

# Immunoprecipitation Sequencing (IP-seq)

IP-seq is used to study the interaction between proteins and nucleic acids by sequencing the protein-binding motifs within DNA and RNA. Depending on the different research objectives, it can include chromatin immunoprecipitation (ChIP) and RNA immunoprecipitation (RIP). Novogene takes DNA or RNA fragments enriched by antibodies specific to the protein of interest, maps them against reference genomes and analyses to call peaks. Our service delivers high quality data, publication-ready analysis results, and personalised pipelines.

## Why Novogene?



**Extensive experience with over 3000 projects**



**Industry-leading data quality guarantee**



**In house pipeline to meet different analysis requirement**

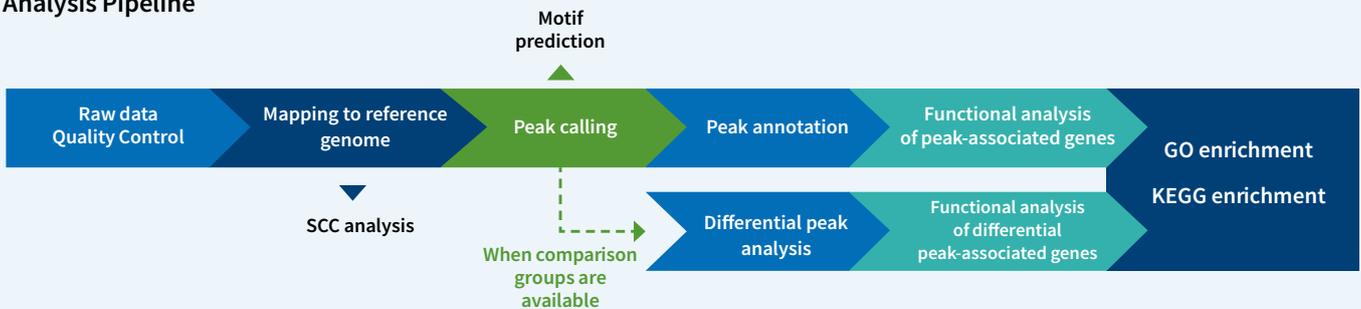
## Sample requirements

Sample Type	Amount	Volume	Concentration	Purity
Immunoprecipitated DNA	≥ 50 ng	≥ 20 µL	≥ 2 ng/µL	Main peak of 100 bp - 500 bp
Immunoprecipitated RNA	≥ 100 ng	≥ 20 µL	≥ 3 ng/µL	Without fragmentation; fragments should be longer than 1000 bp

## Sequencing parameters

Service	Platform	Sequencing strategy	Recommended data amount	Data quality
ChIP-seq	Illumina NovaSeq 6000	Pair-end 150	≥ 20M read pairs per sample for transcription factor studies.	Q30 ≥ 85%
			≥ 40M read pairs per sample for histone studies.	
RIP-seq			≥ 20M read pairs per sample for species with reference genome.	

## Analysis Pipeline



## Publications using Novogene's expertise



**Cell Death & Disease**, 2020.  
LncRNA MNX1-AS1 promotes progression of intrahepatic cholangiocarcinoma through the MNX1/Hippo axis

**bioRxiv**, 2020.  
A study of ALK-positive pulmonary squamous-cell carcinoma: From diagnostic methodologies to clinical efficacy.

**Molecular Cell** 2019.  
Co-transcriptional Loading of RNA Export Factors Shapes the Human Transcriptome

**PNAS**, 2019.  
Coupling of COPII vesicle trafficking to nutrient availability by the IRE1α-XBP1s axis

**Cell**, 2019.  
A Natural Allele of a Transcription Factor in Rice Confers Broad-Spectrum Blast Resistance

**Nucleic Acids Research**, 2020.  
The m6A reader YTHDF1 promotes ovarian cancer progression via augmenting EIF3C translation.

**HEPATOLOGY**, 2017.  
RNA N6-methyladenosine methyltransferase-like 3 promotes liver cancer progression through YTHDF2-dependent posttranscriptional silencing of SOCS2

### mRNA N6-methyladenosine methylation of postnatal liver development in pigs.

He et al., 2017. PLoS ONE. DOI: doi:10.1371/journal.pone.0173421.

**Research objective:**

To explore the impact of N6-methyladenosine (m6A) methylation patterns of RNA on gene expression in porcine liver at different developmental stages.

**Sample collection:**

Methylated RNA collected from newborn, suckling, and adult stage porcine liver.

