In **WP 1.1 (Genome sequencing)**, complete genetic inventories of the rat BN-Lx and SHR strains (founders of the recombinant inbred panel) were published in the open access journal Genome Biology (Simonis (2012), Genome Biology, 13:r31). The effects of this variation on transcriptional characteristics were described. Complete inventories of the founder strains of the rat heterogeneous stock (8 strains) were compiled and used to identify the genetic basis of a wide range of phenotypic traits measured in more than 1,600 derived HS panel animals (Baud et al, Nature Genetics (2013), 45: 767-75). Genome sequencing has been extended to total of 37 rat inbred strains. A describtion of the genome analysis of 27 rat strains as well as the selective processes underlying shared phentoypic traits is published in Cell (Atanur et al. (2013), Cell 154:691-703).

In **WP 1.2 (Transcriptome)**, alternative splicing was quantified on the exon level in liver and left ventricle tissue in the RI strains and parental strains. Expression levels of microRNA were quantified in liver and left ventricle tissue in the RI strains and parental strains. Transcriptome data on exon and gene level were integrated with genetic data and suggests that 30% of RNA-seq eQTL which not only affect the global gene expression level, but also cause differential alternative transcript events in their target genes, preferentially in the last exon. Transcriptome data of microRNA and mRNA were integrated with genetic data and suggests that only a small proportion of mRNA eQTL are regulated directly by microRNA. Results imply that alternative polyadenylation events are a common mechanism to regulate eQTL transcripts

In **WP 1.3 (Epigenetics)**, new human antibodies against transcription factors showing reactivities in rat were obtained, some showing immunoreactivity to highly specific cell types. Some of these factors shuttle between the cytoplasm and the nucleus, potentially regulating gene expression. The genome-wide maps for histone methylation were completed for the parental strains and the RI lines in the liver and left ventricles and in the liver for Pol II. Using the ChIp-seq maps for modified histones and Pol II, allele-specific distribution of the marks or the enzyme were identified. A significant percentage of PolII binds loci associated with H3K4me3 or H3K4me1 has been observed outside known annotated genes in liver. Computational algorithms were developed to integrate genome-wide chromatin state maps, histoneHMM designed to facilitate statistical comparisons of two ChIP-samples and chromSTAR designed to facilitate a simultaneous analysis of a large number of epigenetic marks collected on the same sample. PolII data was integrated with histone mark and gene expression in liver where almost every PolII enriched proximal promoter regions also exhibit enrichment of H3K4me3, by contrast with H3K4me3 which can be enriched in some proximal promoter regions in absence of PolII.

In **WP 1.4 (Proteomics)**, quantitative tissue analysis of various rat tissue samples derived from

the both RI and parental strains were performed. Collectively these analyses have currently identified >10,000 rat proteins differential protein expression between RI liver lines and parental rat. An in-depth proteomic analysis of liver-specific protein expression in parental strains was conducted. Multilevel analysis identified a genomic variant in the promoter of the most differentially expressed gene Cyp17a1. Proteome quantification of 30 RI lines and the investigation of DNA protein interactions of SNPs are ongoing.

In **WP 1.5 (Metabonomics)**, a pipeline for in vitro validation of the biological effects of metabolites in cell lines was developed. ShRNA was applied for the analysis of the biological effects of enzyme dowregulation in vitro in 3T3 adipocytes. A UPLC-MS based tissue metabolite phenotyping pipeline for BHX/HXB RI strains was developed. Novel chemoinformatic strategies were developed for processing NMR-based and MS-based data and mQTL mapping (2 technical papers in preparation). Two candidate genes, Asns and Galm, were identified in the BN:GK panel by an integrative mQTL/eQTL analysis and their functional relevance was confirmed in cell-based knock-down assays. mQTL mapping of tissue-specific UPLC-MS metabotypes for 5 RI panel tissues, showed that >50% of metabotypes are heritable (polygenic and monogenic heritability) and that 50% of liver mQTLs were replicated in the heart, suggesting the existence of conditional mQTLs in tissues. Several candidate metabolites and candidate genes are currently being shortlisted for further functional validation.

In **WPs 1.6/2.1 (Regulatory networking and computational infrastructure)**, the immunological disease portal is online. We built new SNP tracks within the Rat Genome Database to display are EURATRANS data. Data mining tools for mapping discovered variations to network components and the Pinball algorithm as a mechanism for direct integration free of alignment bias and also largely free of artefacts caused by errors or incomplete sections of the reference genome assembly were developed. Substantial heritability of metabolites abundance in the RI strains were determined and hundreds of genetic control points for metabolite abundance (mQTLs) across five rat tissues mapped, identifying several cluster of metabolites controlled by the same genetic loci.

In **WP 2.2 (Functional validation)**, four novel shRNA transgenic rat lines have been developed and all show inducibility of the knockdown effect and interesting phenotypes and may become valuable novel rat models for diseases. The generation of transgenic rats by transposon technology is now well established and very efficient, a result which was prominently published by several consortium members (Katter et al., FASEB J 2013). The rederivation of two functional rat mutants from the ENU archive has successfully been achieved. Since the phenotypes of the established mutants resemble human disorders (epilepsy and metabolic syndrome), their usability for translational research has already been proven. The derivation of a knockout rat by homologous recombination in ES cells. The HGPRT gene was ablated and created a model for the Lesch-Nyhan syndrome.

In **WP 3.1 (Comparatie analysis)**, a large number of eQTLs can be mapped in the HS and biologically meaningful gene networks have been identified. Novel multi-trait methods to increase genetic mapping resolution and power were developed. A novel gene involved in anxiety was identified.

In **WP 3.2 (Integration with GWAS)**, we significantly augmented the annotation, visualisation and underlying data resources for the human GWAS positive regions. An integrated genomics, transcriptomics and proteomics analysis for BN-Lx and SHR liver has identified 41 genes that were differentially expressed at both transcriptome and proteome level, one of which was identified as a top hit in relation to blood pressure and hypertension in human genome-wide association studies.

In **WP 3.3 (Validation)**, high-throughput expression studies and multimodel/pathway analysis are ongoing. Initial analysis of the RNAseq data in human heart biopsies has identified two genes and candidate genes for heart disease were identified through translational investigations.

(31.03.2014)