B2M

Human beta-2-microglobulin (B2M) is an 11.6 kDa non-glycosylated protein of 99 amino acids, which is the light chain of human leucocyte antigen-I (HLA-I) and can accumulate to cause serious dialysis-related amyloidosis (DRA) in long-term hemodialysis patients. It can be used for specific elimination of B2M from serum and can induce apoptosis of several types of tumor cells, and thus has great therapeutic potential. In addition to its physiological function, much clinical interest has been drawn to B2M as its increased serum levels and misfolding have been linked to a pathological condition.

Anti-beta-2-microglobulin (B2M) monoclonal antibody

A new generation of anti-B2M monoclonal antibodies, which was recently produced by CUSAg, makes possible the development of immunoturbidimetric assay (LETIA) and lateral-flow immunoassay (LFIA). Anti-B2M monoclonal antibodies by immunoturbidimetric assay can be used for a full-range B2M assay, which is a simple, rapid, sensitive and homogeneous assay that can be performed using automated systems for high-throughput immunoassays. The sensitivity and specificity of anti-B2M monoclonal antibodies have been repeatedly tested by gold lateral-flow immunoassay (LFIA).

Properties	**Specification**
Target species	Human
Host animal	Mice Balb/c
Cell line used for fusion	Sp2/0
Immunogen	Human beta-2-microglobulin (B2M) protein
Purification method, Purity	Protein G affinity chromatography, >90%
Presentation	MAb solution in NaCl with 15 mM NaN_3 (pH 7.2) or PBS with 15 mM NaN_3 (pH 7.4)
Application	ELISA, CLIA, LEITA, LFIA etc.
Catalog Number	CSB-DA003CmN①
	CSB-DA003CmN②
	CSB-DA003CmN③

CSB-DA003CmN④

CSB-DA003CmN⑤

CSB-DA003CmN⑥

CSB-DA003CmN⑦

Anti-human B2M antibodies were evaluated on tow platforms which are immunoturbidimetric assay (LETIA) and lateral-flow immunoassay (LFIA).

Latex enhanced turbidimetric immunoassay (LETIA)

Calibration curve

The human B2M reacts with the anti-human B2M antibody-coated latex, resulting in agglutination and increase in turbidity. Turbidity changes are then measured using a spectrometer to quantitatively measure the B2M concentration in the sample. Fig.1 shows that the absorbance was increasing gradually along with the B2M concentration.

