nu·Q discover

A valuable tool from target identification to validation in clinical studies.

Empowering drug developers and scientists through a range of state-of-the-art assays for rapid epigenetic profiling in disease, model development, preclinical testing, and clinical studies – from discovery to market ready.

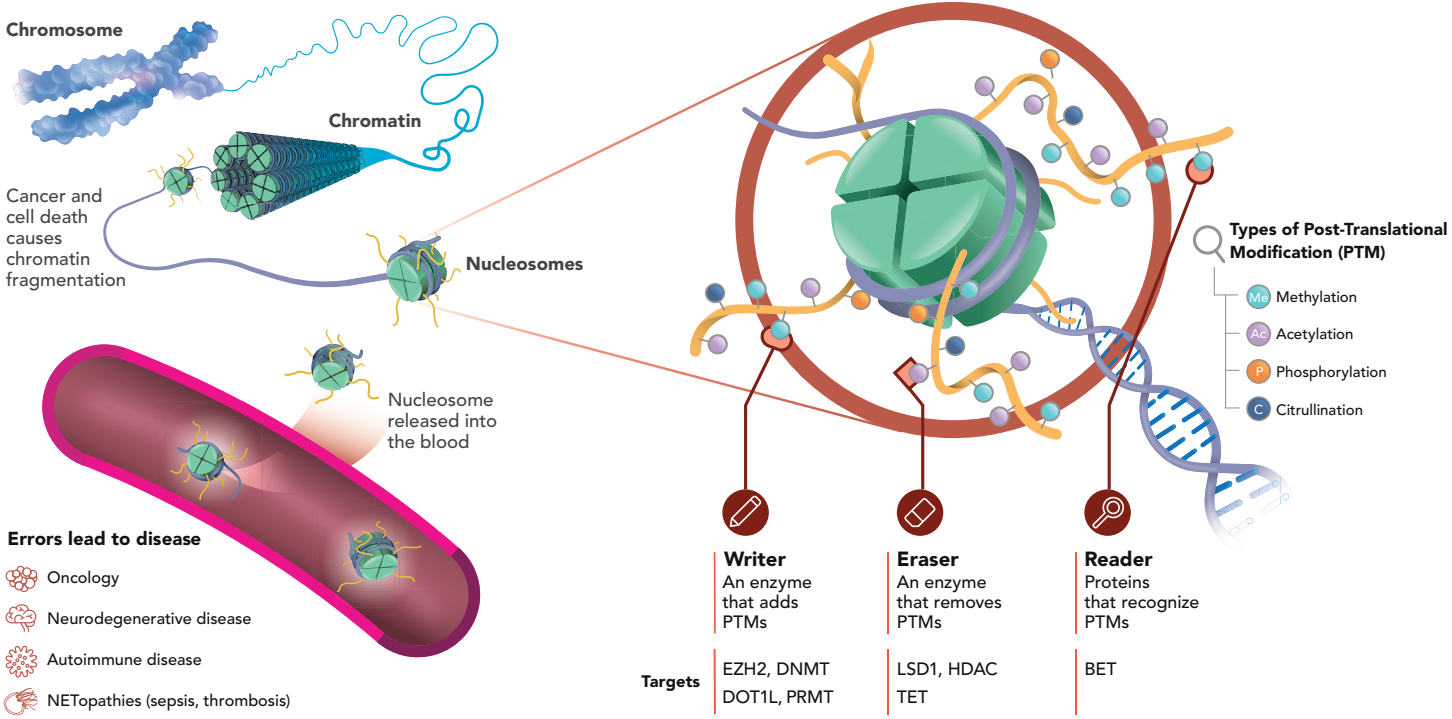
Including research in

- Oncology
- Neurodegenerative diseases
- Autoimmune diseases
- NETopathies

For research use only. Not for use in diagnostic procedures.

Volition

Measuring and monitoring nucleosome levels and modifications in circulating blood has the potential to aid diagnosis, prognosis and monitoring of many human diseases.



Adapted from: Li X and Li XD. Integrative Chemical Biology Approaches to Deciphering the Histone Code: A Problem-Driven Journey. Acc Chem Res 2021 54(19), 3734-3747; Regnier FE, Kim J. Proteins and Proteoforms: New Separation Challenges. Anal Chem 2018 Jan 2;90(1):361-373

Nu.Q® adds value across the Drug Development pipeline.