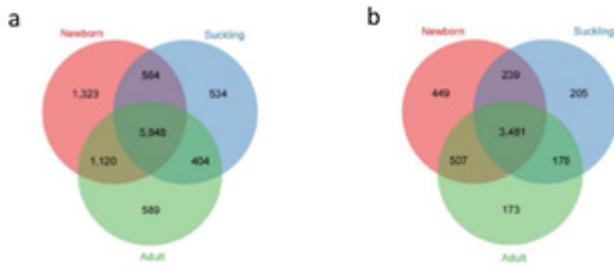
**Sequencing strategy:**

PE150 on Illumina NovaSeq 6000.

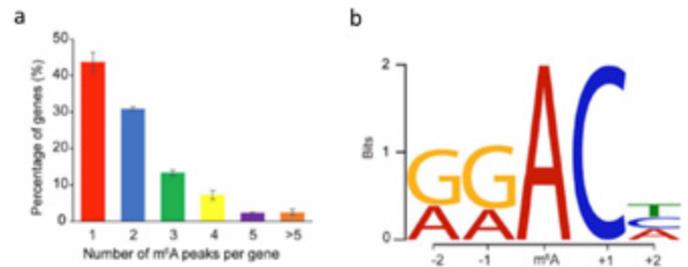
Data amount: > 9 Gb raw data (30M reads) per sample.

Hundreds of genes with differential expression and m6A patterns in each of the 3 development stages were identified. There was found to be a negative correlation between m6A modification and gene expression, suggesting that m6A modification may increase RNA stability of transcripts that are essential but expressed at low levels.

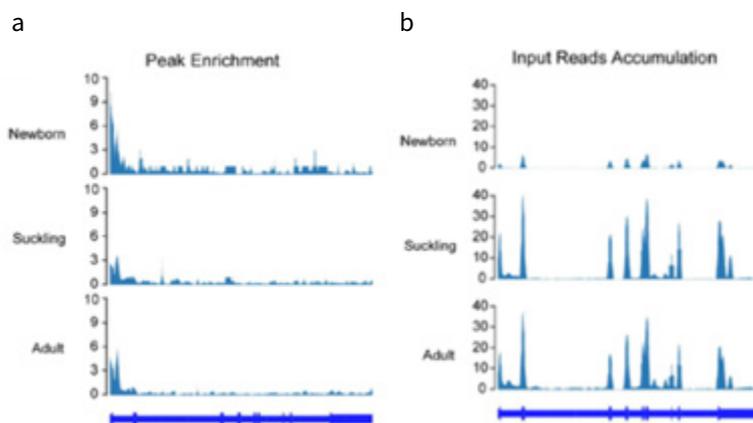
The identified genes were mapped to pathways crucial for liver function at each of the 3 stages e.g. bile acid biosynthesis in newborn piglets for the metabolism of nutrients provided by the placenta; glycosaminoglycan metabolism in suckling piglets for metabolism of milk components; and fatty acid transport and lysine degradation in adult pigs, required for the metabolism of a varied adult diet. In summary, genes with a high degree of m6A methylation showed enriched terms consistent with liver function at a given developmental stage.



**Figure 1**  
Venn diagram showing the overlap of (a) m6A peaks and (b) m6A modified genes in newborn, suckling, and adult samples.



**Figure 2**  
(a) m6A peak distribution in genes; (b) The most common consensus motifs in the m6A peaks.



**Figure 3**  
Venn diagram showing the overlap of (a) m6A peaks and (b) m6A modified genes in newborn, suckling, and adult samples.

The balance of expression of genes involved in central metabolism was controlled by m6A methylation at each of the 3 stages. Figure 3 highlights the opposite trends in m6A methylation (a) and gene expression (b) of GATM, a gene involved in glycine and threonine metabolism. Higher levels of methylation at the newborn stage suppress expression, as the function of this gene is not required. Methylation decreases with age to allow expression and functioning of the gene.

## Conclusions

As growing conditions and diets change in the 3 developmental stages, the transcriptome of the porcine liver adapts to metabolic requirements. m6A methylation plays a role in the negative regulation of post-transcriptional gene expression at the epitranscriptomic level.

Novogene (UK) Company Limited  
25 Cambridge Science Park  
Cambridge, CB4 0FW  
United Kingdom  
www.novogene.com  
info@novogene-europe.com

For more details on our services  
and to download product flyers  
visit: [om.novogene.com/flyers](http://om.novogene.com/flyers)



Search 'Novogene Europe'