

## Lipoteichoic acid

## Kit

(2011 / 9 / 26                      2011 / 5 / 2                      )

LTA

LTA

*St.viridans*

*S.aureus*

Kit

ELISA

$\pm 0.0141$      $1.0700 \pm 0.0141$ ) *St.viridans* – LTA    *S.aureus* - LTA

(1.0100

.(0.9300  $\pm$  0.0141)

## **Preparation of Laboratory Kit for Lipoteichoic acid as One of the Constituents of Gram-Positive Bacterial Cell Wall**

**Tuka M. Al-Sawaf**

**Amera M. Al-Rawi**

*Department of Biology*

*College of Science*

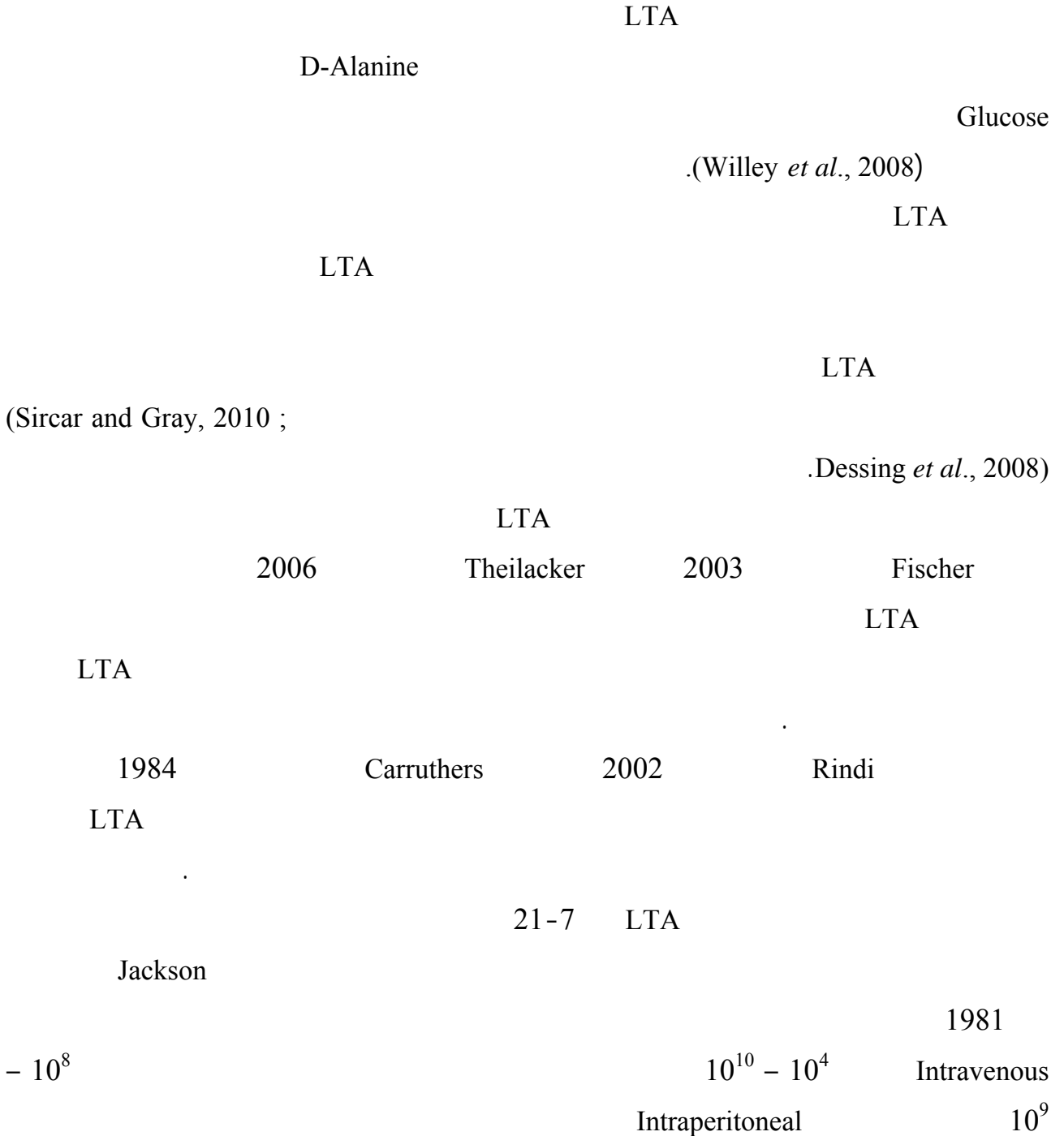
*University of Mosul*

### **ABSTRACT**

The study includes the production of antibody for LTA in the rabbit after isolation and purification from *S.aureus* and *St.viridans* by injecting the animals regularly and consecutively with pure LTA. Kit was prepared to investigate the presence of antibodies in sera of injected animals by ELISA, the absorbance of the test samples for each of *S.aureus*

- LTA and *St.viridans* - LTA were  $1.0700 \pm 0.0141$  and  $1.0100 \pm 0.0141$  respectively, The absorbance of control sample, which represent sera of lab animals injected with distilled water was  $0.9300 \pm 0.0141$ .

