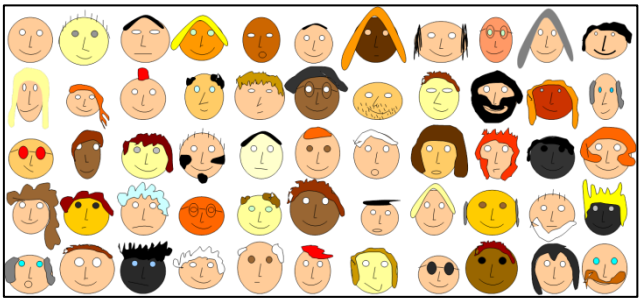
Registration & Shipment Manual

International Patients

INFORM

INdividualized Therapy FOr Relapsed Malignancies in Childhood



Coordinating Investigator Registry: Prof. Dr. med. Olaf Witt

NCT Clinical Trial Center Heidelberg

Dr. Kristian Pajtler / Ruth Witt / Martina Nesper-Brock

Tel: 06221-56-7255 / -7294 / -7439

Email: INFORM_info@DKFZ.de

Website: http://www.dkfz.de/de/inform

Registry Code: NCT-2013-0220

German Clinical Trial Register ID: DRKS00007623

Version and date: Version 1, dated 2.12.2016

Important Notice

Please ensure that all cases are discussed with the respective national coordinator for eligibility for INFORM.

After receiving informed consent it is mandatory to register every patient in the central MARVIN database before sample shipment.

INFORM shipment forms provided with an INFORM patient ID can be generated upon registration of a patient in MARVIN. Only after registration and with these forms samples can be shipped to the INFORM sample processing laboratory at Neuropathology in Heidelberg. Samples shipped without INFORM patient ID and no previous registration in MARVIN cannot be processed.

For international patients, two types of material are required:

I. **Tumor DNA and Tumor RNA**
   from fresh frozen malignant material of the current disease episode
   (tumor cell content >40%)
II. **DNA from non-malignant material (germline, e.g. from blood)**

Please do not send fresh frozen tumor tissue, bone marrow, blasts, blood or saliva to Heidelberg, only extracted nucleic acids will be accepted. Read this manual carefully while incomplete submissions will, without exception, significantly delay the process!

For information regarding sample shipment, please read chapter 2 and contact your national coordinator first. In case of further questions contact the INFORM sample processing laboratory at Neuropathology in Heidelberg:

Petra Fiesel / David Capper
06221-56-4650
INFORM_samples@DKFZ.de

For all other additional information regarding the INFORM registry, please contact your national coordinator.

No verification procedure will be performed for the identified targets. Verification needs to be done by the submitting hospital! The analyses are not clinically validated!