With access to a broad range of state-of-the-art assays built on our proprietary Nucleosomics™ platform, you can answer your drug development, pre-clinical and clinical questions.

Target Identification and Validation	**Identify potential drug targets** by measuring levels of specific histone modifications
Lead Discovery	**Screen and identify lead compounds** that modulate the levels of specific histone modifications
Preclinical Studies	Monitor changes in histone modification levels to **assess the efficacy and Mechanism of Action (MoA) of compounds**
Clinical Studies	Monitor target engagement to determine the **pharmacodynamics (PD) of drugs in patients**
Monitoring	Demonstrates dose/time response in **disease/treatment monitoring**

Identify

- Drug targets/novel inhibitors
- MoA
- Biomarkers

Demonstrate

- PD monitoring of target engagement
- On/Off target activities and effects
- Correlation with gene expression and cancer progression
- Efficacy: in-vitro/in-vivo

Accelerate

- Use same assay for drug screen on cell culture to clinical studies
- Demonstrated in a variety of cells and animal models
- Combine assays for a comprehensive overview

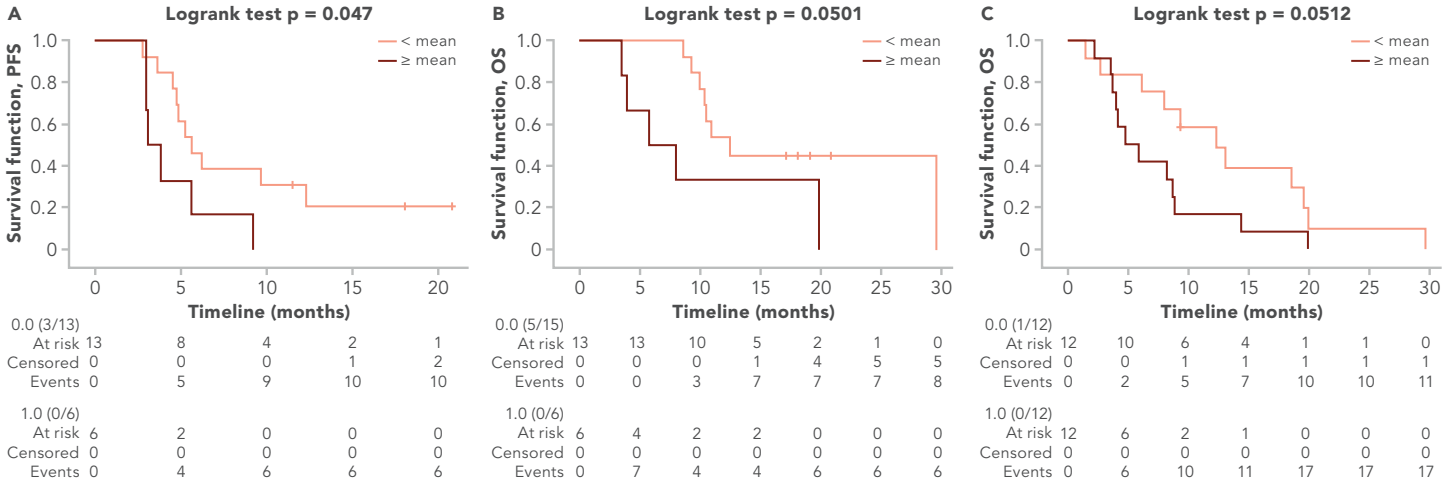
Abbreviations: MoA, mechanism of action; PD, pharmacodynamics.

Our Assays in Action.

Predictive significance of Nu.Q®
Discover assays in hepatocellular carcinoma patients treated with sorafenib

Kaplan–Meier curves for progression-free and overall survival according to H3K27me3/H3K36me3 ratio values at best response and progressive disease

- Kaplan–Meier curves for (A) PFS according to H3K27me3/H3K36me3 ratio mean values at BR, (B) OS according to H3K27me3/H3K36me3 ratio mean values at BR and (C) OS according to H3K27me3/H3K36me3 ratio median values at PD. Data for all curves compared using log rank test.

