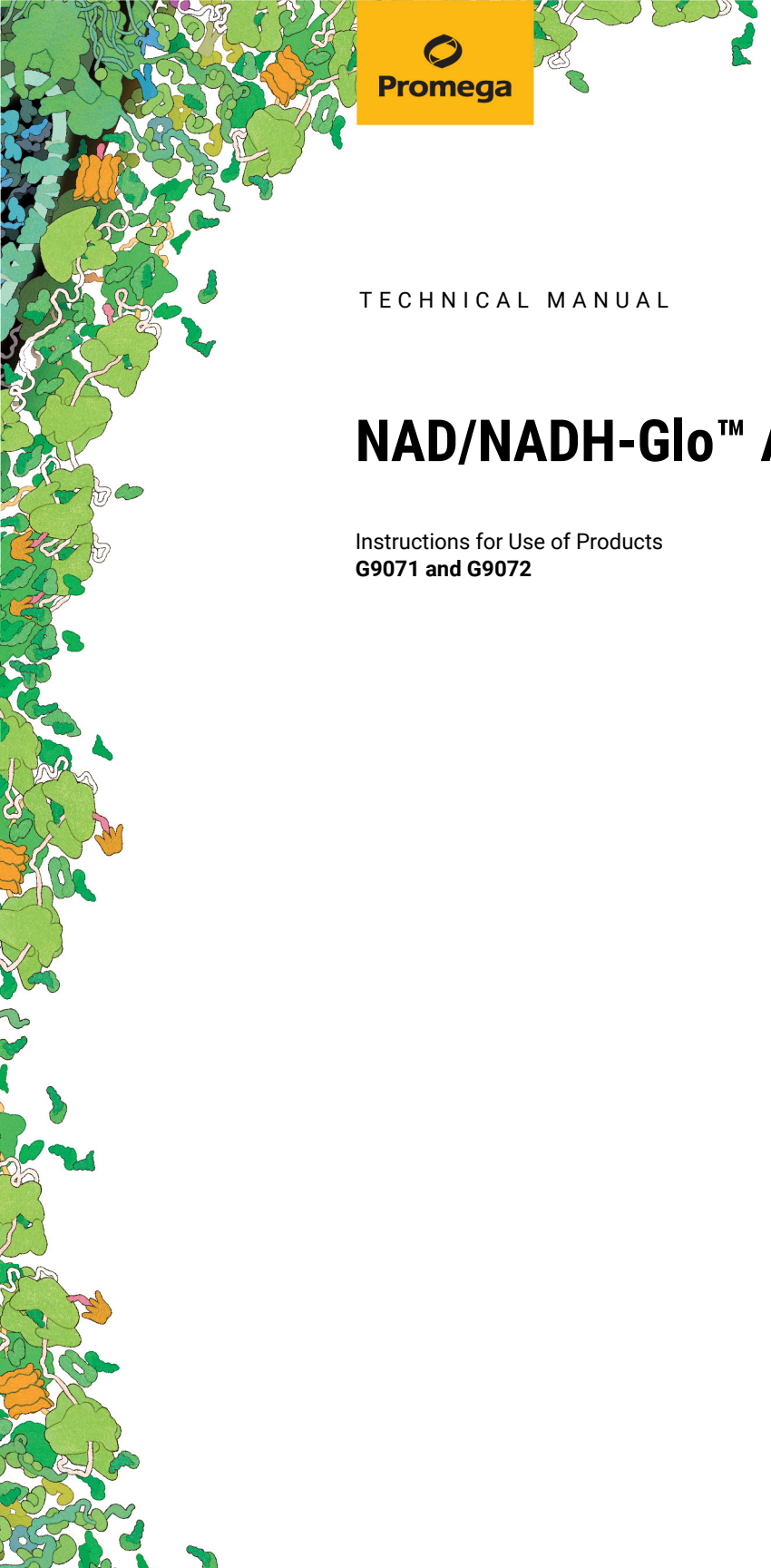

TECHNICAL MANUAL

NAD/NADH-Glo™ Assay

Instructions for Use of Products
G9071 and G9072

Revised 9/23
TM399

NAD/NADH-Glo™ Assay

All technical literature is available at: www.promega.com/protocols/
Visit the web site to verify that you are using the most current version of this Technical Manual.
E-mail Promega Technical Services if you have questions on use of this system: techserv@promega.com

1. Description

The NAD/NADH-Glo™ Assay[a] is a bioluminescent assay for detecting total oxidized and reduced nicotinamide adenine dinucleotides (NAD^+ and NADH, respectively) and determining their ratio in biological samples. NAD^+ and NADH are critical molecules important for major cellular processes including metabolism, signal transduction and epigenetics, and their levels are key indicators of cell health (1,2).

The NAD/NADH-Glo™ Assay is a homogeneous, single-reagent-addition method to rapidly detect NAD^+ and NADH in cells and enzymatic reactions and is easily adaptable for inhibitor screening in high-throughput formats.