**Keywords** : lipoteichoic acid, antibody, ELISA.



3

.....

Kit

Polyglycerol phosphate LTA

LTA

**LTA**

(2010

) LTA

Antibody

LTA

LTA

Succinic Anhydride

3

LTA

LTA-Tyrosine

Succinic Anhydride

0.1 NaCl

Tris-HCl <sup>3</sup> 10

Succinic Anhydride 0.5

0.1) NaCl

<sup>3</sup> 2

<sup>3</sup> 2

LTA 30

1

<sup>3</sup> 30

(

.(Hasty *et al.*, 2006)

*S. aureus* - LTA

9

*St. viridans* - LTA

6

<sup>3</sup> 3-1

%70

3

.(Zamora *et al.*,1963)

7-5

30

15 / 3000

LTA IgG

(Kit) ELISA

LTA

20-

405

LTA

3

%70

*St. viridans* - LTA Antibody

*S. aureus* - LTA Antibody

6 6

( )<sup>3</sup> 0.2 - 0.15 - 0.1

ether

200:1

Phosphate buffer solution – Tween 20 (PBS-T20) 200 1

Alkaline Phosphatase 5

2005 Cuchacovich

LTA 50 LTA -1

.(1)

100 -2

° 37 -3

5

.....

Kit

300

-4

3

PBS-T

100

-5

.(1)

.° 37

-6

.5

-7

50

TMB

-

100

-8

30

-9

100

-10

405

30

-11

LTA

:1

Anti- <i>S.aureus</i> -LTA Antibody	<i>S.aureus</i> -LTA antibody	<i>S.aureus</i> -LTA
Anti- <i>St.viridans</i> -LTA Antibody	<i>St.viridans</i> -LTA antibody	<i>St.viridans</i> -LTA
Anti Serum Rabbit Control Antibody	Seurm Rabbit Control	<i>S.aureus</i> -LTA
Anti- <i>St.viridans</i> -LTA Antibody	<i>St.viridans</i> -LTA antibody	<i>S.aureus</i> -LTA
Anti- <i>S.aureus</i> -LTA Antibody	<i>S.aureus</i> -LTA antibody	<i>St.viridans</i> -LTA
Anti- <i>S.aureus</i> -LTA Antibody	Albumin	<i>S.aureus</i> -LTA
Anti- <i>S.aureus</i> -LTA Antibody		<i>S.aureus</i> -LTA

**LTA**

LTA

2006

Hasty

Ibrahim

LTA

LTA

1987

Lyons

Tyrosin

LTA

**ELISA****LTA****IgG**

ELISA

LTA

Kit

2005

Cuchacovich

*St.viridans* - LTA*S.aureus* - LTA

.(2)

LTA

:2

.ELISA

405			
		±	الامتصاصية العينات
		0.9300 ± 0.0141	( )
8.60	-	* 0.8500 ± 0.0141	
5.37	-	* 0.8800 ± 0.0141	
-	15.05	* 1.0700 ± 0.0141	<i>S.aureus</i> a
-	8.60	* 1.0100 ± 0.0141	<i>St.viridans</i> a
5.37	-	* 0.8800 ± 0.0141	<i>S.aureus</i> b
24.73	-	* 0.7000 ± 0.0141	<i>St.viridans</i> b

# .(P≤0.05)

LTA

LTA

= a

LTA

LTA

= b

.....

Kit

0.05

LTA

LTA

2004

Parmentier

LTA

21

LTA

2002

Yasuaki

Yuji

LTA

LTA

IgG

IgA IgG

ELISA

Phosphate Buffer Solution

1980

Beining

LTA

LTA

2003

Fischer

2002

Rindi

Endocarditis

IgG

*S. aureus*

LTA

1984

Carruthers

Endocarditis

*S. aureus*

LTA

Serological diagnosis

Osteomyelitis

(non- *S.aureus* osteomyelitis)

.LTA

*S. aureus*-LTA

(2)

*St. viridans* - LTA

LTA

LTA

2007

Toma

LTA

LTA

2006

Theilacher

LTA

*E. faecalis*

*E. faecium*

*E. faecalis*

*E. faecalis*

LTA

LTA

1986

Stashenko

*St. mutans*

LTA

*St. sanguis St. salivarius*

*. Actinomyces viscosus St. mitis*

1983

Jackson

Hether

LTA

*Listeria monocytogenes*

LTA

*.Streptococcus*

LTA

2005

Fischer

*St. mutans*

LTA

LTA

*.St. faecalis S. aureus*

0.05

.LTA



	.....	Kit	
Cuchacovich	2001	Gargir	
			2005
	%5	(LTA	)
			LTA
			LTA
	1989	Tollefsen Monefeldt	
	Gingivitis	Periodontitis	
LTA	0.05		
			.ELISA
			.(2010)

