

**Wissenschaftliches Symposium „Best Abstracts“  
Montag, 10. Oktober, 11:45 - 13:15 Uhr**

**V868 - Einfluss der Resektionsränder nach neoadjuvanter  
Chemotherapie auf Lokalrezidivrate und das Überleben  
beim Mammakarzinom /  
Impact of Surgical Margins in Breast Cancer After  
Preoperative  
>Systemic Chemotherapy on Local  
Recurrence and Survival**

**Kerstin Wimmer (Wien, A)**

*Wimmer K.<sup>1</sup>, Bolliger M.<sup>1</sup>, Bago-Horvath Z.<sup>2</sup>, Steger G.<sup>3</sup>, Kauer-Dorner D.<sup>4</sup>,  
Helfgott R.<sup>5</sup>, Gruber C.<sup>6</sup>, Moinfar F.<sup>6</sup>, Mittelböck M.<sup>7</sup>, Fitzal F.<sup>1</sup>*

<sup>1</sup>Medizinische Universität Wien, Klinische Abteilung für Viszeralchirurgie, Wien, Österreich, <sup>2</sup>Medizinische Universität Wien, Klinische Abteilung für Pathologie, Wien, Österreich, <sup>3</sup>Medizinische Universität Wien, Klinische Abteilung für Onkologie, Wien, Österreich, <sup>4</sup>Medizinische Universität Wien, Klinische Abteilung für Strahlentherapie, Wien, Österreich, <sup>5</sup>Ordendklinikum Linz, Chirurgie, Linz, Österreich, <sup>6</sup>Ordendklinikum Linz, Pathologie, Linz, Österreich, <sup>7</sup>Medizinische Universität Wien, Center for Medical Statistics, Informatics and Intelligent Systems, Wien, Österreich

**Background:** While “no tumour on ink” is an accepted margin width for R0 resection in primary surgery, it's unclear if it's oncologically safe after neoadjuvant chemotherapy (NAC). Only limited data demonstrate that surgery within new margins in cases of a pathological complete response (pCR) is safe. We therefore investigated the influence of different margins and pCR on local recurrence and survival rates after NAC.

**Methods:** We retrospectively analysed data of 406 women with invasive breast cancer, treated with NAC and breastconserving therapy between 1994 and 2014 in two certified Austrian breast health centres. We compared R B 1 mm, R[1 mm and RX (pCR) for local recurrence-free survival (LRFS), disease-free survival (DFS) and overall survival (OS).

**Results:** After a median follow-up of 84.3 months, the 5-year LRFS (R B 1 mm: 94.2%, R[1 mm: 90.6%, RX: 95.0%;  $p = 0.940$ ), the 5-year DFS (R B 1 mm: 71.9%, R[1 mm: 74.1%, RX: 87.2%;  $p = 0.245$ ) and the 5-year OS (R B 1 mm: 85.1%, R[1 mm: 88.0%, RX: 96.4%;  $p = 0.236$ ) did not differ significantly between narrow, wide, nor RX resections. Regarding DFS and OS, a negative nodal status reduced the hazard ratio significantly.

**Conclusion:** There is no significant difference in LRFS, DFS and OS comparing close, wide or unknown margins after pCR. We suggest that resection in new margins after NAC is safe according to “no tumour on ink”. Resection of the clipped area in cases of pCR is emphasized.