The NAD Cycling Enzyme is used to convert NAD^+ to NADH. In the presence of NADH, the enzyme Reductase reduces a proluciferin reductase substrate to form luciferin. Luciferin then is quantified using Ultra-Glo™ Recombinant Luciferase (rLuciferase), and the light signal produced is proportional to the amount of NAD^+ and NADH in the sample (Figure 1). Cycling between NAD^+ and NADH by the NAD Cycling Enzyme and Reductase increases assay sensitivity and provides selectivity for the nonphosphorylated NAD^+ and NADH compared to the phosphorylated forms $NADP^+$ and NADPH.

1. Description (continued)

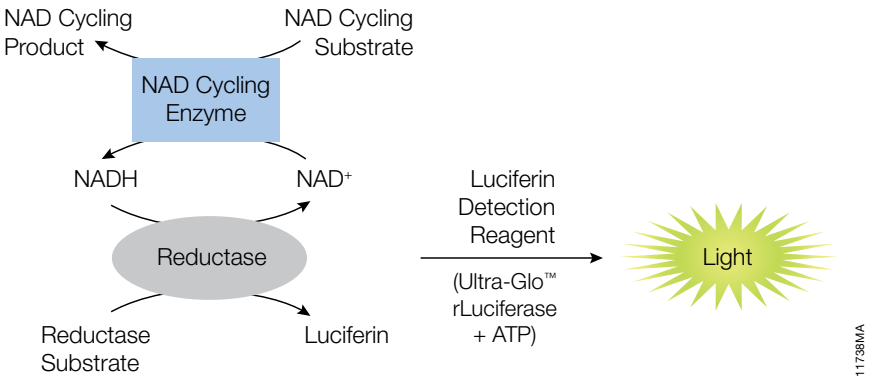

Figure 1. Schematic diagram of the NAD/NADH-Glo™ Assay technology. NAD Cycling Enzyme converts NAD+ to NADH. In the presence of NADH, Reductase enzymatically reduces a proluciferin reductase substrate to luciferin. Luciferin is detected using Ultra-Glo™ rLuciferase, and the amount of light produced is proportional to the amount of NAD^+ and NADH in a sample.

The NAD Cycling Enzyme, Reductase and luciferase reactions are initiated by adding an equal volume of NAD/NADH-Glo™ Detection Reagent, which contains NAD Cycling Enzyme and Substrate, Reductase, Reductase Substrate and Ultra-Glo™ rLuciferase, to an NAD^+- or NADH-containing sample (Figure 2). Detergent present in the reagent lyses cells, allowing detection of total cellular NAD^+ and NADH in a multiwell format with addition of a single reagent. An accessory protocol is provided to allow separate measurements of NAD^+ and NADH and calculation of the NAD^+ to NADH ratio (Section 5.A).

Due to the cycling of the coupled enzymatic reactions, the light signal will continue to increase after adding the NAD/NADH-Glo™ Detection Reagent to the sample (see Section 6). The luminescent signal remains proportional to the starting amount of NAD^+ and NADH within the linear range of the assay. The assay has a linear range of 10nM to 400nM NAD^+ and high signal-to-background ratios and is specific for the nonphosphorylated forms (Figure 3). The assay is compatible with 96-, 384-, low-volume 384- and 1536-well plates and is well suited to monitor the effects of small molecule compounds on NAD^+ and NADH levels in enzymatic reactions or directly in cells in high-throughput formats.

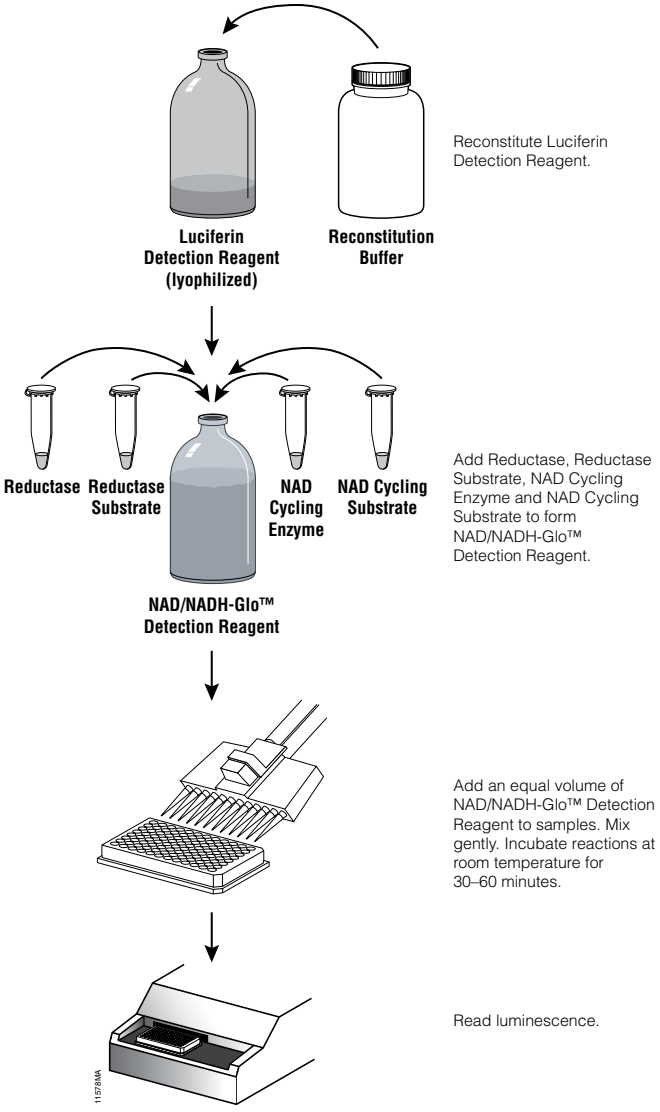

Figure 2. Schematic diagram of the NAD/NADH-Glo™ Assay protocol.