Flow Chart

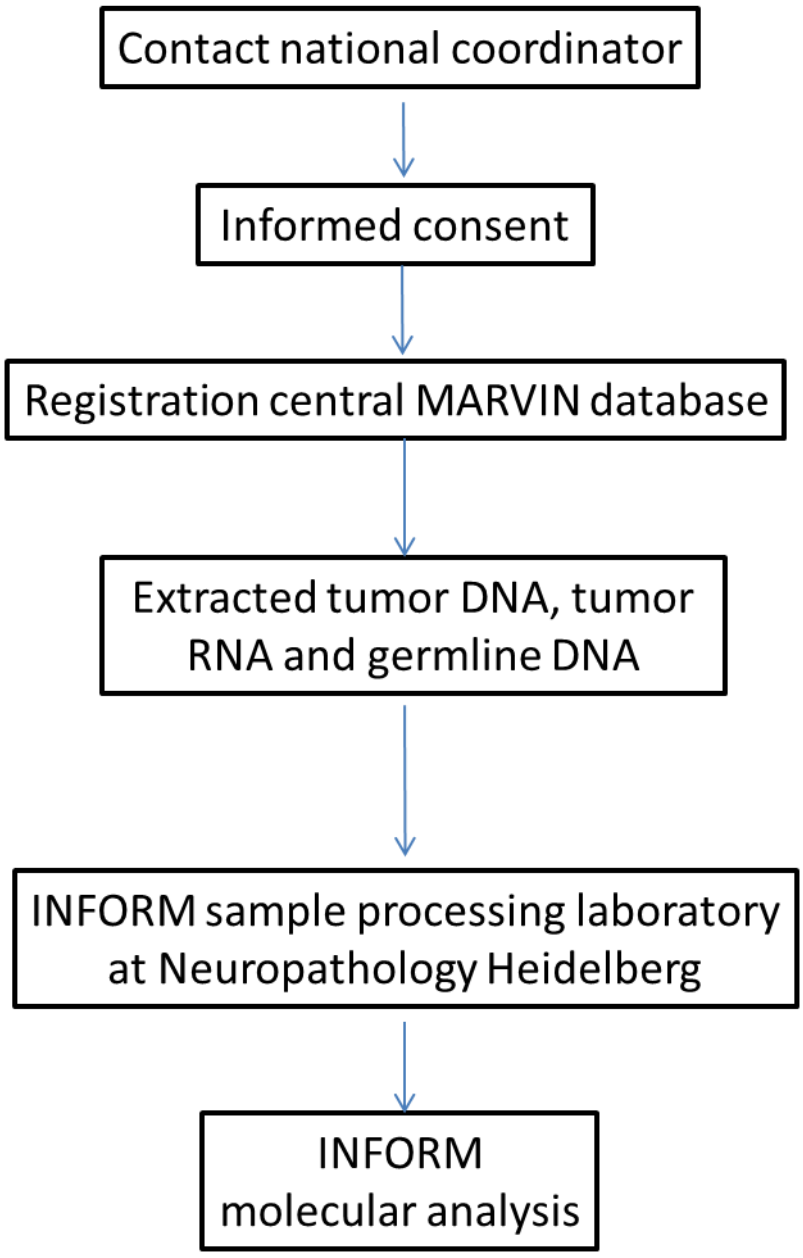

Contact national coordinator
Informed consent
Registration central MARVIN database
Extracted tumor DNA, tumor RNA and germline DNA
INFORM sample processing laboratory at Neuropathology Heidelberg
INFORM molecular analysis

Table of Contents

1. Registration

1.1 Central MARVIN database

In the INFORM registry, patient data is captured and stored via remote data entry system in the central MARVIN database of the Society for Pediatric Oncology and Hematology (GPOH). Before registration, all cases need be discussed for eligibility with the respective national coordinator as there may be open Phase I/II trials available for certain patient subpopulations.

After receiving informed consent it is mandatory to register every patient in the central MARVIN database before sample shipment. INFORM shipment forms, provided with an INFORM patient ID can be generated upon registration of a patient in MARVIN. Only after registration and with these forms, samples can be shipped to the INFORM sample processing laboratory at Neuropathology in Heidelberg.

1.2 Access to the central MARVIN database

Every pediatric oncology center in the partner countries (stated by the respective national coordinator) can request for an INFORM registry MARVIN access via the NCT Clinical Trial Center in Heidelberg. Name and email address of the people, who should be entitled to have access to the INFORM Registry in MARVIN and the name and the postal address of the requesting center have to be sent to the INFORM study center by email (**INFORM_info@DKFZ.de**). MARVIN training is mandatory, before access can be provided as it is routine practice within all GPOH trials that use the MARVIN platform. If the person has already been trained, access will be granted within short time.

For questions regarding MARVIN and access please contact the NCT Clinical Trial Center Data Management group:

Angelika Freitag / Christine Grasy

Tel. 06221-56-6237 / 7457

Email: INFORM_info@DKFZ.de

1.3 INFORM patient ID

The patients' name and all confidential information are subject to medical confidentiality and to the regulations of German Law (Bundesdatenschutzgesetz, BDSG). Only the treating physician/hospital will be able to link the registry data to personal information like name and address. The registry does not keep record on patients' name or address.