**V872 - 10-Tage Decitabin vs. Standard-Induktion, mit nachfolgender allogener Stammzelltransplantation (HSZT) bei AML-Patienten  $\geq 60$  Jahre: eine randomisierte Phase III Intergroup-Studie der EORTC Leukemia Group, GIMEMA, CELG und GMDS-SG / 10-day decitabine vs. Conventional chemotherapy ("3+7") followed by allografting (hsct) in aml patients  $\geq 60$  years: a randomized phase iii intergroup study of the eortc leukemia group, gimema, celg, and gmds-sg**

**Michael Lübbert (Freiburg i. Br., D)**

*Lübbert M.<sup>1</sup>, Wijermans P.<sup>2</sup>, Kicinski M.<sup>3</sup>, Chantepie S.<sup>4</sup>, van der Velden W.<sup>5</sup>, Noppeney R.<sup>6</sup>, Griskevicius L.<sup>7</sup>, Neubauer A.<sup>8</sup>, Crysandt M.<sup>9</sup>, Vrhovac R.<sup>10</sup>, Luppi M.<sup>11</sup>, Fuhrmann S.<sup>12</sup>, Audisio E.<sup>13</sup>, Candoni A.<sup>14</sup>, Legrand O.<sup>15</sup>, Foá R.<sup>16</sup>, Gaidano G.<sup>17</sup>, van Lameren-Venema D.<sup>2</sup>, Posthuma E.F.<sup>18</sup>, Hoogendoorn M.<sup>19</sup>, Giraut A.<sup>3</sup>, Stevens-Kroef M.<sup>5</sup>, Jansen J.<sup>5</sup>, Ammatuna E.<sup>20</sup>, Vilque J.-P.<sup>4</sup>, Wäsch R.<sup>1</sup>, Becker H.<sup>21</sup>, Blijlevens N.<sup>5</sup>, Dührsen U.<sup>6</sup>, Baron F.<sup>22</sup>, Suciu S.<sup>3</sup>, Amadori S.<sup>23</sup>, Venditti A.<sup>23</sup>, Huls G.<sup>20</sup>*

<sup>1</sup>University Medical Center Freiburg, Department of Hematology, Oncology and Stem Cell Transplantation, Freiburg, Deutschland, <sup>2</sup>Haga Teaching Hospital, Department of Hematology, The Hague, Niederlande, <sup>3</sup>EORTC Headquarters, Brussels, Belgien, <sup>4</sup>Centre Hospitalo-Universitaire de Caen, Caen, Frankreich, <sup>5</sup>Radboud University Medical Centre, Nijmegen, Niederlande, <sup>6</sup>Klinik für Hämatologie, Universitätsklinikum Essen, Essen, Deutschland, <sup>7</sup>Oncology and Transfusion Medicine Center, Vilnius University Hospital Santaros Klinikos, Vilnius, Litauen, <sup>8</sup>Department of Internal Medicine, Hematology, Oncology and Immunology, Philipps University Marburg, Marburg, Deutschland, <sup>9</sup>Department of Hematology, Oncology, Hemostaseology and Stem Cell Transplantation, Medical Faculty, Center for Integrated Oncology Aachen Bonn Cologne Duesseldorf, Aachen, Deutschland, <sup>10</sup>Department of Haematology, University Hospital Centre Zagreb, Zagreb, Kroatien, <sup>11</sup>Dipartimento di Scienze Mediche e Chirurgiche Materno-Infantili e dell'Adulto, University of Modena and Reggio Emilia, Modena, Italien, <sup>12</sup>HELIOS Hospital Berlin-Buch, Berlin, Deutschland, <sup>13</sup>SC Ematologia Città della Salute e della Scienza Torino, Torino, Italien, <sup>14</sup>Clinica Ematologica Azienda Sanitaria Universitaria Integrata di Udine, Udine, Italien, <sup>15</sup>Service d'Hématologie Clinique et de Thérapie cellulaire, Hôpital Saint Antoine, APHP, Paris, Frankreich, <sup>16</sup>Ematologia, Dipartimento di Medicina Traslazionale e di Precisione, "Sapienza" Università di Roma, Rom, Italien, <sup>17</sup>Division of Hematology, Department of Translational Medicine, Università del Piemonte Orientale and Azienda Ospedaliero-Universitaria Maggiore della Carità, Novara, Italien, <sup>18</sup>Department of Internal Medicine, Reinier de Graaf Hospital, Delft, Niederlande, <sup>19</sup>Department of Hematology, Medical Center Leeuwarden, Leeuwarden, Niederlande, <sup>20</sup>University Medical Center Groningen, Groningen, Niederlande, <sup>21</sup>University Medical Center Freiburg, Department of Hematology, Oncology and Stem Cell Transplantation, Freiburg, Deutschland, <sup>22</sup>University of Liège, Liège, Belgien, <sup>23</sup>Department of Biomedicine and Prevention, University of Rome Tor Vergata, Rom, Italien