1. Description (continued)

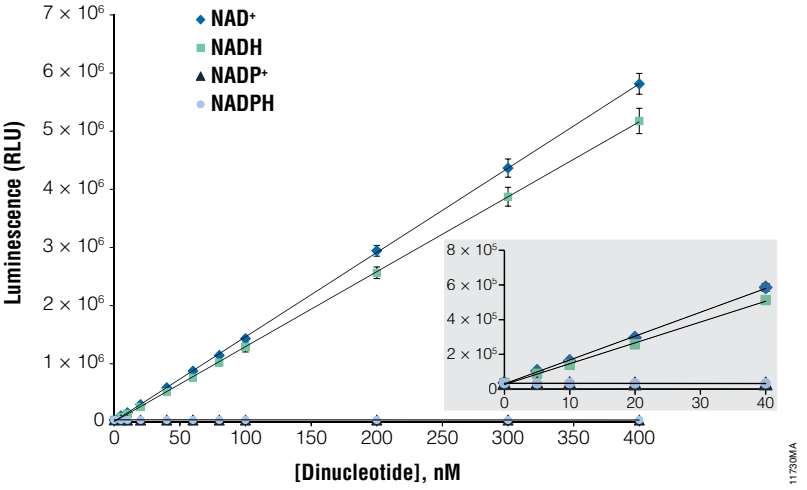

Figure 3. Linear range and specificity of the NAD/NADH-Glo™ Assay. Individual purified nicotinamide adenine dinucleotides were assayed following the protocol described in Section 3.C. NADH, NADPH, NAD^+ and $NADP^+$ stocks were prepared fresh from powder (Sigma Cat.# N6660, N9910, N8285 and N8035, respectively) and diluted to the indicated concentrations in phosphate-buffered saline (PBS). Fifty-microliter samples at each dinucleotide concentration were incubated with 50µl of NAD/NADH-Glo™ Detection Reagent in white 96-well luminometer plates. After a 30-minute incubation, luminescence was measured with a GloMax® 96 Microplate Luminometer. Each point represents average luminescence of quadruplicate reactions measured in relative light units (RLU). Error bars are ± 1 standard deviation. The limit of detection was approximately 1nM for this experiment. The data used to generate this figure are shown in Table 1.

Table 1. Titration of Purified Dinucleotides.

	NAD+		NADH	
Dinucleotide Concentration (nM)	Luminescence (RLU)	Signal-to-Background Ratio[1]	Luminescence (RLU)	Signal-to-Background Ratio[1]
400	5,813,895	163.7	5,175,684	145.1
300	4,364,286	122.9	3,872,261	108.5
200	2,942,975	82.9	2,564,713	71.9
100	1,431,256	40.3	1,288,099	36.1
80	1,141,183	32.1	1,020,158	28.6
60	872,819	24.6	758,605	21.3
40	585,845	16.5	512,531	14.4
20	295,865	8.3	257,097	7.2
10	161,209	4.5	138,984	3.9
5	104,505	2.9	86,631	2.4
0	35,513	1.0	35,681	1.0

[1]Signal of sample divided by signal of the 0nM control.

2. Product Components and Storage Conditions

PRODUCT	SIZE	CAT.#
NAD/NADH-Glo™ Assay	**10ml**	**G9071**

The system contains sufficient reagents to perform 100 reactions in 96-well plates (100µl of sample + 100µl of NAD/NADH-Glo™ Detection Reagent), 400 assays in 384-well plates (25µl of sample + 25µl of NAD/NADH-Glo™ Detection Reagent) or 2,000 assays in low-volume 384-well plates (5µl of sample + 5µl of NAD/NADH-Glo™ Detection Reagent). Assay volumes can be varied depending on plate format as long as you maintain a 1:1 ratio of sample to NAD/NADH-Glo™ Detection Reagent. Includes:

- 55µl Reductase
- 55µl Reductase Substrate
- 1 vial NAD Cycling Enzyme (lyophilized)
- 1.25ml NAD Cycling Substrate
- 1 vial Luciferin Detection Reagent (lyophilized)
- 10ml Reconstitution Buffer

PRODUCT	SIZE	CAT.#
NAD/NADH-Glo™ Assay	**50ml**	**G9072**

The system contains sufficient reagents to perform 500 reactions in 96-well plates (100µl of sample + 100µl of NAD/NADH-Glo™ Detection Reagent), 2,000 assays in 384-well plates (25µl of sample + 25µl of NAD/NADH-Glo™ Detection Reagent) or 10,000 assays in low-volume 384-well plates (5µl of sample + 5µl of NAD/NADH-Glo™ Detection Reagent). Assay volumes can be varied depending on plate format as long as you maintain a 1:1 ratio of sample to NAD/NADH-Glo™ Detection Reagent. Includes:

- 275µl Reductase
- 275µl Reductase Substrate
- 1 vial NAD Cycling Enzyme (lyophilized)
- 1.25ml NAD Cycling Substrate
- 1 vial Luciferin Detection Reagent (lyophilized)
- 50ml Reconstitution Buffer

Storage Conditions: Store all components below –65°C. Alternatively, store the Reductase Substrate below –65°C and all other components at –30°C to –10°C. Minimize freeze-thaw cycles of all reagents.

