

Read the package insert carefully before using the product

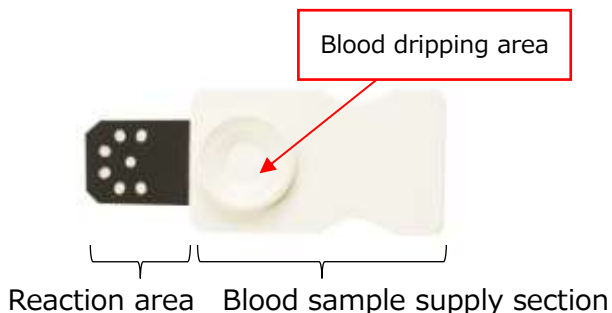
BoviLab Liver Fxn Slide 2 (AST, ALB, GLU, GGT, TC, TP)
for BoviLab Blood analyzer

[General precautions]

1. For Research use only. Not for human use.
2. This product is a reagent used for bovine blood component analysis. Do not use it for any other purpose.
3. Comprehensively judge the condition based on the analytical finding of this product as well as the results and symptoms of other related analysis.
4. We do not guarantee any method other than that described in the package insert, or its usage for purposes other than the intended one.
5. This product is a dedicated reagent for BoviLab Blood analyzer. Read the Quick Start Guide and the User Guide carefully before using the device.

[Shape, structure etc. (kit configuration)]

This product is a slide type and consists of a blood sample supply section and a reaction section.



(Components involved in the reaction system)

BoviLab AST	· Sodium L (+)-aspartate monohydrate
BoviLab ALB	· Bromocresol green
BoviLab GLU	· Glucose dehydrogenase
BoviLab GGT	· L-γ-glutamyl-3-carboxy-4-nitroanilide
BoviLab TC	· Cholesterol dehydrogenase
BoviLab TP	· Copper (II) sulfate

[Intended use]

Quantitatively measure the concentration of the

following components in bovine whole blood, serum and plasma.

- AST (aspartate aminotransferase)
- ALB (albumin)
- GLU (glucose)
- GGT (γ-glutamyltranspeptidase)
- TC (total cholesterol)
- TP (total protein)

[Measurement principle]

Whole blood dropped into the blood dripping area is pressurized by the device and passes through the blood cell separation membrane to separate the plasma. When plasma or serum is dropped directly, it passes through the blood cell separation membrane as it is. Plasma or serum moves to the reaction area and reacts by dissolving the applied and dried reagent. The coloration generated by the reaction is optically measured according to the measurement principle of each item. The measurement principle of each item in this product is as follows.

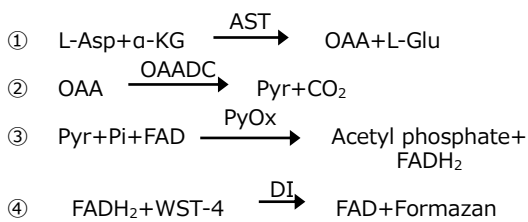
● **BoviLab AST**

It is measured by an enzymatic method using L-aspartic acid (L-Asp) and α-ketoglutaric acid (α-KG) as substrates.

- ① AST present in the blood sample produces oxaloacetic acid (OAA) and L-glutamic acid (L-Glu) from L-Asp and α-KG
- ② Oxaloacetate decarboxylase (OAADC) decomposes the resulting OAA to produce pyruvic acid (Pyr)
- ③ Pyruvate oxidase (PyOx) produces FADH₂ from the resulting Pyr, the phosphoric acid (Pi) and flavin adenine dinucleotide (FAD)
- ④ The resulting FADH₂ acts on the reductive dye WST-4 in the presence of Diaphorase (DI) to produce the green formazan

The AST concentration in the blood sample is

determined by measuring the coloration of the resulting formazan at a wavelength of 630 nm.



● BoviLab ALB

It is measured based on the bromocresol green (BCG) method.

① Albumin in the blood sample binds to BCG around pH4 to form a blue complex

The albumin concentration in the blood sample is determined by measuring the resulting complex at a wavelength of 630 nm.

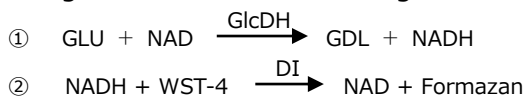


● BoviLab GLU

It is measured by the glucose dehydrogenase (GlcDH) method.