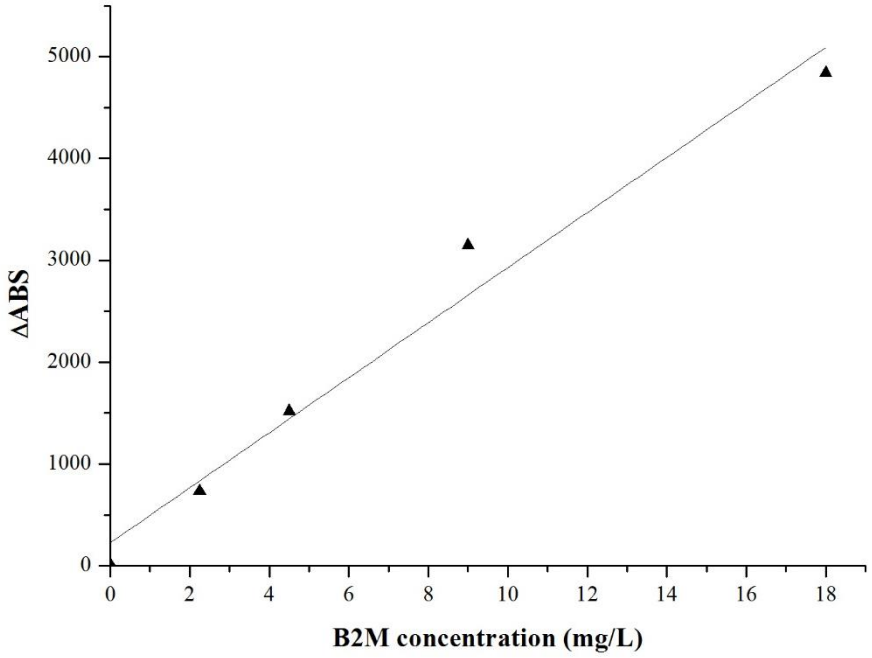

Fig.1 Linear calibration for B2M LETIA assay

Thermal Stability

Our anti-B2M monoclonal antibodies (CSB-DA003CmN③ and CSB-DA003CmN⑦) were respectively stored at 4℃ and 37℃ for 14 days, which were prepared into our CUSAg in-house B2M reagent to test the B2M control with interval of 7 days. Fig.2 shows our B2M antibodies were themal stable.

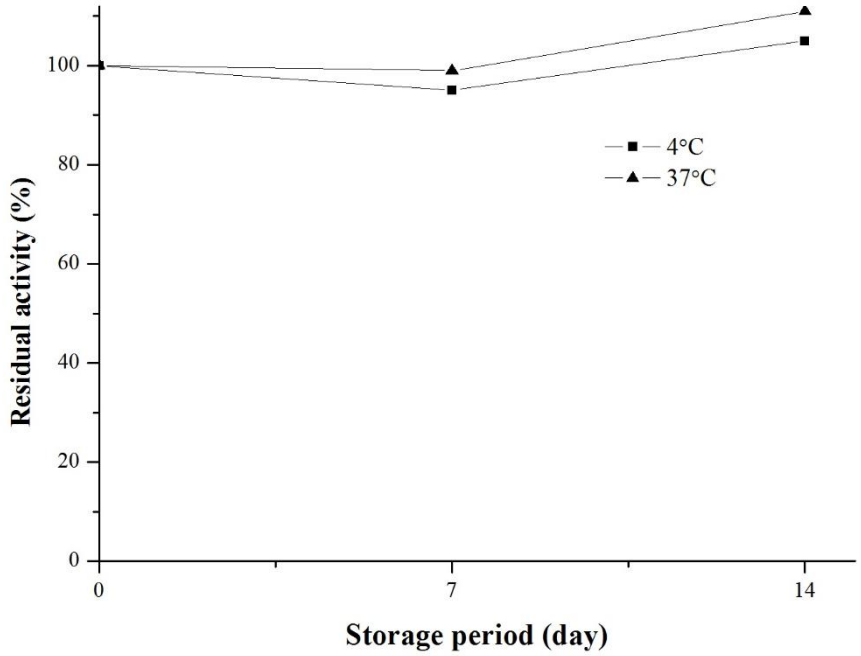

Fig.2 Thermal stability of anti-B2M monoclonal antibodies

Precision

Five members of serum samples based panel were assayed, using a single lot of reagents with our antibodies (CSB-DA003AmN③ and CSB-DA003AmN⑦), in replicates of ten on the CUSAg immunoturbidimetric assay. As shown in table.1, the system shows excellent precision with CV≤5%.

Table.1 Precision profile of B2M reagent

Panel Member	n	Mean Conc. (mg/L)	SD	%CV
1	10	0.54	0.04	4.24
2	10	0.70	0.02	0.27
3	10	0.92	0.02	1.95
4	10	1.59	0.01	0.83
5	10	2.30	0.09	4.09

Clinical Comparison

Anti-B2M monoclonal antibodies (CSB-DA003CmN③ and CSB-DA003CmN⑦) were also evaluated in medium-scale clinical trials with random blood samples from donations (n=44). Fig.3 shows that the correlation coefficient (r) is as high as 0.98 between in-house B2M assay and commercial immunoturbidimetric assay. These results show good agreement between the two systems.