3. NAD/NADH-Glo™ Assay Protocol

3.A. Preparing the Luciferin Detection Reagent

1. Thaw the Reconstitution Buffer, and equilibrate the Reconstitution Buffer and lyophilized Luciferin Detection Reagent to room temperature.
2. Transfer the entire contents of the Reconstitution Buffer bottle to the amber bottle of lyophilized Luciferin Detection Reagent.
3. Mix by swirling or inversion to obtain a uniform solution. Do not vortex. The Luciferin Detection Reagent should go into solution easily in less than 1 minute.

   Note: Store the reconstituted Luciferin Detection Reagent at room temperature while preparing the NAD/NADH-Glo™ Detection Reagent. If the reconstituted Luciferin Detection Reagent is not used immediately, the reagent can be stored at room temperature (approximately 22°C) for up to 24 hours or dispensed into single-use aliquots and stored at 4°C for up to 1 week or –20°C for up to 3 months with no change in activity.

3.B. Preparing the NAD/NADH-Glo™ Detection Reagent

Determine the number of NAD/NADH-Glo™ Assays being performed, and calculate the volume of NAD/NADH-Glo™ Detection Reagent needed. An equal volume of NAD/NADH-Glo™ Detection Reagent will be added to each sample containing NAD^+ or NADH. We recommend preparing extra reagent to compensate for pipetting error. Do not store unused NAD/NADH-Glo™ Detection Reagent.

1. Equilibrate the reconstituted Luciferin Detection Reagent to room temperature.
2. Thaw the Reductase, Reductase Substrate and NAD Cycling Substrate at room temperature or on ice just prior to use. Briefly centrifuge the tubes to bring the contents to the bottom of the tubes, and store on ice.
3. Reconstitute the NAD Cycling Enzyme by adding 275µl of water. Mix by gently swirling the vial, and store on ice.

3.B. Preparing the NAD/NADH-Glo™ Detection Reagent (continued)

4. Prepare the required amount of NAD/NADH-Glo™ Detection Reagent by adding the volumes of Reductase, Reductase Substrate, NAD Cycling Enzyme and NAD Cycling Substrate indicated in Table 2 per 1ml of reconstituted Luciferin Detection Reagent.

For best results, we recommend preparing the NAD/NADH-Glo™ Detection Reagent immediately before use. If necessary, the prepared NAD/NADH-Glo™ Detection Reagent can be kept at room temperature and used within 6 hours.

Table 2. Preparing the NAD/NADH-Glo™ Detection Reagent.

Component	Volume
Reconstituted Luciferin Detection Reagent	1ml
Reductase	5µl
Reductase Substrate	5µl
NAD Cycling Enzyme	5µl
NAD Cycling Substrate	25µl

5. Mix by gently inverting five times.
6. Return unused Reductase, NAD Cycling Enzyme and NAD Cycling Substrate to –20°C storage. Return unused Reductase Substrate to storage at less than –65°C. Do not store unused NAD/NADH-Glo™ Detection Reagent. Minimize the number of freeze-thaw cycles for all reagents.

3.C. Protocol

Perform a titration of your particular cell line to determine the linear range and optimal number of cells to use with the NAD/NADH-Glo™ Assay (see Section 6). Include control wells without cells to determine background luminescence.

This protocol is for a reaction of 50µl of sample and 50µl of NAD/NADH-Glo™ Detection Reagent in a 96-well plate. The reaction volume can be varied as long as you maintain a 1:1 ratio of sample to NAD/NADH-Glo™ Detection Reagent. Throughout this manual, sample refers to the starting material such as tissue culture cells.

Note: Avoid the presence of DTT and other reducing agents in the samples. Reducing agents will react with the Reductase Substrate and increase background. Also avoid the presence of chelating compounds such as EDTA.

1. Plate cells in a white-walled tissue culture plate, and treat with the compounds of interest. The final volume per well should be 50µl.

2. If cells were incubated at 37°C during treatment, remove plate from the incubator, and equilibrate at room temperature for 5 minutes.

   Note: The assay is compatible with most complete media, making it unnecessary to remove the medium. The medium can be removed and replaced with 50µl of PBS per well if desired.

3. Add 50µl of NAD/NADH-Glo™ Detection Reagent to each well.

4. Gently and briefly shake the plate to mix and lyse cells.

5. Incubate for 30–60 minutes at room temperature.

   Note: The light signal will continue to increase with time. Changes in light output can be monitored over time, or luminescence can be measured at a single time point. Be sure to determine the optimal incubation time for your particular application (see Section 6).