Older, fit AML patients (pts) receiving induction chemotherapy (IC) have poor long-term survival unless HSCT is performed. DNA-hypomethylating agents have become the backbone of AML therapy in pts unfit for IC, with promising results for the 10-day (d) decitabine (DEC) schedule as bridging prior to HSCT.

**Aims:** To compare efficacy and safety of 10-d DEC vs. IC, followed by allografting, in older fit AML pts (NCT02172872).

Key inclusion criteria: newly diagnosed AML, age  $\geq 60$  years (yr), eligible for IC, WHO PS 0-2. DEC was administered over 10 d in cycle 1 (20 mg/m<sup>2</sup>), 10 or 5 d subsequently (depending on bone marrow blast clearance). IC: daunorubicin 60 mg/m<sup>2</sup> x3, cytarabine 200 mg/m<sup>2</sup> x7, followed by 1-3 further chemotherapy cycles. Pts with an HLA-matched donor and at least stable disease were encouraged to undergo HSCT after  $\geq 1$  cycle. Pts from the DEC arm not receiving HSCT could continue DEC. Primary endpoint: overall survival (OS). The statistical design aimed to detect a hazard ratio (HR) for OS of 0.75 (one-sided alpha 0.025, 85% power).

Between 12/2014 and 8/2019, 606 pts were randomized 1:1. Median follow-up was 4.0 yr, median age 68 yr (range 60-81), 34% of pts were  $\geq 70$  yrs old, 32% had adverse risk by ELN 2017. A median of 3 DEC cycles (Q1-3: 2-5) and 2 IC cycles (Q1-3: 1-2) were administered. CR/CRi rate: 48% with DEC and 61% with IC. HSCT as part of the protocol was performed in 122 pts (40%, 30 pts not in CR/CRi) from the DEC and 118 (39%, 11 pts not in CR/CRi) from the IC arm, and in 52% in both arms at any time. OS (after 423 deaths) was not significantly different after DEC vs IC (HR=1.04, 95% CI: 0.86-1.26 2-sided p=0.68). Median OS was 15 months (95% CI: 13-18) in the DEC and 18 months (95% CI: 14-22) in the IC group. OS rates (%) after 1, 2, 3 and 4 yr for DEC vs IC were 58 vs 59, 38 vs 40, 30 vs 33, and 26 vs 30, respectively. In age subgroups, the estimated HR for OS for DEC vs IC was 1.34 (99% CI: 0.79-2.28) for pts 60-64, 1.14 (99% CI: 0.77-1.69) for pts 65-69, and 0.84 (99% CI: 0.55-1.26) for pts  $\geq 70$  yr (p-value for trend: 0.058). Notable differences in grade 3-5 adverse events (%) reported (before HSCT) with DEC vs. IC: febrile neutropenia (37 vs 57%), decreased platelets (24 vs 32%), oral mucositis (2 vs 10%), diarrhea (1 vs 8%), decreased neutrophils (19 vs 13%). The 30-day mortality rate was 3.6% for DEC and 6.4% for IC.

In **conclusion**, DEC resulted in similar OS and HSCT rates but a better safety profile compared to IC.