An INFORM patient ID will be automatically generated after registration of a new patient in MARVIN. Samples can be shipped to Heidelberg only after registration of a patient and accompanied by a shipment form created in MARVIN. Samples without INFORM patient ID cannot be processed.

1.4 Patient registration step 1: In- and Exclusion criteria

CAVE: Every started registration is charged with 50 €! Exception: the patient is already recorded in MARVIN and has a unique MARVIN-ID, which can be used to register the patient in MARVIN for INFORM. Therefore it is strongly recommended to check whether a patient is eligible before starting the registration procedure in MARVIN. This includes discussion of the case with the respective national coordinator. Patient registration in the central MARVIN database takes place via a two-step process. In the first step, general and entity specific in- and exclusion criteria are screened. When eligible for INFORM, the patient will be registered. After this first registration step, an INFORM patient ID and INFORM shipment forms can be generated. From this moment on, samples can be shipped to the INFORM sample processing laboratory at Neuropathology in Heidelberg.
For in- and exclusion criteria, please see our website (http://www.dkfz.de/en/inform/protocol-synopsis.html).

1.5 Patient registration step 2: Baseline assessment

To speed up the process, it is not necessary to enter the full baseline characteristics into the database to obtain an INFORM patient ID, which enables sample shipment within a relatively short time span. It is however mandatory, to finish the second registration step (baseline characteristics) within 14 days after sample submission in order to allow the INFORM registry to report potential targets. In case of an incomplete documentation of baseline assessment, no targets can be reported.

1.6 Additional information

For further information concerning the INFORM registry, please see the registry protocol, contact the respective national coordinator.

Website: http://www.dkfz.de/en/inform/protocol-synopsis.html

2. Shipment of international patient material

2.1 General information

It is mandatory to register every patient in the central MARVIN database before sample shipment. For detailed registration information, please see chapter 1. Only samples accompanied by shipment forms, and provided with the INFORM patient ID, will be processed (see chapter 2.3).

Mandatory material requirements:

I) Tumor DNA and Tumor RNA
from fresh frozen malignant material of the current disease episode
(tumor cell content >40%)

II) DNA from non-malignant material (germline, e.g. from blood)

Please send samples as soon as possible. The time between biopsy/puncture of the current disease and receipt of all required samples and information in the INFORM sample processing laboratory at Neuropathology in Heidelberg should not exceed 8 weeks; otherwise the analyses will not be performed.

In case of questions please always contact your national coordinator first. For further questions regarding sample shipment:
INFORM sample processing laboratory at Neuropathology in Heidelberg
Petra Fiesel / David Capper
Tel: 06221-56-4650
Fax: 06221-56-4566
Email: INFORM_samples@DKFZ.de

Whenever possible, please also provide extracted **Tumor DNA and Tumor RNA** from fresh frozen malignant material **of the primary tumor or a previous relapse** (please send accompanied by shipment forms, and provided with the INFORM patient ID). If only FFPE material is available of the primary or a previous relapse, please send extracted Tumor DNA or provide 20 unstained sections of 10µm instead (not mandatory, **for research purposes** only). Please do not send FFPE blocks, as we cannot guarantee the return of material.

No verification procedure will be performed for the identified targets. Verification needs to be done by the submitting hospital! The analyses are not clinically validated!

2.2 Mandatory material requirements for international patients

Please do not send fresh frozen tumor tissue, bone marrow, blasts, blood or saliva to Heidelberg, only extracted nucleic acids will be accepted. The INFORM sequencing analyses cannot be performed with nucleic acids from Formalin fixed paraffin embedded (FFPE) material.

It is mandatory to send **Tumor DNA and Tumor RNA from suitable fresh frozen malignant material** of the current refractory/relapsed/progressive disease episode, with a tumor cell content of at least 40%.

Additionally, **DNA from non-malignant material (germline)** is required (see also Table 1). Please make sure to send germline DNA which is completely free of tumor content (non-malignant material). In case of leukemia the germline DNA must derive from complete remission bone marrow.