6. Record luminescence using a luminometer.

4. General Considerations

Plates and Luminometers

Use opaque, white multiwell tissue-culture-treated sterile plates that are compatible with your luminometer (e.g., Corning® 96-well solid white flat-bottom polystyrene TC-treated microplates, Cat.# 3917, or Corning® 384-well low-flange white flat-bottom polystyrene TC-treated microplates, Cat.# 3570). For cultured cell samples, white-walled clear-bottom tissue culture plates (e.g., Corning® 96-well flat clear-bottom white polystyrene TC-treated microplates, Cat.# 3903) are acceptable. If using clear tissue culture plates, you must transfer reactions to white luminometer plates before measuring luminescence. Light signal is diminished in black plates, and increased well-to-well cross-talk is observed in clear plates. All standard instruments capable of measuring luminescence are suitable for this assay. Instrument settings depend on the luminometer manufacturer. Use an integration time of 0.25–1 second per well as a guide. Although relative light output will vary with different instruments, variation should not affect assay performance.

Temperature

The intensity and stability of the luminescent signal from the NAD/NADH-Glo™ Assay depend on the reaction rates of the Reductase, NAD Cycling Enzyme and luciferase enzyme. Environmental factors such as temperature affect reaction rates and the intensity of light output. For consistent results, equilibrate the NAD/NADH-Glo™ Detection Reagent to room temperature (approximately 22°C) before using, and equilibrate assay plates at room temperature for 5 minutes before adding the NAD/NADH-Glo™ Detection Reagent. Insufficient equilibration may result in a temperature gradient and variability across the plate.

4. General Considerations (continued)

Chemical Environment

The chemical environment of the sample containing NAD^+ or NADH (e.g., cell type, medium and buffer) can affect the Reductase, NAD Cycling Enzyme and luciferase enzymatic rates and light signal intensity. Some media contain ingredients such as pyruvate that can slow down the enzymatic rate. If necessary, increase the incubation time after adding the NAD/NADH-Glo™ Detection Reagent until sufficient sensitivity is achieved. We recommend testing your particular cell type and medium to determine the optimal cell number and incubation time for your application. The assay is compatible with phenol red.

We recommend a pH of ~7–8 for the NAD^+- and NADH-containing samples. Avoid the presence of chelating compounds such as EDTA in the samples. The luciferase reaction requires the divalent magnesium cation, which is included in the Luciferin Detection Reagent. Also, avoid the presence of DTT and other reducing agents in the samples. Reducing agents will react with the Reductase Substrate and increase background.

The NAD/NADH-Glo™ Assay is compatible with samples containing up to 10% DMSO.

5. Measuring NAD^+ or NADH Individually

5.A. Protocol for Sample Preparation

The protocol to separate oxidized (NAD^+) and reduced (NADH) forms takes advantage of the differential stabilities of the forms at acidic and basic pH. In general, oxidized forms are selectively destroyed by heating in basic solution, while reduced forms are not stable in acidic solution (3). Levels of cellular dinucleotides can be individually measured after treatment with acid or base conditions.

The following sample preparation protocol is recommended for use with the NAD/NADH-Glo™ Assay to measure NAD^+ and NADH separately (Figure 4). With this protocol, cells can be processed directly in wells of a 96-well plate. We recommend lysing cells in the preferred base solution with dodecyltrimethylammonium bromide (DTAB), which lyses cells and preserves the stability of the dinucleotides, then splitting the sample into separate wells for acid and base treatments. An advantage of this method is that NAD^+ and NADH can be measured from one cell sample with in-plate processing. The same treated samples can be used to measure $NADP^+$ and NADPH using the NADP/NADPH-Glo™ Assay (Cat.# G9081, G9082).

After sample preparation, all neutralized samples have the same final buffer formulation, which facilitates direct comparison of luminescence values. The direct correlation between luminescence and NAD⁺ or NADH amount in the samples allows calculation of the NAD⁺ to NADH ratio by dividing luminescence obtained from samples heated in acid by luminescence obtained from samples heated in base solution. Representative data are shown in Figure 5 and Table 3. A standard curve can be generated to quantitate the levels of NAD⁺ and NADH (see Section 5.B).