# V869 - Phase-I/II-Studie eines Peptid-basierten COVID-19-Impfstoffs zur Induktion von T-Zell-Antworten gegen SARS-CoV-2 bei Krebspatienten mit B-Zell-Defizienz / Phase I/II trial of a peptide-based COVID-19 vaccine to induce SARS-CoV-2-specific T-cell responses in cancer patients with B-cell deficiency

Juliane Walz (Tübingen, D)

Heitmann J.<sup>1,2</sup>, Tandler C.<sup>3,1,2,4</sup>, Marconato M.<sup>1</sup>, Habibzade T.<sup>5</sup>, Rittig S.M.<sup>6</sup>, Tegeler C.M.<sup>1,7</sup>, Nelde A.<sup>3,1,2,4</sup>, Maringer Y.<sup>3,1,2,4</sup>, Jäger S.<sup>8,1,9</sup>, Denk M.<sup>3,4</sup>, Richter M.<sup>3,4</sup>, Oezbek M.T.<sup>3,4</sup>, Wiesmüller K.-H.<sup>10</sup>, Bauer J.<sup>3,1,4,2</sup>, Riehl J.<sup>3,1,4</sup>, Wacker M.<sup>3,1,4,2</sup>, Schroeder S.<sup>3,4,11</sup>, Hörber S.<sup>12</sup>, Peter A.<sup>12</sup>, Meisner C.<sup>13,14</sup>, Fischer I.<sup>13</sup>, Löffler M.W.<sup>15,2,4,16</sup>, Peuker C.A.<sup>6</sup>, Habringer S.<sup>6</sup>, Goetze T.O.<sup>5</sup>, Jäger E.<sup>5</sup>, Rammensee H.-G.<sup>16,2,4</sup>, Salih H.R.<sup>1,2</sup>, Walz J.S.<sup>3,1,2,4</sup>

<sup>1</sup>University Hospital Tübingen, Clinical Collaboration Unit Translational Immunology, Tübingen, Deutschland, <sup>2</sup>University of Tübingen, Cluster of Excellence iFIT (EXC2180) "Image-Guided and Functionally Instructed Tumor Therapies", Tübingen, Deutschland, <sup>3</sup>University and University Hospital Tübingen, Peptide-based Immunotherapy, Tübingen, Deutschland, <sup>4</sup>University of Tübingen, Institute for Cell Biology, Department of Immunology, Tübingen, Deutschland, <sup>5</sup>UCT-University Cancer Center Frankfurt, Institute of Clinical Cancer Research, Krankenhaus Nordwest, Frankfurt, Deutschland, <sup>6</sup>Charité-Universitätsmedizin Berlin, Department of Hematology, Oncology and Cancer Immunology, Campus Benjamin Franklin, Berlin, Deutschland, <sup>7</sup>University Hospital Tübingen, Department of Obstetrics and Gynecology, Tübingen, Deutschland, <sup>8</sup>Dr. Margarete Fischer-Bosch-Institute for Clinical Pharmacology, Stuttgart, Deutschland, <sup>9</sup>University Hospital Tübingen, Department of Clinical Pharmacology, Tübingen, Deutschland, <sup>10</sup>EMC Microcollections GmbH, Tübingen, Deutschland, <sup>11</sup>University Hospital Tübingen, Department of Otorhinolaryngology, Head & Neck Surgery, Hearing Research Centre, Tübingen, Deutschland, <sup>12</sup>University Hospital Tübingen, Institute for Clinical Chemistry and Pathobiochemistry, Department for Diagnostic Laboratory Medicine, Tübingen, Deutschland, <sup>13</sup>University Hospital Tübingen, Institute for Clinical Epidemiology and Applied Biometry, Tübingen, Deutschland, <sup>14</sup>Robert Bosch Hospital, Robert Bosch Society for Medical Research, Stuttgart, Deutschland, <sup>15</sup>Dr. Margarete Fischer-Bosch-Institute for Clinical Pharmacology, Stuttgart, Stuttgart, Deutschland, <sup>16</sup>German Cancer Consortium (DKTK) and German Cancer Research Center (DKFZ), partner site Tübingen, Tübingen, Deutschland

