
- Dabrafenib,
- Trametinib,
- Pazopanib,
- Vandetanib,
- Roaccutan,
- Rosi-/ Pioglitazon,
- Cabozantinib,
- Selumetinib

Countries of recruitment

- DE **Germany**

Locations of Recruitment

- University Medical Center **Universitätsklinikum Magdeburg A.ö.R., Magdeburg**

Recruitment

- Planned/Actual: **Actual**
- (Anticipated or Actual) Date of First Enrollment: **2018/05/24**
- Target Sample Size: **80**
- Monocenter/Multicenter trial: **Monocenter trial**
- National/International: **National**

Inclusion Criteria

- Gender: **Both, male and female**

Gender: **Both, male and female**

- Minimum Age: **18 Years**
- Maximum Age: **79 Years**

Additional Inclusion Criteria

• men and women (no specific gender distribution planned)
• age: ≥ 18 / <80 years
• capacity to consent
• elective clinically indicated surgery
• diagnosis of differentiated thyroid carcinoma or clinically high-grade suspected differentiated thyroid carcinoma
• signed consent form

Exclusion criteria

• known HIV, HBV or HCV infection

Addresses

- **Primary Sponsor**

Universitätsklinikum Magdeburg A.ö.R. Universitätsklinik für Radiologie und Nuklearmedizin Bereich Nuklearmedizin
Mr. Prof. Dr. med Michael C. Kreißl
Leipziger Str. 44
39120 Magdeburg
Germany

Telephone: **+49 391 6713003**
Fax: **+49 391 6714078**
E-mail: **michael.kreissl at med.ovgu.de**
URL: **http://www.krn.ovgu.de/**

- **Contact for Scientific Queries**

Universitätsklinikum Magdeburg A.ö.R. Universitätsklinik für Radiologie und Nuklearmedizin Bereich Nuklearmedizin
Ms. Dr. rer. nat. Franziska Stöber
Leipziger Str. 44
39120 Magdeburg
Germany

Telephone: **+49 391 6721920**
Fax: **+49 391 6714078**
E-mail: **franziska.stoeber at med.ovgu.de**
URL: **http://www.krn.ovgu.de/**

- **Contact for Public Queries**

Universitätsklinikum Magdeburg A.ö.R. Universitätsklinik für Radiologie und Nuklearmedizin Bereich Nuklearmedizin
Ms. Dr. rer. nat. Franziska Stöber
Leipziger Str. 44
39120 Magdeburg
Germany

Telephone: **+49 391 6721920**
Fax: **+49 391 6714078**
E-mail: **franziska.stoeber at med.ovgu.de**
URL: **http://www.krn.ovgu.de/**

- **Collaborator, Other Address**

CPO - cellular phenomics & oncology Berlin-Buch GmbH
Mr. Dr. rer. nat. Christian Regenbrecht
Robert-Rössle-Straße 10
13125 Berlin
Germany

Telephone: **[---]***
Fax: **[---]***
E-mail: **christian.regenbrecht at cellphenomics.com**
URL: **https://www.cellphenomics.com/**

Sources of Monetary or Material Support

- **Institutional budget, no external funding (budget of sponsor/PI)**

Universitätsklinikum Magdeburg A.ö.R. Universitätsklinik für Radiologie und Nuklearmedizin Bereich Nuklearmedizin
Mr. Prof. Dr. med Michael C. Kreißl
Leipziger Str. 44
39120 Magdeburg
Germany

Telephone: **+49 391 6713003**
Fax: **+49 391 6714078**
E-mail: **michael.kreissl at med.ovgu.de**
URL: **http://www.krn.ovgu.de/**

Status

- Recruitment Status: **Recruiting ongoing**
- Study Closing (LPLV): **[---]***

Trial Publications, Results and other documents

- trial protocol (mandatory for transfer to Studybox) **Studienprotokoll**

** This entry means the parameter is not applicable or has not been set.*

**** This entry means that data is not displayed due to insufficient data privacy clearing.*