① Glucose (GLU) in the blood sample reacts with nicotinamide adenine dinucleotide (NAD) by GlcDH to produce D-glucono- δ -lactone (GDL) and NADH

② The resulting NADH acts on the reductive dye WST-4 in the presence of Diaphorase (DI) to produce green formazan
The GLU concentration in the blood sample is determined by measuring the coloration of the resulting formazan at a wavelength of 630 nm.



● BoviLab GGT

It is measured by the L- γ -glutamyl-3-carboxy-4-nitroanilide (GluCANA) method.

① GGT present in the blood sample produces L- γ -glutamylglycylglycine (L- γ -glutamylGly-Gly) and 5-amino-2-nitrobenzoic acid (5-ANB) from GluCANA and glycylglycine (Gly-Gly)

The GGT concentration in the blood sample is determined by measuring the coloration of the 5-ANB at a wavelength of 405 nm.



● BoviLab TC

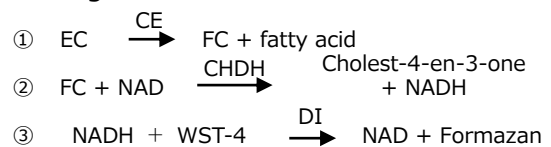
It is measured by the enzymatic method.

① Esterified cholesterol (EC) in the blood sample is hydrolyzed using cholesterol esterase (CE) and converted to free cholesterol (FC)

② Cholesterol Dehydrogenase (CHDH) produces cholest-4-en-3-one and reduced form of nicotinamide adenine dinucleotide (NADH) from the converted FC as well as the FC and oxidized form of nicotinamide adenine dinucleotide (NAD) originally contained in the blood sample

③ The resulting NADH acts on the reductive dye WST-4 in the presence of Diaphorase (DI) to produce green formazan

The total cholesterol concentration in the blood sample is determined by measuring the coloration of the resulting formazan at a wavelength of 630 nm.

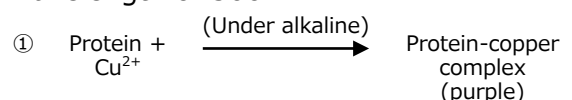


● BoviLab TP

It is measured by the biuret method.

① The protein in the blood sample reacts with the copper ion (Cu^{2+}) generated by the dissolution of copper sulfate to form a purple protein-copper complex

The TP concentration in the blood sample is determined by measuring the amount of increase in coloration of the resulting complex at a wavelength of 560 nm.



[Operational precaution]

1. Blood samples

- (1) Measure immediately after blood collection.
- (2) Heparin can be used as an anticoagulant. Do not use EDTA, citric acid, oxalic acid and sodium fluoride.
- (3) If the measured value exceeds the upper limit of the measurement range, dilute it twice with saline solution and measure again. When diluted and measured, an error may occur, so treat it as estimation.
- (4) When using a blood sample that has been refrigerated or frozen, bring it to room temperature, invert carefully and mix it thoroughly before measuring.

2. Substances, drugs, etc. that affect the measurement results

● BoviLab ALB

- Measurements of hemolyzed blood sample or blood sample containing high levels of acute phase protein may produce positive error.

● BoviLab GLU

- If whole blood is left as it is, the blood glucose level will drop due to the glycolytic reaction of red blood cells. Measure immediately after blood collection. If measurement cannot be performed immediately, separate the blood into plasma or serum as soon as possible and store refrigerated.

● BoviLab TC

- If the blood sample is left at room temperature, the cholesterol level will change over time due to the action of lecithin-cholesterol acyltransferase in the blood, so store it in a refrigerator. In case of long-term storage of serum or plasma, preservation by freezing is preferable.

● BoviLab TP

Since fibrinogen is removed from the serum sample, it may have a negative error of about 0.2 to 0.4 g/dL.

[Operation method]

1. Reagent preparation and handling

Remove this product from the refrigerator, bring it to room temperature, and then open the aluminum pouch. Once taken out of the aluminum pouch, use the product as it is. In

addition, open the aluminum pouch just before using the product.