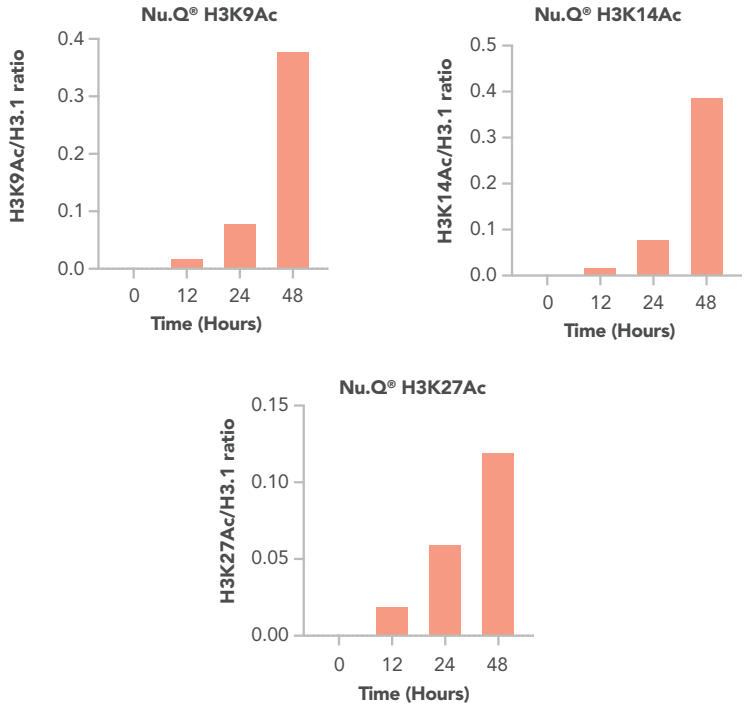


Abbreviations: BR, best response; OS, overall survival; PD, progressive diseases; PFS, progression-free survival

Reference: Salani F et al. Predictive significance of circulating histones in hepatocellular carcinoma patients treated with sorafenib. Epigenomics 2022 14(9), 507–517

Nu.Q® Discover Assays*.

- In this study we evaluated numerous Nu.Q® Discover Assays on the K562 leukemia cell line treated with a variety of drugs targeting epigenetic regulatory enzymes. Our goal was to demonstrate the ability of Nu.Q® Discover to discern fluctuations in epigenetic marks following drug treatment, utilising this widely employed cell line.
- We demonstrated that Nu.Q® Discover assays can show changes in histone PTM levels in a time and dose dependent manner following treatment with drugs targeting histone methyltransferases, histone demethylases as well as histone deacetylases.
- This investigation confirms the suitability of the Nu.Q® Discover assay as a reliable tool for elucidating epigenetic modifications and their potential implications in cellular responses to drug treatments.



Histone PTM Changes in response to treatment with the HDACi Valproic Acid. Time course of increasing acetylation at a variety of histone residues upon treatment with Valproic Acid.

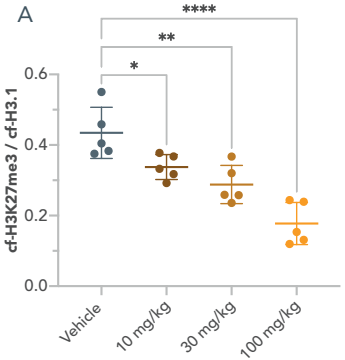
Reference: Data on file

Case Study.

Leveraging our cutting-edge Nu.Q® platform, a compelling use case has demonstrated how Volition's Nu.Q® Discover cell-free (cf)-nucleosomal H3K27me3 assay enables robust, quantitative analysis of histone modifications in circulating cf-nucleosomes.

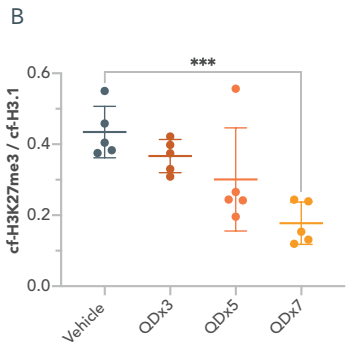
- ORIC-944 induced a dose- and time-dependent H3K27me3 reduction in cf-nucleosomes in-vivo.
- Since dying tumor cells release nucleosomes to circulation, cf-nucleosomal H3K27me3 levels normalized to cf-H3.1 were assessed in plasma using a magnetic bead-based sandwich immunoassay (Belgian Volition SRL). 22Rv1-tumor bearing mice were treated with Vehicle QDx7, 100 mg/kg ORIC-944 QDx3, QDx5 or QDx7, and 10 or 30 mg/kg ORIC-944 QDx7. Shown are average cf-nucleosomal H3K27me3 levels normalized to cf-H3.1, error bars indicate SD, with n=5/group. One-way ANOVA, followed by Dunnett's multiple comparison test, was used to compare ORIC-944 groups to Vehicle. *, p<0.05; **, p<0.01; ***, p<0.001; ****, p<0.0001 vs. Vehicle.
- Our Nu.Q® Discover assays will be used in the human clinical trial setting to monitor response.