The analyses only start when tumor and germline samples as well as all required information have arrived in the INFORM sample processing laboratory at Neuropathology in Heidelberg. It is mandatory to send within 8 weeks after biopsy/puncture.

Mandatory material requirements	Nucleic acids	Isolated from
Malignant material	**Tumor DNA** **>2500ng** minimum concentration of 25ng/µl (optimally in 30 - 100µl) & **Tumor RNA** **>2500ng** minimum concentration of 45ng/µl (optimally in 30 - 55µl) from **fresh frozen** malignant material of the current disease episode **tumor cell content >40%**	extracted from **suitable* fresh frozen tumor tissue** (tumor cell content >40%) or extracted from enriched or sorted leukemic blasts or neuroblasts (blast content >40%) or extracted from bone marrow aspirate containing >40% neuroblast infiltration (% after cytospin, not in bone marrow smear)
Non-malignant material (germline)**	**Non-malignant DNA** **>2000ng** minimum concentration of 25ng/µl (optimally in 30 - 80µl)	extracted from **blood** (store blood cooled and isolate DNA within 4 days) or extracted from **complete remission bone marrow** (in case of leukemia) or extracted from saliva (only in exceptional cases, as low yield and bad quality is expected)

Table 1: Mandatory material requirements for international patients. If possible please send a higher amount and a higher concentration. The concentration should be measured with Qubit (not e.g. nano drop). Please ship on dry ice per overnight express.

*** Please make sure that the fresh frozen tumor tissue used for extraction of nucleic acids is suitable:**

- do not use FFPE material
- use **snap frozen fresh tumor tissue** (frozen optimally within 30 minutes)
- extract from **vital tumor material** with as little necrosis as possible
- necrosis may result in degradation of nucleic acids
- **select areas with highest tumor cell content** (aim for >80% of total cells)
- tumor cell content has to be at least 40%.
- It is recommended to use nuclease free H_2O or low buffered TE for elution
- always keep RNA on ice and freeze as soon as possible
- avoid repeated freezing and thawing
- the concentration should be measured with Qubit (not e.g. nano drop)
- please ship on dry ice per overnight express.

** Furthermore, if the patient ever received allogeneic stem cell transplantation special germline controls are necessary.

In case of **post allogeneic stem cell transplantation patients**, please send:

I) patient germline:

DNA from blood of the patient before SCT

or DNA from saliva (only in exceptional cases, as bad quality is expected)

AND II) donor germline:

DNA from blood of the donor

or DNA from blood of the patient post SCT (with complete donor chimerism).

In case of **Leukemia post allogeneic SCT**, please send:

I) patient germline:

DNA from complete remission bone marrow of the patient before SCT

or DNA from saliva (only in exceptional cases, as bad quality is expected)

AND II) donor germline:

DNA from blood of the donor

or DNA from complete remission bone marrow of the patient post SCT

(with complete donor chimerism, MRD negative).

2.3 Shipment of Material

Only samples accompanied by shipment forms, and provided with the INFORM patient ID, will be processed (please do not send patients' names). Also provide a shipment form for the non-malignant material:

- ➔ **Fill in all required sample and contact information into the MARVIN form "Shipment of Material".**
- ➔ **After completing the form click on: "Transform: Print Shipment Sheet" and send it together with the material to the address stated on the sheet.**

Please ship samples **on dry ice**. Fill box completely with dry ice to ensure sufficient freezing of samples during shipment.
Ship **per overnight express** and consider that **we cannot receive samples on weekends and on German public holidays!**

For further information about shipments (company, declaration, labeling, customs clearance, payment, etc.) contact the respective national coordinator.

Please send the samples to the following address:

Institute of Pathology
Department Neuropathology
INFORM sample processing laboratory
Im Neuenheimer Feld 224
69120 Heidelberg
Germany

Contact: Petra Fiesel / David Capper
Tel: +49 6221 56 4650
Email: INFORM_samples@DKFZ.de

Once the samples are on the way to Heidelberg, please **send an email to INFORM_samples@DKFZ.de stating the shipment company and the tracking number / Way Bill.**