Lipoprotein-associated phospholipase A2 (Lp-PLA2) also known as platelet-activating factor acetylhydrolase (PAF-AH) is a phospholipase A2 enzyme in humans. Lp-PLA2 is a 45 kDa protein of 441 amino acids encoded by the *PLA2G7* gene.

Some recent studies have shown that Lp-PLA2 is an independent risk marker for cardiovascular disease (CVD), including coronary heart disease (CHD), and ischemic stroke. In these studies, increased concentrations of Lp-PLA2 were seen in many people who were diagnosed with CHD and ischemic stroke, regardless of other risk factors. These findings make relatively the new test potentially useful among the growing number of cardiac risk markers used to determine a person's risk of developing CVD.

Lp-PLA2 is specific for vascular inflammation and is a circulating measure of the progression of rupture-prone plaque. Following production by inflammatory cells, this enzyme cleaves oxidized phospholipids, generating pro-inflammatory molecules and oxidized fatty acids. Elevated Lp-PLA2 levels can predict the development of coronary artery disease in apparently healthy individuals and the risk of future adverse cardiac and cerebrovascular events. The Lp-PLA2 tests are meaningful for assessing risk of coronary artery disease and stroke.

Anti-Human Lp-PLA2 Monoclonal Antibodies

Four highly selective anti-human Lp-PLA2 monoclonal antibodies were recently developed by CUSAg, which make possible the development of highly sensitive and rapid sandwich immunoassays. Our in-house assays have a linear detection range from 0.13 to 1000 ng/mL. All recommended MAb combinations were evaluated in medium-scale clinical trials with blood samples from normal and patients with atherosclerosis or cerebral infarction.

Properties	Specification
Target species	Human
Host animal	Mice Balb/c
Cell line used for fusion	Sp2/0
Immunogen	Human Lp-PLA2
Purification method	Protein G affinity chromatography
Presentation	MAb solution in PBS(pH 7.4)
Application	CLIA,LETIA,LFIA,ELISA and others
Catalog Number	CSB-DA113BmN① CSB-DA113BmN③ CSB-DA113BmN④ CSB-DA113BmN⑤

❶ Calibration Curve

All MAbs were tested in pairs as capture and detection antibodies to select the best two-site MAb combinations for the development of a quantitative sandwich immunoassay. Calibration curves for several best two-site combinations, which utilized different anti Lp-PLA2 MAbs, are shown in Fig.1. Detection antibodies were labeled with horse reddish peroxidase (HRP). The best selected MAb combinations for the development of quantitative human Lp-PLA2 immunoassays are (capture-detection respectively):

MAb combination A: CSB-DA113BmN①- CSB-DA113BmN③
MAb combination C: CSB-DA113BmN③- CSB-DA113BmN①
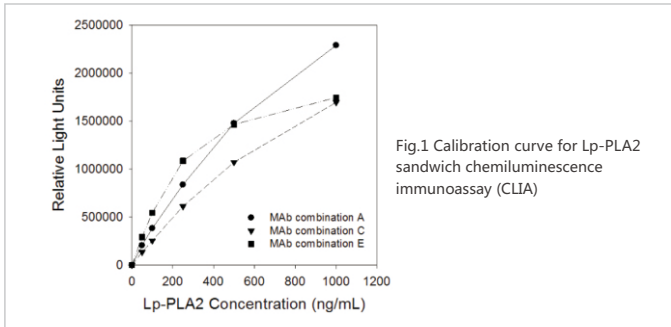MAb combination E: CSB-DA113BmN④- CSB-DA113BmN⑤

Fig.1 Calibration curve for Lp-PLA2 sandwich chemiluminescence immunoassay (CLIA)

