**REFERENCE NUMBER TO 32-00071** 

# A scalable and highly sensitive assay for measuring extracellular NAD<sup>+</sup> level in human body fluids

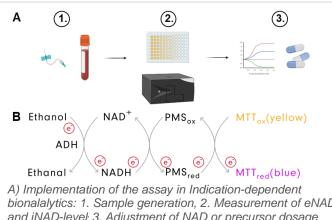
Keywords: nicotiname adenine dinucleotide, niacin, supplement, B3, pyridine nucleotides

#### INVENTION NOVELTY

Researchers at Charité Universitätsmedizin developed a novel, robust assay for detection of low extracellular NAD<sup>+</sup> (eNAD<sup>+</sup>) levels in human heparinized plasma and other bodily fluids. In contrast to existing gold-standard analytical methods like liquid chromatography-based mass spectrometry, this tool provides a simple, time- and cost-effective method to record eNAD<sup>+</sup> with a peerless sensitivity in the nanomolar range.

#### VALUE PROPOSITION

NAD<sup>+</sup> is a central co-enzyme involved in manifold cellular biochemical processes. NAD<sup>+</sup> levels are crucial for human health and monitoring of such is of high interest, especially in the field of aging and metabolic diseases. The proposed standardized method is suitable for high throughput measurements of eNAD<sup>+</sup> levels in human heparinized plasma. This highly sensitive colorimetric assay facilitates the quantification of eNAD<sup>+</sup> concentrations in the low micromolar, extending into the nanomolar range, and its protocol is ideally suited for translatory development of a direct-to-consumer lab testing product.



bionalalytics: 1. Sample generation, 2. Measurement of eNAD<sup>+</sup> and iNAD-level<sup>,</sup> 3. Adjustment of NAD or precursor dosage based on the levels B) Cycling principle of the assay. ADH: alceholdehydrogenase; PMS: peroxymonosulfate; ox: oxidzied; red: reduced

## TECHNOLOGY DESCRIPTION

The underlying methodology is based on the NAD+ dependent ADH catalyzed dehydrogenation of ethanol to ethanal. The inventors modified this colorimetric assay in an innovative fashion to accurately and reliably quantify very low eNAD<sup>+</sup> levels through a two-step cycling method in heparinized plasma samples. Notably, assay conditions can be adapted and miniaturized for high throughput analysis and for use in other bodily fluids. Because of its high-sensitivity and scalability, the proposed technology can be employed in a diverse range of applications, including as kit-format in research settings to replace timecost- and skill-demanding standard methods or as directto-consumer lab test.

## COMMERCIAL OPPORTUNITY

This opportunity is available for licensing or acquisition.

#### **DEVELOPMENT STATUS**

Assay characteristics (linearity and reproducibility) were systematically evaluated with human heparinized blood samples. Clinical application has been established. Validation according to FDA M10 guidelines and clinical proof-of-principle are ongoing.

## PATENT SITUATION

Non-provisional US patent application USA (US20210087603A1) with priority of 2019 is pending.

## FURTHER READING

Brunnbauer, P. et al. (2018) "The nanomolar sensing of nicotinamide adenine dinucleotide in human plasma using a cycling assay in albumin modified simulated body fluids", Scientific Reports, volume 8.



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