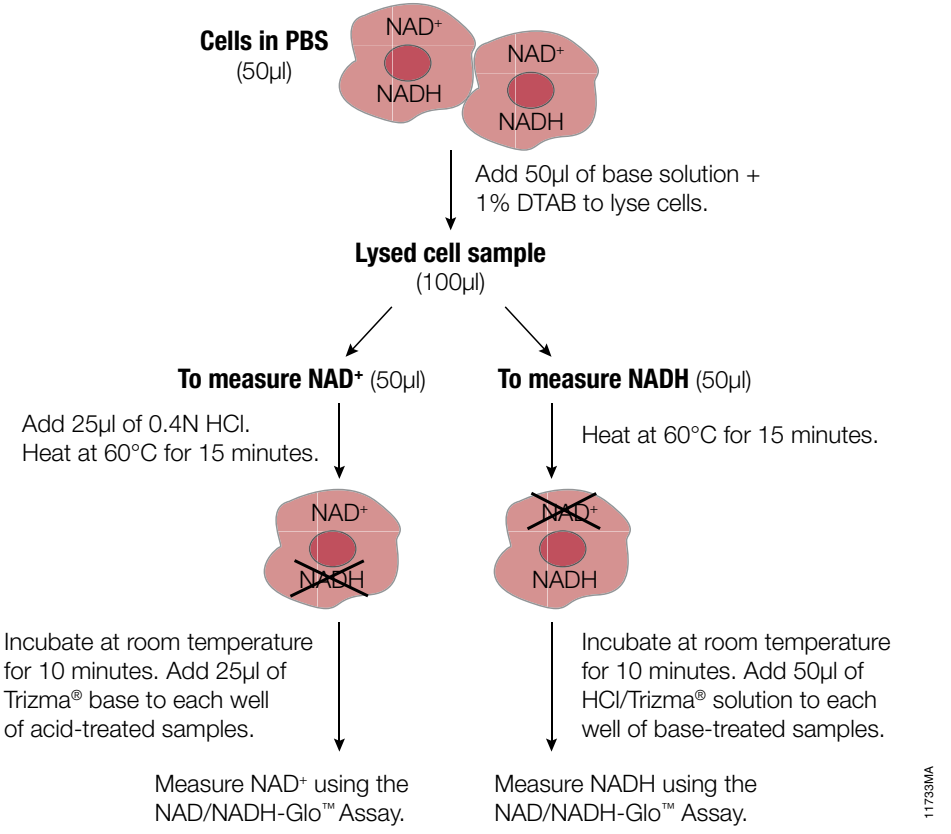

Figure 4. Schematic diagram of the sample preparation protocol for measuring NAD⁺ and NADH individually.

5.A. Protocol for Sample Preparation (continued)

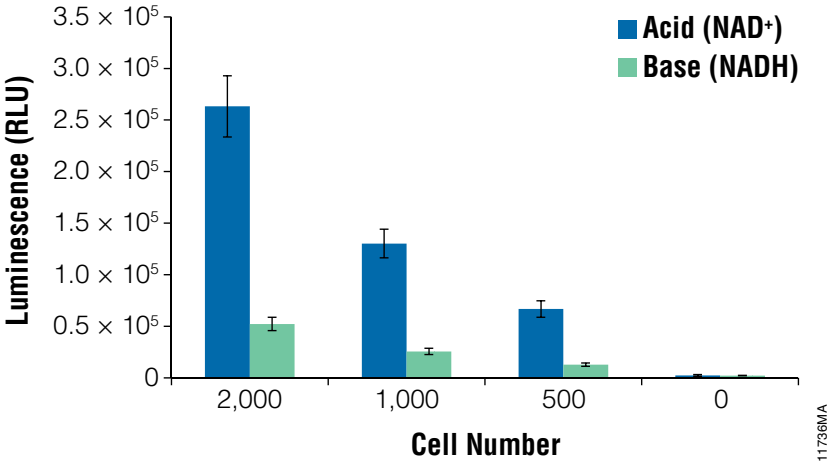

Figure 5. Separate measurement of cellular NAD⁺ and NADH from a single cell sample. K562 cells were centrifuged and resuspended in PBS at a density of 4 × 10^5 cells/ml. After twofold serial dilutions in PBS, 50µl of diluted cells was transferred to each well of a white 96-well plate. Cells were lysed by adding 50µl of bicarbonate base buffer with 1% DTAB and processed as described in Section 5.A. The plate was weighed before and after heating to quantify evaporation. A ≤2% change in weight was observed, indicating minimal sample loss due to evaporation. Twenty microliters of each neutralized sample, containing the indicated number of cell equivalents, was transferred to a 384-well plate, and the NAD/NADH-Glo™ Assay protocol was performed as described in Section 3.C. The average of quadruplicate reactions is plotted. Error bars are ± 1 standard deviation.

Table 3. Calculation of the NAD⁺ to NADH Ratio.

	Cell Number[1]			
	2,000	**1,000**	**500**	**0**
Luminescence of acid-treated samples (NAD⁺) (RLU)	263,217	130,205	66,700	2,278
Luminescence of base-treated samples (NADH) (RLU)	52,215	25,604	12,766	2,135
Ratio of NAD⁺ to NADH	5.0	5.1	5.2	

[1]Number of cell equivalents in 20µl of neutralized sample combined with 20µl of NAD/NADH-Glo™ Detection Reagent.

Materials to Be Supplied by the User

(Solution compositions are provided in Section 8.)

- phosphate-buffered saline (e.g., Sigma Cat.# D8537 or Gibco Cat.# 14190)
- base solution: bicarbonate base buffer or 0.2N NaOH
  Note: Two base solutions, the bicarbonate base buffer and 0.2N NaOH, have been tested in the protocol and perform similarly. For 100 samples, 4.8ml of either base solution is required.
- 0.4N HCl
- base solution with 1% DTAB (bicarbonate base buffer with 1% DTAB or 0.2N NaOH with 1% DTAB)
- 0.5M Trizma® base
- HCl/Trizma® solution

This protocol is for assaying cells in 50µl of PBS per well in 96-well white luminometer plates. Each well of cells is split into two samples: One sample is treated with acid to quantify NAD^+, and the other is treated with base to quantify NADH (see Figure 4). When plating cells, reserve wells on the plate for splitting samples. Alternatively, use a second plate when splitting samples.