T-cell immunity is central for the control of COVID-19 in particular in patients incapable to mount humoral immune response, comprising cancer patients with disease- or treatment-related B-cell deficiency. We have previously reported on the favorable safety profile and efficacy in terms of induction of SARS-CoV-2-specific T-cell responses by CoVac-1, a peptide-based T-cell activator, composed of SARS-CoV-2 T-cell epitopes derived from various viral proteins, combined with the toll-like receptor 1/2 agonist XS15 emulsified in Montanide<sup>TM</sup> ISA51 VG. In the Phase I trial in healthy adults (NCT04546841), CoVac-1 induced profound and long-lasting T-cell immunity after single dose administration in 100% of participants, mediated by multifunctional Th1 CD4<sup>+</sup> and CD8<sup>+</sup> T cells. CoVac-1-induced T-cell responses surpassed those after SARS-CoV-2 infection as well as after vaccination with approved vaccines and were unaffected by SARS-CoV-2 variants of concern (VOC, Heitmann *et al.* Nature 2021).

We here report on a Phase I/II open-label trial (NCT04954469), recruiting 54 patients with congenital or acquired B-cell deficiency, who received one single subcutaneous dose of CoVac-1. The immunogenicity in terms of CoVac-1-induced T-cell response until day 28 was

the primary endpoint, while safety analyzed until day 56 was assessed as secondary endpoint.

92 % of study patients suffered from cancer-related B-cell deficiency, with chronic lymphocytic leukemia (30%), mantle cell lymphoma (26%) and follicular lymphoma (20%) being the most common diagnoses. No serious adverse events and no grade 4 adverse events were observed. Expected local granuloma formation was observed in 95% of study subjects, while systemic reactogenicity was absent or mild. SARS-CoV-2-specific T-cell responses were induced in 86% of these highly immunocompromised study patients and directed to multiple CoVac-1 peptides, not affected by any of current Omicron variants and mediated by multifunctional T-helper 1 CD4<sup>+</sup>T cells. CoVac-1-induced T-cell responses exceeded spike-specific T-cell responses after vaccination with mRNA vaccines in B-cell deficient patients as well as those in immunocompetent COVID-19 convalescents with and without seroconversion.

Together, CoVac-1 induces broad and potent T-cell responses in patients with B-cell/anti-body deficiency independently of current variants of concern, with a favorable safety profile. Our data warrant advancement to a pivotal Phase II/III safety and efficacy evaluation.

**V870 - INTRIGUE: Eine randomisierte, offene Phase-3-Studie zur Bewertung der Wirksamkeit und Sicherheit von Ripretinib im Vergleich zu Sunitinib bei Patienten mit fortgeschrittenem gastrointestinalem Stromatumor, die zuvor mit Imatinib behandelt wurden /**  
**INTRIGUE: A phase 3, randomized, open-label study to evaluate the efficacy and safety of ripretinib vs sunitinib in patients with advanced gastrointestinal stromal tumor previously treated with imatinib**

**Sebastian Bauer (Essen, D)**

*Bauer S.<sup>1</sup>, Jones R.L.<sup>2</sup>, Gelderblom H.<sup>3</sup>, George S.<sup>4</sup>, Schöffski P.<sup>5</sup>, von Mehren M.<sup>6</sup>, Zalberg J.<sup>7</sup>, Kang Y.-K.<sup>8</sup>, Razak A.A.<sup>9</sup>, Trent J.<sup>10</sup>, Attia S.<sup>11</sup>, Le Cesne A.<sup>12</sup>, Su Y.<sup>13</sup>, Meade J.<sup>13</sup>, Wang T.<sup>13</sup>, Sherman M.L.<sup>13</sup>, Ruiz-Soto R.<sup>13</sup>, Blay J.-Y.<sup>14</sup>, Heinrich M.C.<sup>15</sup>*