2. Necessary apparatus/tools and materials/blood samples etc BoviLab Blood analyzer

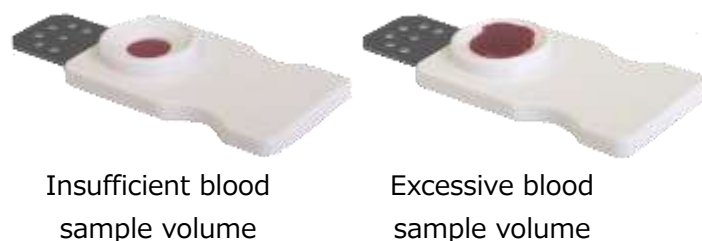
3. Measurement (operation) method

Drop approximately 150 μ L of whole blood or plasma/serum onto the blood dripping area of the product, and immediately set it in the BoviLab Blood analyzer. The device reads the QR code affixed to the back of the product, automatically measures the blood sample, and displays the result. (For details on how to operate the BoviLab Blood analyzer, see the Quick Start Guide as well as the User Guide of the device.)

Appropriate blood sample dropping volume



Inappropriate blood sample dropping amount



[Reference values]

Refer to the following website for reference range of each item in bovine blood.

<http://ib-holdings.com/en/bovilab-support>

[Clinical significance]

The metabolic profile test using blood test is used as a method to judge the health and nutritional status of the cattle.^{1), 2)} Each of the specialized reagents in the BoviLab Blood analyzer is a reagent for analyzing blood components in dairy cattle and beef cattle and

can be used for metabolic profile testing. Six tests can be performed simultaneously with one reagent by using whole blood, plasma or serum. The BoviLab Blood analyzer can be used at farms and veterinary clinics to obtain analysis results.

[Performance]

Repeatability

The reproducibility range, when low-concentration and high-concentration controlled blood samples were measured 16 times simultaneously for 3 lots of each item, is as follows.

Items	Blood sample Concentration	C.V.(%)
AST	46 U/L	2.4-4.1%
	248 U/L	1.7-2.9%
ALB	3.28 g/dL	1.6-2.7%
	5.32 g/dL	0.7-3.0%
GLU	55 mg/dL	1.2-3.0%
	339 mg/dL	0.7-2.6%
GGT	31 U/L	4.6-8.9%
	139 U/L	1.6-3.6%
TC	116 mg/dL	1.5-2.5%
	316 mg/dL	1.3-3.3%
TP	4.16 g/dL	1.9-2.8%
	6.50 g/dL	1.4-2.4%

Correlation

The correlation when measuring bovine plasma blood samples is as follows. Comparative method (X) is an automated analyzer.

Items	r	Regression equation	Concentration range
AST	0.995	y=0.969x+2.94	29-348 U/L
ALB	0.949	y=0.921x+0.31	2.64-4.61 g/dL
GLU	0.999	y=0.999x-0.01	25-336 mg/dL
GGT	0.998	y=1.053x-1.91	8-332 U/L
TC	0.999	y=0.996x+0.79	37-338 mg/dL
TP	0.970	y=1.002x-0.01	5.90-10.66 g/dL

Measurement range

The measurement range of each item is as follows.

Items	Measurement range
AST	10-1000 U/L

ALB	0.50-7.00 g/dL
GLU	10-400 mg/dL
GGT	10-600 U/L
TC	10-600 mg/dL
TP	1.00-11.00 g/dL

Interference of coexisting substances

No significant effects were observed up to the following concentrations for each of the substances listed below.

Items	Concentration	Hemoglobin (Hemolysis)	Bilirubin (Icterus)
AST	78 U/L	100 mg/dL	4 mg/dL
	300 U/L	500 mg/dL	4 mg/dL
ALB	3.10 g/dL	500 mg/dL	4 mg/dL
	4.95 g/dL	500 mg/dL	4 mg/dL
GLU	52 mg/dL	400 mg/dL	4 mg/dL
	137 mg/dL	500 mg/dL	4 mg/dL
GGT	18 U/L	significant interference	1 mg/dL
	99 U/L	500 mg/dL	4 mg/dL
TC	84 mg/dL	350 mg/dL	4 mg/dL
	263 mg/dL	500 mg/dL	4 mg/dL
TP	2.53 g/dL	significant interference	4 mg/dL
	7.35 g/dL	100 mg/dL	4 mg/dL

There were no significant effects on Chyle (Turbidity) and ascorbic acid up to the following concentrations.

Chyle (Turbidity): 1000 formazin turbidity unit
Ascorbic acid: 2.0 mg/dL

[Information related to reference material on calibration]

The reference material applies to our company's basic rules, and cannot be used directly on this product.