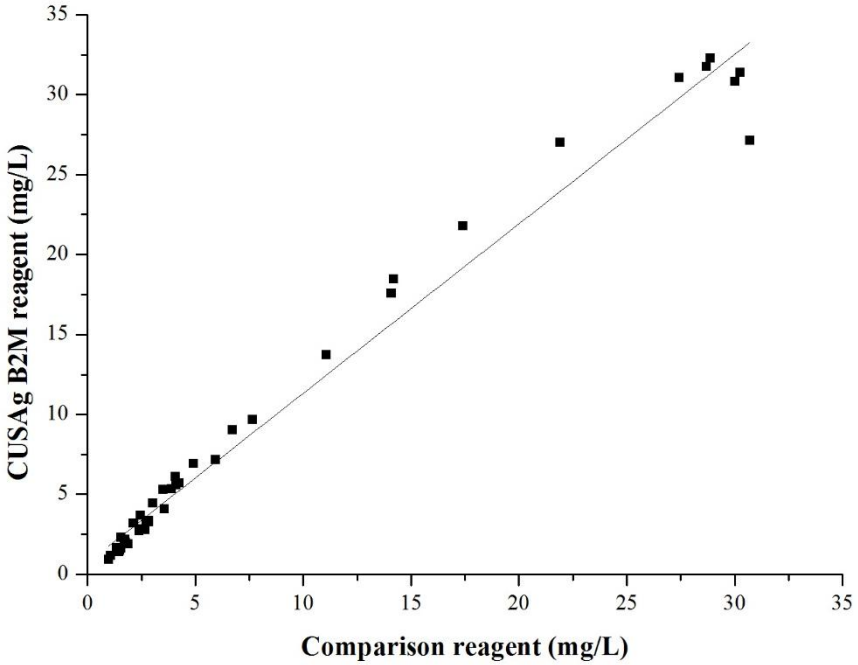

Fig.3 Clinical comparison of in-house B2M reagent and commercial kit

Lateral-flow immunoassay (LFIA)

Clinical analysis

At an in-house laboratory, the CUSAg B2M LFIA assay using CSB-DA003CmN⑤(labeled with gold)and CSB-DA003CmN⑥(capture antibody) was compared to commercial B2M LETIA kit with 6 blood samples, which were diluted by 50 fold. Fig.4 shows that there was 100% agreement between the results obtained from commercial LETIA kit and CUSAg LFIA test.

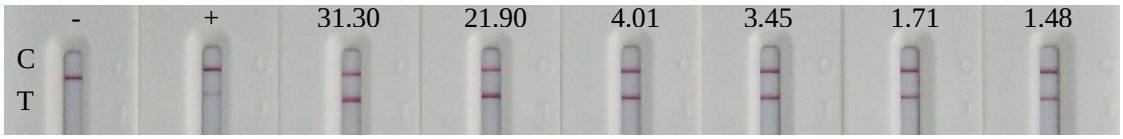

Fig.4 Clinical comparison of in-house B2M reagent and commercial LETIA kit

("+": general positive, "-": negative, LETIA: mg/L, "C": control, "T": test)

Thermal Stability

CUSAg anti-B2M antibodies (CSB-DA003AmN⑤ and CSB-DA003AmN⑥) were stored at -20 ℃, 4 ℃ and 37 ℃ for 14 days. After then, these samples were detected, respectively (Table.2). The results show that the stability of CUSAg anti-B2M antibody is perfect.

Table.2 Thermal stability of CUSAg anti-B2M monoclonal antibodies

Measurement of LETIA (mg/L)	-20 ℃	4 ℃	37 ℃

		(14 days)	(14 days)
31.30	+++	+++	+++
21.90	+++	+++	+++
4.01	++	++	++
3.45	++	++	++
1.71	+	+	+
1.48	+	+	+
Positive Control	+	+	+
Negative Control	-	-	-

“+++”: intense positive, “++”: positive, “+”: general positive, “-”: negative

References

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3. Jae Ryung Shin, Seung Min Kim, Jung Sun Yoo, Ji Yoon Park, Seul Ki Kim, Joo Hee Cho, Kyung Hwan Jeong, Tae Won Lee and Chun Gyoo Ihm: Urinary excretion of beta2-microglobulin as a prognostic marker in immunoglobulin A in nephropathy. 2014; 29; 334-340
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