compared to commercially available diagnostic kits. As shown in Fig.2, these results reveal our home-grown MAb combinations can be applied on double-MAb-sandwich-immunoassays.
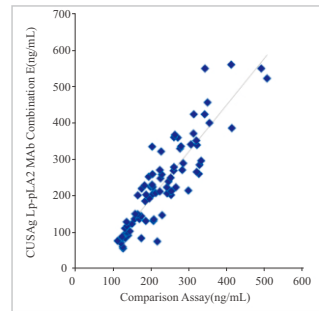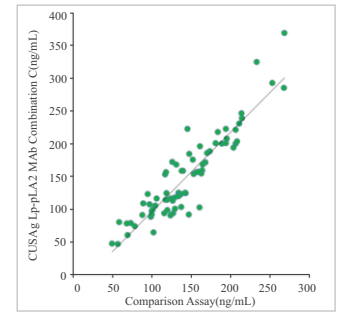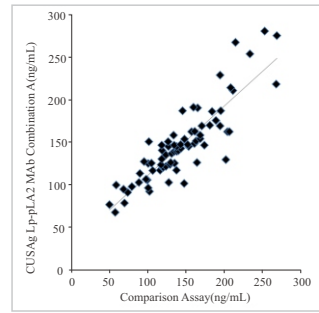

Fig.2 Determination of clinical samples on CLIA using CUSAg MAb combinations

❷ Precision

In order to assure the qualities of Lp-PLA2 monoclonal antibodies, different batches of MAbs were evaluated via our microplate-chemiluminescent immunoassay. The precisions of CUSAg CLIA Lp-PLA2 assays are all ≤5% in intra assays and ≤10% in inter assays. Data from these tests are summarized in the following table.

Pairs	Control	Intra assay (n=10)			Inter assay (n=30)		
		Mean Conc. (ng/mL)	SD	%CV	Mean Conc. (ng/mL)	SD	%CV
A	Control Low	152.94	4.49	2.9	152.38	10.51	6.9
	Control High	344.52	10.67	3.1	345.93	30.10	8.7
C	Control Low	157.21	5.76	3.7	157.67	14.51	9.2
	Control High	349.97	11.93	3.4	351.09	31.60	9.0
E	Control Low	150.49	6.19	4.1	149.81	8.53	5.7
	Control High	338.01	23.81	7.0	337.35	26.02	7.7

❸ Recovery

Known concentrations of Lp-PLA2 were added to five aliquots of human serum. The concentration of Lp-PLA2 was determined using the CUSAg CLIA platform and the resulting recovery percentage was calculated. The recovery percentage mean values of the Lp-PLA2 immunoassays using four MAb combinations were 99.9%, 100.7% and 101.7%, respectively.

❹ Clinical Comparison

81 clinical samples from apparently healthy donors and apparently patients with atherosclerosis or cerebral infarction were detected with the CUSAg CLIA Lp-PLA2 assays using varied MAb combinations, and

❺ Thermal Stability

All the anti-human Lp-PLA2 monoclonal antibodies presented in PBS buffer without any preservative were stored at -20°C, 2-8°C and 37°C for 15 days, respectively. During this period, the titers of four MAbs were determined, respectively. Fig.3 shows the relative titers of CSB-DA113BmN① and CSB-DA113BmN③ treated at 37°C or 2-8°C were similar with that at -20°C, but the titers of CSB-DA113BmN④ and CSB-DA113BmN⑤ weakly decrease at 37°C.
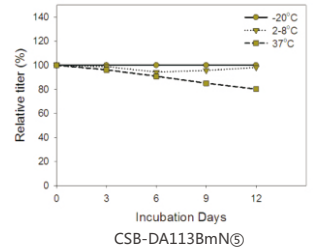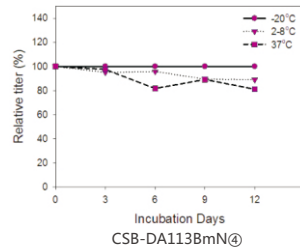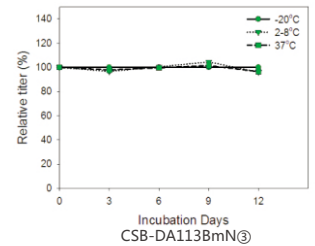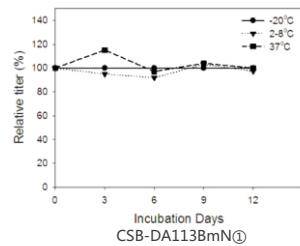

Fig.3 Thermal stability profiles of four monoclonal antibodies

Lp-PLA2 protein

A certain amount of excellent Lp-PLA2 protein (Cat: CSB-DP113B) is also offered by CUSAg. It could be used as calibrator in immunoassay.