<sup>1</sup>Sarcoma Center/West German Cancer Center, University Hospital Essen, University Duisburg-Essen and German Cancer Consortium (DKTK), Partner Site University Hospital Essen, Medical Oncology, Essen, Deutschland, <sup>2</sup>The Royal Marsden NHS Foundation Trust and Institute of Cancer Research, Sarcoma Unit, London, Vereinigtes Königreich, <sup>3</sup>Leiden University Medical Center, Leiden, Niederlande, <sup>4</sup>Dana-Farber Cancer Institute, Boston, Vereinigte Staaten, <sup>5</sup>University Hospitals Leuven, Leuven Cancer Institute, KU Leuven, Department of General Medical Oncology, Leuven, Belgien, <sup>6</sup>Fox Chase Cancer Center, Philadelphia, Vereinigte Staaten, <sup>7</sup>Monash University and Alfred Health, Medical Oncology, Melbourne, Australien, <sup>8</sup>Asan Medical Center, University of Ulsan, Seoul, Korea, Republik, <sup>9</sup>Toronto Sarcoma Program, Princess Margaret Cancer Center, Toronto, Kanada, <sup>10</sup>Sylvester Comprehensive Cancer Center, University of Miami Health System, Miami, Vereinigte Staaten, <sup>11</sup>Mayo Clinic, Jacksonville, Vereinigte Staaten, <sup>12</sup>Gustave Roussy, Villejuif, Frankreich, <sup>13</sup>Deciphera Pharmaceuticals, LLC, Waltham, Vereinigte Staaten, <sup>14</sup>Centre Léon Bérard, Lyon, Frankreich, <sup>15</sup>Portland VA Health Care System and Knight Cancer Institute, Portland, Vereinigte Staaten

**Background:** Sunitinib is approved for advanced gastrointestinal stromal tumor (GIST) after imatinib failure. Ripretinib, a broad-spectrum KIT and PDGFRA switch-control tyrosine kinase inhibitor (TKI), is indicated for the treatment of adult patients (pts) with GIST who received prior treatment with 3 or more TKIs, including imatinib. We compared the efficacy and safety of ripretinib vs sunitinib in pts with advanced GIST who progressed on or were intolerant to imatinib.

**Methods:** This multicenter, global, randomized, open-label phase 3 study (NCT03673501) enrolled adult pts with GIST who progressed on or had intolerance to imatinib. Pts were randomized 1:1 to ripretinib 150 mg once daily (QD) or sunitinib 50mg QD (4wks on/2wks off). Randomization was stratified by *KIT* mutational status and imatinib intolerance. Primary endpoint was progression-free survival (PFS). Key secondary endpoints were objective response rate (ORR) and overall survival (OS). Hierarchical testing was performed for primary and key secondary endpoints in a prespecified sequence; testing pts with a *KIT* exon 11 primary mutation (Ex11 intention-to-treat [ITT] population) preceded the all-patient (AP) ITT population. Data cutoff was 1 Sep 2021; final analyses of PFS and ORR and the first interim analysis of OS were conducted.



**Results:** A total of 453 pts were randomized to ripretinib (n=226; Ex11 ITT, n=163) or sunitinib (n=227; Ex11 ITT, n=164). Median age was 60 yrs (range 18–88); most pts were white (66.2%) males (62.0%). PFS was not statistically different for ripretinib vs sunitinib in the Ex11 ITT (hazard ratio [HR] 0.88, 95% CI 0.66, 1.16;  $P=0.36$ ; median 8.3 vs 7.0 mos) or in the AP populations (HR 1.05, 95% CI 0.82, 1.33;  $P=0.72$ ; median 8.0 vs 8.3 mos). ORR was numerically higher for ripretinib vs sunitinib in the Ex11 ITT (23.9% vs 14.6%; 95% CI 0.7, 17.8; nominal  $P=0.03$ ) and AP ITT populations (21.7% vs 17.6%; 95% CI –3.2, 11.5; nominal  $P=0.27$ ). OS data was highly immature; median OS was not reached in either arm. Fewer pts in the ripretinib arm experienced Grade 3-4 (G3-4) treatment-emergent adverse events (TEAEs) vs sunitinib (41.3% vs 65.6%).

