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# **Serum Creatinine Kit**

Publications



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Quantity:	2 x 96 tests
Target:	Creatinine (CR)
Reactivity:	Human, Mouse, Rat, Rabbit, Sheep, Mammalian
Application:	Biochemical Assay (BCA)

Product Details		
Purpose:	The DetectX® Serum Creatinine kits is designed to quantitatively measure creatinine present inserum samples.	
Brand:	DetectX®	
Sample Type:	Serum, Plasma	
Detection Method:	Colorimetric	
Specificity:	Species Independent. Samples Types validated: Mammalian Serum and Plasma	
Sensitivity:	0.81 μg/mL	
Characteristics:	The Serum Creatinine Kit measures creatinine in serum and plasma samples. A creatinine standard, calibrated to a NIST creatinine standard, is provided to generate a standard curve. The color generating reaction is initiated with the Creatinine Detection Reagent. The assay utilizes a kinetic absorbance method to overcome interference by colored compounds in the sample. The concentration of creatinine is calculated using the delta of the optical density readings at 1 and 30 minutes. We offer a free Excel worksheet for concentration calculations. Creatinine is a metabolite of phosphocreatine (p-creatine), a high-energy phosphate that can be utilized by tissues for the production of ATP. Creatine either comes from the diet or synthesized from the amino acids arginine, glycine, and methionine. Creatine and p-creatine are converted	

non-enzymatically to the metabolite creatinine, which diffuses into the blood and is excreted by the kidneys. In vivo, this conversion appears to be irreversible and in vitro it is favored by higher temperatures and lower pH. Measuring serum creatinine is one of the most commonly used indicators of renal function. A rise in blood creatinine levels is observed only with marked damage to functioning nephrons. A good indicator of kidney function is given by the creatinine clearance test. Creatinine clearance can be accurately calculated using serum creatinine concentration and taking into account the variables of sex, age, weight, and race.

Calibated - N-Cal Kit, NIST-Calibrated

#### Components:

Clear 96 well Plates Bag containing 2 by 96 well or 4 by 96 well Half-Area plates. 2 or 4 plates Creatinine Standard A 100 mg/dL creatinine solution in deionized water. Calibrated to NIST Standard Reference Material Lot Number 914a 100 µL or 1 mL Assay Diluent A special diluent for use in the serum kit. 6 mL or 11 mL Allow to warm completely to Room Temperature prior to use.

DetectX® Creatinine Reagent 20 mL or 50 mL

#### Material not included:

Distilled or deionized water.

Creatinine (CR)

Repeater pipet, such as an Eppendorf repeater, with disposable tips to accurately dispense 25 and  $100 \, \mu L$ .

Colorimetric 96 well microplate reader capable of reading optical density at 490 nm.

Software for converting raw relative optical density readings from the plate reader and carrying out four parameter logistic curve (4PLC) fitting.

Contact your plate reader manufacturer for de-tails.

# **Target Details**

Target:

Alternative Name:	Creatinine (CR Products)		
Target Type:	Amino Acid		
Background:	Creatinine (2-amino-1-methyl-5H-imadazol-4-one) is a metabolite of phosphocreatine (p-		
	creatine), a molecule used as a store for high-energy phosphate that can be utilized by tissues		
	for the pro- duction of ATP1. Creatine either comes from the diet or synthesized from the amino		
	acids arginine, glycine, and methionine. This occurs in the kidneys and liver, although other		
	organ systems may be involved and species-specific differences may exist2. Creatine and p-		
	creatine are converted non-enzymatically to the metabolite creatinine, which diffuses into the		
	blood and is excreted by the kidneys. In vivo, this conversion appears to be irreversible and in		
	vitro it is favored by higher temperatures and lower pH 2. Creatinine forms spontaneously from		

p-creatine3. Under normal conditions, its formation occurs at a rate that is relatively constant and as intra-individual variation is <15 % from day to day, creatinine is a useful tool for normalizing the levels of other molecules found in urine. Additionally altered creatinine levels may be associated with other conditions that result in decreased renal blood flow such as diabetes and cardiovascular disease4-6. 2- PO3 ATP ADP NH NH2 HOOC HOOC NH NH Creatine Kinase N N Creatine H Phosphocreatine O N NH N Creatinine

# **Application Details**

Application	Notes:
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This assay has been validated for human, mouse, rabbit, rat and sheep serum and EDTA and hepa- rin plasma samples.

The end user should evaluate recoveries of creatinine in other plasma and serum samples being used.

For measuring Creatinine in urine samples, please refer to our DetectX® Urinary Creatinine Detection kits, Catalog Number K002-H1 or K002-H5.

Hemolyzed or lipemic samples should not be used with this kit.

Hemolyzed samples have shown a decrease in creatinine concentration with increasing hemoglobin, whereas lipemic samples have been shown to yield artificially high creatinine concentrations.

Please see our Hemoglobin Detec- tion kit, K013-H1 for details of a convenient method to measure Hb levels in whole blood.

#### Comment:

Sample values: Eleven serum samples from a variety of different species were tested in the assay.

Values ranged from 0.78 to 1.45 mg/dL with an average of 1.00 mg/dL.

# Protocol:

A creatinine standard, calibrated to a NIST creatinine standard, is provided to generate a standard curve for the assay and all samples should be read off the standard curve.

Standards or samples are pipetted into a clear microtiter plate.

An assay diluent is added to all standards, controls and samples.

The color generating reaction is initiated with the DetectX® Creatinine Reagent, which is pipetted into each well.

The assay utilizes a kinetic absorbance method to overcome interference by colored compounds in serum.

The absorbance of the colored product is read after 1 minute in a microtiter plate reader capable of measuring 490nm wavelength.