ORIC-944 decreased cf-H3K27me3 levels after 7 days in a dose-dependent manner



A. Normalized cf-H3K27me3 levels significantly decreased with increasing dose (one-way ANOVA test for linear trend, p<0.0001).

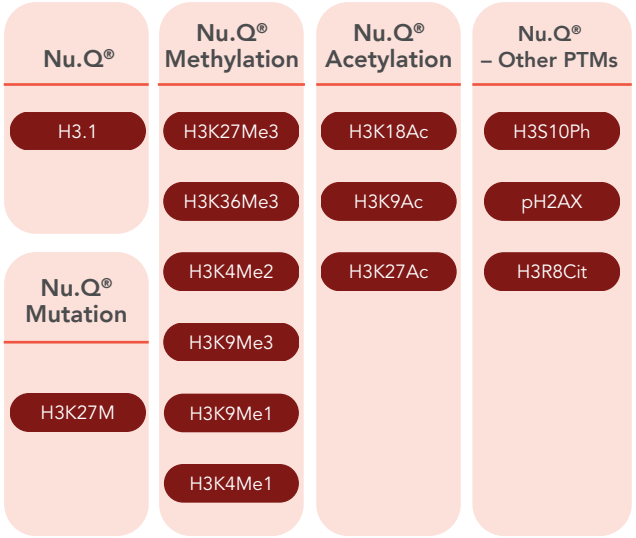
ORIC-944 decreased cf-H3K27me3 levels with 100 mg/kg in a time dependent manner



B. Normalized cf-H3K27me3 levels significantly decreased with increasing treatment duration (p=0.0024).

Reference: Daemen A et al. Biomarker strategy for a phase 1 study of ORIC-944, a potent and selective allosteric PRC2 inhibitor, in patients with metastatic prostate cancer [abstract]. In: Proceedings of the American Association for Cancer Research Annual Meeting 2023; Part 1 (Regular and Invited Abstracts); 2023 Apr 14-19; Orlando, FL. Philadelphia (PA): AACR; Cancer Res 2023;83(7_Suppl):Abstract nr 2791.

Nu.Q® Discover Assays*.



*Contact us for custom assay development at asknu.qdiscover@volition.com

Convenience:

- cf-nucleosome quantification technology run manually and on fully automated magnetic bead-based sandwich immunoassay ChLIA platform.
- No assay development required, assays ready to run.
- Easy to interpret report.
- Assays compatible with multiple animals (murine, lapine, porcine, canine and human) and cell models to provide constancy and confidence in results.

Sensitivity & Specificity:

- Low sample volumes. Use with EDTA plasma, cell culture extract, supernatant.
- Antibodies screened extensively to ensure antibodies have limited cross reactivity to non-targeted histone PTMs.
- Detection antibodies recognize a nucleosome specific epitope ensuring detection of only intact nucleosomes.
- Typical reproducibility:
  - Precision for Nu.Q® H3.1 intra-run less than 5%CV.
  - Precision for Nu.Q® H3.1 inter-run less than 10%CV.
- Lower limit of quantification: 3ng/ml for Nu.Q® H3.1.

Quality:

- Assays developed based on CLSI guidelines.
- Expert support for your histone PTM research needs.

Abbreviations: cf, cell-free, ChLIA, chemiluminescence immunoassay; CLSI, Clinical and Laboratory Standards Institute; CV, coefficient of variation; EDTA, ethylenediaminetetraacetic acid; PTM, post-translational modifications.

Volition

Volition is a multinational epigenetics company, powered by Nu.Q®, our proprietary nucleosome quantification platform. Our Nu.Q® Discover program enables drug developers and scientists access to a range of state-of-the-art assays for rapid epigenetic profiling in disease, model development, preclinical testing, and clinical studies.

Our expert team are on hand to offer guidance and support.

Get in touch for more information:

asknu.qdiscover@volition.com

volition.com

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For research use only. Not for use in diagnostic procedures.

CLSI

Developed based on CLSI standard