**Conclusion:** The PFS in both arms was longer than PFS achieved by sunitinib in its pivotal phase 3 trial. While PFS for ripretinib did not meet the primary endpoint of superiority vs sunitinib, meaningful clinical activity and fewer G3-4 TEAEs were observed in pts with advanced GIST treated with ripretinib after imatinib failure.

# **V871 - Super-Enhancer Analyse definiert IRF4 als Zielstruktur im Anaplastisch-großzelligem Lymphom und rationales Design von IRF4 Degradier Molekülen / Targeting Core Super-Enhancer Circuitries of Anaplastic Large Cell Lymphoma by Rational Design of IRF4 Degradier Drugs**

**Catello Giordano (Vienna, A)**

*Giordano C.<sup>1</sup>, Prutsch N.<sup>2</sup>, Gurnhofer E.<sup>1</sup>, Aplenc A.<sup>1</sup>, Kenner L.<sup>3,1</sup>, Look A.T.<sup>2</sup>, Touaibia M.<sup>4</sup>, Merkel O.<sup>1</sup>*

<sup>1</sup>Medical University of Vienna, Department of Pathology, Wien, Österreich, <sup>2</sup>Boston Children's Hospital, Dana-Farber Cancer Institute, Department of Pediatrics, Boston, Vereinigte Staaten, <sup>3</sup>University of Veterinary Medicine Vienna, Unit of Laboratory Animal Pathology, Wien, Österreich, <sup>4</sup>Université de Moncton, Department of Chemistry and Biochemistry, Moncton, Kanada

Anaplastic Large Cell Lymphoma (ALCL) is an aggressive CD30+ non-Hodgkin T-cell lymphoma that is characterized by the presence of the typical NPM-ALK fusion protein in half of the patients. Despite the high efficiency of CHOP chemotherapy and the availability of effective second generation ALK inhibitors like ceritinib, about 30% of patients relapse with dire prognosis. By using H3K27ac ChIP-seq analysis in ALCL tissue of primary patients as well as in ALCL cell lines we identified IRF4, BATF3, JunB and IKZF1 as ALCL specific, highly expressed super-enhancer transcription factors. We hypothesized that inhibiting these transcription factors would perturb ALCL growth, however transcription factors have been notoriously difficult to target by small molecules. The immunomodulatory multiple myeloma drug lenalidomide has recently been shown to be a specific degrader of IKZF1 by facilitating binding to the cereblon ubiquitin ligase but has no effect on ALCL cells. In contrast, we describe here a small molecule CM that leads to sequential degradation of first IRF4 and then IKZF1 and finally apoptosis induction in ALCL cell lines. Inspired by bee glue derived substances, CM was developed through multiple rounds of chemical modifications and *invitro* testing establishing clear structure function relationships. RNA-seq analysis after challenge with CM revealed activation of oxidative stress and unfolded protein response pathways. Finally, significant tumor size reduction was observed in murine ALCL engraftment models that received CM by oral gavage over two weeks. In the future we want to test CM in PDX models of ALCL or other lymphoma types that have been shown to highly express IRF4 including diffuse large B-cell lymphoma, multiple myeloma, Burkitt lymphoma or Hodgkin disease. Thus, we have identified a specific, orally available degrader drug for the IRF4 transcription factor in ALCL that may be useful also in other lymphoma types.