1. Prepare the Luciferin Detection Reagent as described in Section 3.A.
2. To each well of cells in 50µl of PBS, add 50µl of base solution with 1% DTAB.
3. Briefly mix plate on a plate shaker to ensure homogeneity and cell lysis.
4. Remove 50µl of each sample to an empty well for acid treatment. To these samples, add 25µl of 0.4N HCl per well; these wells contain the acid-treated samples. The original sample wells are the base-treated samples; do not add 0.4N HCl to those wells.
5. Cover the plate, and incubate all samples for 15 minutes at 60°C.
6. Equilibrate the plate for 10 minutes at room temperature.
7. Add 25µl of 0.5M Trizma® base to each well of acid-treated cells to neutralize the acid.
8. Add 50µl of HCl/Trizma® solution to each well containing base-treated samples.
   Note: At this point, the total volume per well is 100µl. To perform the NAD/NADH-Glo™ assay, you may add 100µl of NAD/NADH-Glo™ Detection Reagent directly to each well in Step 10. Alternatively, you may remove a portion of the sample to another plate before adding an equal volume of NAD/NADH-Glo™ Detection Reagent (e.g., transfer 20µl of sample to a 384-well plate, and add 20µl of NAD/NADH-Glo™ Detection Reagent).
9. Prepare the NAD/NADH-Glo™ Detection Reagent as described in Section 3.B.
10. Add an equal volume of NAD/NADH-Glo™ Detection Reagent (e.g., 100µl) to each well.
11. Gently shake the plate to mix.
12. Incubate for 30–60 minutes at room temperature.
13. Record luminescence using a luminometer.
    Note: The oxidized form (NAD^+) is selectively destroyed by heating in basic solution, while the reduced form (NADH) is not stable in acidic solution. Thus, luminescence from acid-treated samples is proportional to the amount of NAD^+. Luminescence from base-treated samples is proportional to the amount of NADH.

5.B. Generating a Standard Curve

A standard curve allows conversion of luminescence (in RLU) to NAD^+ or NADH concentration by directly comparing luminescence from samples to the light signals generated from purified NAD^+ or NADH. For the standard curve, we recommend using purified NAD^+ to prepare a concentrated stock of 2mM NAD^+ in PBS. (If Sigma Cat.# N8285 is used, the stock solution can be prepared directly in the vial.) Immediately before the assay, prepare standard samples at the desired concentrations by diluting the 2mM NAD^+ stock in the same buffer used to prepare the experimental samples, as pH and some buffer components can affect the light signal (see Section 4). If experimental samples were generated using the sample preparation protocol in Section 5.A, dilute the NAD^+ in a mixture of equal volumes of PBS, base solution with 1% DTAB, 0.4N HCl and 0.5M Trizma® base. Assay each standard sample on the same plate as the experimental samples. Include control wells that lack NAD^+.

For each point on the standard curve, calculate average luminescence, and subtract average luminescence of the blank reactions (reactions at 0nM NAD^+) to obtain net luminescence. Use the net luminescence values to generate the standard curve and perform linear regression analysis. Interpolate the amount of NAD^+ or NADH by comparing net luminescence values of the experimental samples to the values in the standard curve.

6. Establishing the Linear Range with Cells

Luminescence is directly proportional to cell number over the linear range of the NAD/NADH-Glo™ Assay. The NAD/NADH-Glo™ Assay is compatible with many cell types and media. However, absolute light signal intensity and linear range depend on specific cell type and medium (Figure 6).

We recommend testing your particular cell type and medium to determine the linear range, optimal cell number and optimal incubation time for your application.

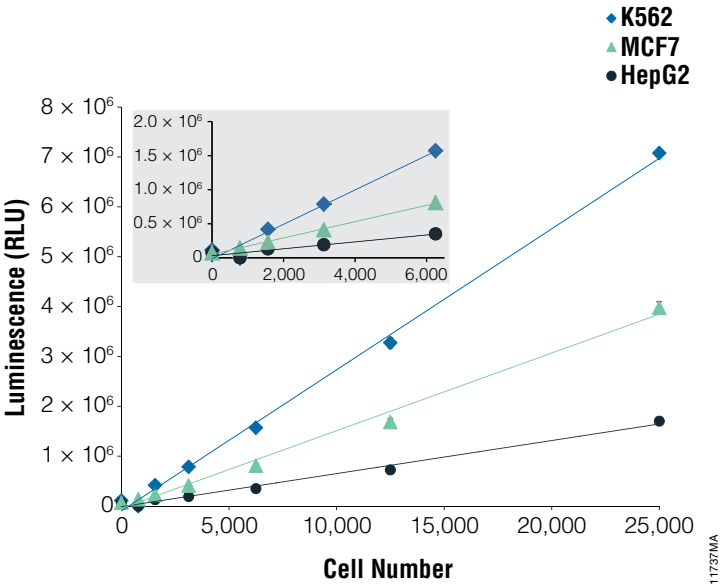

Figure 6. Linear relationship between light signal and cell density. The indicated number of cells were assayed in medium [RPMI 1640 supplemented with 10% fetal bovine serum (FBS) for K562 cells and EMEM supplemented with 10% FBS for HEPG2 and MCF7 cells] in wells of 96-well white plates. Fifty microliters of NAD/NADH-Glo™ Detection Reagent was added to 50µl of each cell type at each dilution. After a 30-minute incubation, the light signal was measured in a GloMax® 96 Microplate Luminometer. The values represent the average of quadruplicate reactions, and error bars are ± 1 standard deviation. The CV values were ≤5%.