At 30 minutes the optical density is read again.

The concentration of creatinine is calculated using the delta of the optical density readings at

# **Application Details**

30 and 1 minute compared to the curve generated from the standards, or by using the Excel worksheet available for free download at our web site. The Jaffe reaction used in this kit has been modified to read creatinine levels in serum,8. Reagent Preparation: Allow the kit reagents to come to room temperature for 30 minutes. We recommend that all standards and samples be run in duplicate to allow the end user to accurately determine creatinine concentrations. Ensure that all samples have reached room temperature and have been diluted as appropriate prior to running them in the kit. Standard Preparation Label four glass test tubes #1 through #4. Pipet 240 μL of water into tube #1 and 100 μL into tubes #2-#4. Carefully add 10 µL of the Creatinine stock solution to tube #1 and vortex completely. Take 100 µL of the creatinine solution in tube #1 and add it to tube #2 and vortex completely. Repeat these serial dilutions for tubes #3 and #4. The concentration of creatinine in tubes 1 through 4 will be 4, 2, 1 and 0.5 mg/dL. Water is used as a sample blank of 0 mg/dL. Use all Standards within 2 hours of preparation. Sample Preparation: All samples should be centrifuged for 15 minutes at 14,000 rpm in an Eppendorf type centrifuge prior to running in the assay. Assay Procedure: 1. Use the plate layout sheet on the back page to aid in proper sample and standard identification. 2 Pipet 25 µL of samples, water as the blank, or standards into wells in the clear plate. 3. Add 25 µL of Assay Diluent to all wells used. Allow to warm completely to Room Temperature prior to use. Set a timer to read 30 minutes and ensure that the plate reader is set to read optical density at 490 nm. 4. Observe wells, checking for bubbles. If bubbles are present, tap the plate gently to remove prior to addition of Reagent. 5. Add 100 µL of the DetectX® Creatinine Reagent to each well using a repeater pipet. Immediately start the timer after adding the Creatinine Reagent to the last well. 6. Incubate at room temperature. 7. At 1 minute, read the optical density generated from each well in a plate reader capable of reading at 490 nm. 8. At 30 minutes, again read the 490 nm optical density generated from each well in the plate reader. Subtract the average Optical Density of the standards at 1 minute from the average Optical Calculation of Results:

Den- sity of the standards at 30 minutes and plot the result (Average Delta OD) versus the creatinine concentration of the standards.

Generate a linear regression line and use the equation, y=mx+b (y=Average delta OD,

x=Creatinine Concentration: m=slope and b= intercept) to calculate the concentrations in the unknown samples.

Alternatively go to our website and download a sample concentration spreadsheet at:

www.ArborAssays.com/resources/lit.asp Or use the online tool from

www.myassays.com/arbor-assays-creatinine-serum-kit.assay to calcu- late the data. \*The

 $\label{thm:myAssays} \mbox{ logo is a registered trademark of MyAssays Ltd.}$ 

Restrictions:

For Research Use only

# Handling

Precaution of Use:

As with all such products, this kit should only be used by qualified personnel who have had

labo- ratory safety instruction.

The complete insert should be read and understood before attempting to use the product.

The Creatinine Reagent contains hazardous chemicals.

It contains a solution of basic picric acid in a stabilizing solution.

The solution should not come in contact with skin or eyes.

Picric acid is an irritant and, if dried, potentially explosive.

Avoid contact with metals and use large volumes of water during disposal.

Take appropriate precautions when handling these reagents.

Storage:

4 °C,RT

Storage Comment:

All components of this kit should be stored at 4°C until the expiration date of the kit.

### **Publications**

Product cited in:

Zou, Kwon, Jiang, Ferguson, Puranik, Zhu, Lerman: "Renal scattered tubular-like cells confer protective effects in the stenotic murine kidney mediated by release of extracellular vesicles." in: **Scientific reports**, Vol. 8, Issue 1, pp. 1263, (2018) (PubMed).

Small, Sanchez, Roy, Morais, Brooks, Coombes, Johnson, Gobe: "N-acetyl cysteine increases cellular dysfunction in progressive chronic kidney damage after acute kidney injury by dampening endogenous antioxidant responses." in: **American journal of physiology. Renal physiology**, (2018) (PubMed).

Uddin, Pak, Ha: "Carbon monoxide releasing molecule-2 protects mice against acute kidney injury through inhibition of ER stress." in: **The Korean journal of physiology & pharmacology: official journal of the Korean Physiological Society and the Korean Society of Pharmacology**, Vol. 22, Issue 5, pp. 567-575, (2018) (PubMed).

Call, Donet, Martin, Sharma, Chen, Zhang, Cai, Galarreta, Okutsu, Du, Lira, Zhang, Mehrad, Annex, Klibanov, Bowler, Laubach, Peirce, Yan: "Muscle-derived extracellular superoxide dismutase inhibits endothelial activation and protects against multiple organ dysfunction syndrome in mice." in: **Free radical biology & medicine**, Vol. 113, pp. 212-223, (2017) (PubMed).

Hong, Bae, Ahn, Kim, Kwon, Jung, Ko: "Resveratrol Ameliorates Contrast Induced Nephropathy Through the Activation of SIRT1-PGC-1α-Foxo1 Signaling in Mice." in: **Kidney & blood pressure research**, Vol. 42, Issue 4, pp. 641-653, (2017) (PubMed).

There are more publications referencing this product on: Product page

## **Images**

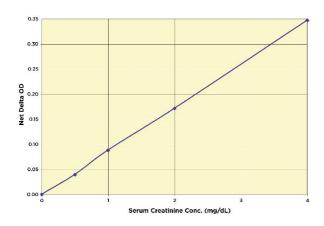


Image 1.