Items	Reference material on calibration
AST	JCCLS certificated reference material Reference Standard-JSCC Enzyme
ALB	IRMM ERM-DA470
GLU	ReCCS Certified Reference Material for Measurement of Nitrogen-containing substance, Glucose (JCCRM 521)
GGT	JCCLS certificated reference material Reference Standard-JSCC Enzyme
TC	ReCCS Total Cholesterol/Glyceride Actual Sample Primary Standard Material (JCCRM 211)

JCCLS: Japanese Committee for Clinical Laboratory Standards

IRMM: Institute for Reference Materials and Measurements

ReCCS : Reference Material Institute for Clinical Chemistry Standards

NIST: National Institute of Standards and Technology

[Precautions for use and handling]

1. Precautions for blood sample handling (hazard prevention)

- (1) Bovine blood samples are susceptible to Crimean-Congo hemorrhagic fever, Rift Valley fever, brucellosis, anthrax, etc. In order to avoid the risk of infection, wear disposable gloves when handling blood samples. Also, do not pipette by mouth.
- (2) Just like the blood sample, considering the risk of infection, handle the equipment carefully that comes in contact with the blood sample.
- (3) If the blood sample is scattered or adhered to any surface, wipe it off and disinfect the surface thoroughly with a disinfectant such as Hypochlorous acid (effective chlorine concentration 1,000 ppm, 0.1%) or 80% ethanol.

2. Precautions to be taken when using slides

- (1) BoviLab Slides that have been brought back to room temperature should be stored at room temperature and used within 3 days. They cannot be returned to refrigeration and stored again.
- (2) Do not freeze this product. Do not use if stored in the freezer, as it may not give correct results.
- (3) Do not use this product after the expiration date.
- (4) Use a new product for each measurement. Do not reuse the used reagent slide.
- (5) If the aluminum pouch is damaged, do not use it. Moreover, when opening the outer box, take care not to damage the aluminum pouch

present inside.

- (6) Aluminum pouch contains this product and desiccant. Do not use this product if it does not contain a desiccant.
- (7) Do not directly touch the blood sample supply section (blood dripping area) and the reaction section of the product.
- (8) Do not stain the label (QR code) on the back of this product.
- (9) Use following method to disinfect used products and equipments that came into contact with blood samples.
 - Disinfect with Hypochlorous acid (effective chlorine concentration 1,000 ppm or more, and soaked for 1 hour or more)
 - Disinfection treatment using glutaraldehyde (2%, soaked for an hour or more)
 - Autoclave sterilization (121°C(249.8°F), for 20 minutes or more)
- (10) Do not reuse this product or its packaging for any other purpose.

3. Precautions when disposing the slides

- (1) When disposing of this product, consider the risk of infection and dispose of the product by incineration, melting, sterilization, disinfection, etc. according to local rules and regulations and at the responsibility of each facility.

[Storage method/validity period]

1. Storage method

Store in a refrigerator at (2-8°C(35.6-46.4°F)).

2. Validity period

6 months to 1 year from the date of manufacture (depending on the item configuration)

The expiration date is stated on the outer box as well as on the individual packaging.

[Product Specifications]

Size of

the aluminum pouch: W 60 mm × L 90 mm

the slide: W 20 mm × L 50 mm

Weight: Slide main unit: 2.3 g

Material: ABS resin/polyethylene terephthalate

(PET)/glass fiber/cellulose acetate film/
polycarbonate film
Made in Japan

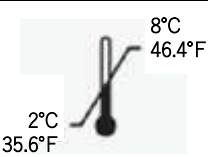
[Package unit]

15 slides/box

[References]

- 1) Keiji Okada. *Juunaikagaku the 2nd edition Daidobutuhon* : Chapter17, Seisann juuiryo sisutemu(生産獣医療システム/PRODUCTION MEDICINE SYSTEM), 2014, Bun-Eido Publishing Co.,Ltd., p.359-369
ISBN:978-4-8300-3252-3
- 2) Fukumori R, Taguchi T, Oetzel GR, Oikawa S. *Res Vet Sci.* 2021;135:247-252

[Explanation of symbols]

Symbols	Title
	Store refrigerated (2-8°C) (35.6-46.4°F)

[Contact information/Manufacturer]



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