Due to the cycling of the coupled enzymatic reactions, the light signal will continue to increase after adding the NAD/NADH-Glo™ Detection Reagent to the sample. Changes in light output can be monitored over time, or luminescence can be measured at a single time point. Optimal light signal will usually be generated within 30–60 minutes. The linear range changes with time, and at later time points, samples at higher cell numbers may be out of the linear range of the assay. Light output remains proportional to the amount of NAD^+ or NADH in the sample until all of the Reductase Substrate is converted to luciferin.

Note: If a stable light signal is preferred (for example, when batch processing multiwell plates), the increase in signal after adding the NAD/NADH-Glo™ Detection Reagent can be stopped at any time by adding the reductase inhibitor menadione. Add 10% of the reaction volume (i.e., 10µl to a 100µl reaction) of 2.75mM menadione prepared in 20% DMSO for a final concentration of 0.25mM menadione.

7. References

1. Chiarugi, A. *et al*. (2012) The NAD metabolome—a key determinant of cancer cell biology. *Nature Reviews Cancer* **12**, 741–52.
2. Houtkooper, R.H. *et al*. (2010) The secret life of NAD+: An old metabolite controlling new metabolic signaling pathways. *Endocrine Reviews* **31**, 194–223.
3. Lowry, O.H., Passonneau, J.V. and Rock, M.K. (1961) The stability of pyridine nucleotides. *J. Biol. Chem.* **236**, 2756–9.

8. Composition of Buffers and Solutions

Base solution with 1% DTAB
To one of the base solutions (i.e., bicarbonate base buffer or 0.2N NaOH), add 20% DTAB to a final concentration of 1% (v/v). For example, to 4.75ml of base solution, add 0.25ml of 20% DTAB.

Bicarbonate base buffer

100mM	sodium carbonate
20mM	sodium bicarbonate
10mM	nicotinamide
0.05%	Triton® X-100

The pH of the prepared buffer will be approximately 10–11.

20% DTAB
Prepare a 20% DTAB (Sigma Cat.# D8638) solution in water. Warm the solution in a 37°C water bath to completely solubilize the DTAB. Store at room temperature or –20°C.

0.4N HCl
Prepare 0.4N HCl from a concentrated stock solution such as 1N HCl by diluting with water. No pH adjustment is required.

HCl/Trizma® solution
Add equal volumes of 0.4N HCl and 0.5M Trizma® base. Mix by vortexing.

0.2N NaOH
Prepare 0.2N NaOH from a concentrated stock solution such as 1N NaOH by diluting with water to 0.2N. No pH adjustment is required.

0.5M Trizma® base
Dissolve 12.1g Trizma® base powder (Sigma Cat.# T1503) in 200ml of water. The final pH will be approximately 10.7. No pH adjustment is required.

9. Related Products

Energy Metabolism Assays

Product	Size	Cat #
NAD(P)H-Glo™ Assay	10ml	G9061
NADP/NADPH-Glo™ Assay	10ml	G9081
Glucose Uptake-Glo™ Assay	5ml	J1341
Lactate-Glo™ Assay	5ml	J5021
Malate-Glo™ Assay	5ml	JE9100
Pyruvate-Glo™ Assay	5ml	J4051
Glucose-Glo™ Assay	5ml	J6021
Glycogen-Glo™ Assay	5ml	J5051
Glutamine/Glutamate-Glo™ Assay	5ml	J8021
Glycerol-Glo™ Assay	5ml	J3150
Triglyceride-Glo™ Assay	5ml	J3160
Cholesterol/Cholesterol Ester-Glo™ Assay	5ml	J3190
BCAA-Glo™ Assay	5ml	JE9300
BHB-Glo™ Assay	5ml	JE9400
Metabolite-Glo™ Detection System	5ml	J9030
ROS-Glo™ H2O2 Assay	10ml	G8820
GSH/GSSG-Glo™ Assay	10ml	V6611

*Additional sizes available.

Cell Viability Assays

Product	Size	Cat #
CellTiter-Glo® 2.0 Cell Viability Assay	10ml	G9241
CellTiter-Glo® 3D Cell Viability Assay	10ml	G9681
RealTime-Glo™ MT Cell Viability Assay	100 assays	G9711
CellTiter-Fluor™ Cell Viability Assay	10ml	G6080

*Additional sizes available.

9. Related Products (continued)

Cytotoxicity and Apoptosis Assays

Product	Size*	Cat #
LDH-Glo™ Cytotoxicity Assay	10ml	J2380
CellTox™ Green Cytotoxicity Assay	10ml	G8741
Caspase-Glo® 3/7 Assay System	2.5ml	G8090
RealTime-Glo™ Annexin V Apoptosis and Necrosis Assay	100 assays	JA1011

*Additional sizes available.

10. Summary of Changes

The following changes were made to the 9/23 revision of this document:

1. Updated patent statements.
2. Changed font and cover image.
3. Made minor text edits.

[a]U.S. Pat. Nos. 9,273,343 and 9,951,372, European Pat. No. 2751089, Japanese Pat. No. 6067019 and other patents pending.