# V873 - Einzelzell-MultiOmic Analyse der Stamm- und Progenitorzellen von CHIP Knochenmark gibt Hinweise auf maligne Transformation im Frühstadium /

## Multiomic analysis of CHIP bone marrow hematopoietic stem/progenitor cells reveals a potential early stage of malignancy

**Mark van der Garde (München, D)**

*van der Garde M.<sup>1,2</sup>, Thomas M.<sup>3</sup>, Rivière J.<sup>1</sup>, Metzeler K.H.<sup>2,4</sup>, Bassermann F.<sup>1</sup>, Hecker J.<sup>1</sup>, Marr C.<sup>3</sup>, Götze K.<sup>1</sup>*

<sup>1</sup>Klinikum Rechts der Isar, Technische Universität München, Klinik und poliklinik für Innere Medizin III, München, Deutschland, <sup>2</sup>German Cancer Consortium (DKTK), Partner Site Munich, Heidelberg, Deutschland, <sup>3</sup>Helmholtz Zentrum München–German Research Center for Environmental Health, Institute of Computational Biology, Neuherberg, Deutschland, <sup>4</sup>University Hospital Leipzig (UHL), Department of Hematology and Cell Therapy, Leipzig, Deutschland

**Introduction:** Clonal hematopoiesis of indeterminate potential (CHIP) increases the risk to develop hematological cancers such as acute myeloid leukemia (AML) tenfold. Since AML has been shown to originate from a hematopoietic stem/progenitor cell (HSPC) we set out to compare the transcriptome and chromatin accessibility profile of bone marrow HSPCs from a healthy donor and a CHIP carrier.

**Methods:** Single cell RNA seq and ATAC seq libraries were prepared from isolated CD34+ cells of a healthy and CHIP donor (mutations in DNMT3A, 1.1% VAF and TET2, 5.3% VAF) according the protocol of 10X Genomics (Fig 1a). Sequencing was performed on a Novaseq, and data was analyzed with R. HSPC subpopulations were defined by analysis of conserved genes in each cluster after dimensional reduction with UMAP (Fig 1b, c).

**Results:** Cell frequency analysis of the HSPC subpopulations showed that the proportion of HSC was twofold higher in the CHIP donor whereas the lymphocyte progenitor and pre-B cells compartment was fourfold lower (Fig 1d), indicating an overall myeloid bias in the CHIP donor.

Gene Ontology analysis of differentially expressed genes in the HSC compartment showed an upregulation of genes involved in metabolic processes in CHIP HSCs, among which genes that are associated with the onset of AML such as MEIS1, PBX1 and PBX3 (Fig 1d). Furthermore, the expression levels of genes that are involved in niche retention, such as CD44 and VLA-4 were significantly higher in CHIP HSCs (Fig 1e), suggesting alterations in the architecture of CHIP bone marrow.

In addition to differences in gene expression, chromatin accessibility analysis showed unique genomic peaks in the CHIP HSC. Analysis of these sequences for binding motifs of transcription factors showed an overabundance of GATA, CEBP and RUNX1 motifs (Fig 1f), all believed to play major roles in the onset of AML.

**Conclusions:** The transcriptional landscapes of HSPC from healthy and CHIP donors show distinct differences, indicating that even the absence of signs of hematological disease,

**a**

CHIP  
72 years  
BMI 24.3  
CH mutations: TET2, DNMT3A

Healthy  
76 years  
BMI 30.1

Archived frozen single cells from biobank

CD34+ sorting

Stem and progenitor cells

Single cell sequencing

Gene expression  
Chromatin accessibility

**b**

scRNA

scATAC

- Stem cells
- Myeloid lymphoid progenitor
- Megakaryocyte erythroid progenitors
- Megakaryocyte progenitors
- Erythroid progenitors
- Granulocyte progenitors
- Eosinophil basophile progenitors
- Dendritic monocyte progenitors
- Dendritic progenitors
- Monocyte progenitors
- Lymphocyte progenitors
- Lymphocyte progenitors late preB

28% 14% 13% 13% 9% 7% 6% 10%