- Beining, P. R.; Flanery, G. M.; Prescott, B. ; Baker, P. J. (1980). Influence of Carrier-Specific, Thymus-Derived Cells on the Immunoglobulin M Antibody Response to Staphylococcal Lipoteichoic acid. *Infect. Immun.*, **29**(1), 132-139.
- Carruthers, M. M.; Jenkins, K. E.; Kabat, W. J. ; Buranosky, T. (1984). Detection of Antibody to Staphylococcal Lipoteichoic acid with a Microenzyme-Linked Immunosorbent Assay. *J. Clin. Microbiol.*, **19**(4), 552-554.
- Cuchacovich, M.; Merino, G.; Yamamoto, J. H.; Villarroel, F.; Saavedra, T.; Jofre, S.; Gatica, H.; Velasquez, V.; Pizzo, S. V. ; Gonzalez-Gronow, M. (2005). Behget's disease patients present high levels of deglycosylated anti-lipoteichoic acid IgG and high IL-8 production after lipoteichoic acid stimulation. *Clin. Exp. Rheumatol.*, **23**, 27-34.
- Dessing, M. C.; Schouten, M.; Draing, C.; Levi, M.; Aulock, S. ; Poll, T. (2008). Role played by Toll-like receptors 2 and 4 in lipoteichoic acid-induced lung inflammation and coagulation. *J. Infect. Dis.*, **197**(2), 245-252.
- Fischer, G. W.; Schuman, R. F.; Wong, H. ; Stinson, J. R. (2003). Opsonic and protective monoclonal and chimeric antibody specific for lipoteichoic acid of gram positive bacteria. WWW. free patents on line. Com / 6610293.
- Fischer, G. W.; Schuman, R. F.; Wong, H. ; Stinson, J. R. (2005). Humanized and chimeric antibodies specific for lipoteichoic acid of gram positive bacteria . WWW. free patents on line. Com / 7511122.

- Gargir, A.; Ofek, I.; Hasty, D.; Meron-Sudai, S.; Tsubery, H.; Keisari, Y.; Nissim, A. (2001). Inhibition of antibody-dependent stimulation of lipoteichoic acid-treated human monocytes and macrophages by polyglycerolphosphate-reactive peptides. *J. Leukoc. Biol.*, **70**, 537-542.
- Hasty, D. L.; Meron-Sudai, S.; Cox, K. H.; Nagorna, T.; Ruiz-Bustos, E.; Losi, E.; Courtney, H. S.; Mahrous, E. A.; Lee, R. ; Ofek, I. (2006). Monocyte and Macrophage activation by Lipoteichoic acid Is Independent of Alanine and is Potentiated by Hemoglobin. *J. Immunol.*, **176**, 5567-5576.
- Hether, N. W. ; Jackson, L. L. (1983). Lipoteichoic acid from *Listeria monocytogenes*. *J. Bacteriol.*, **156**(2), 809–817.
- Ibrahim, G. F. ; Lyons M. J. (1987). Production of potent polyclonal antibodies to bacterial membrane amphiphiles. *Med. Microbiol. Immunol.*, **176**, 305-313.
- Jackson, D. E.; Wicken, A. J. ; Jackson, G. D. F. (1981). Immune responses to lipoteichoic acid: Comparison of antibody responses in Rabbits and Mice. *Int. Archs. Allergy appl. Immun.*, **65**, 203-211.
- Monefeldt, K. ; Tollefsen, T. (1989). Serum IgG antibodies reactive with lipoteichoic acid in adult patients with periodontitis. *J. Clin.Periodontol.*, **16**(8), 519-524.
- Parmentier, H. K.; Kieboom, W. J. A. V. D.; Nieuwland, M. G. B.; Reilingh, G. D. V.; Hangalapura, B. N.; Savelkoul, H. F. J. ; Lammers, A. (2004). Differential Effects of Lipopolysaccharide and Lipoteichoic Acid on the Primary Antibody Response to Keyhole Limpet Hemocyanin of Chickens Selected for High or Low Antibody Responses to Sheep Red Blood Cells. *Poultry Science*, **83**, 1133-1139.
- Rindi, S.; Cicalini, S.; Pietrocola, G.; Petrosillo, N. ; Speziale, P. (2002). Antibody response to Staphylococcal slime and lipoteichoic acid. *The Lancet*, **360**, 1977-1978.
- Sircar, A. ; Gray, J. J. (2010). Snug Dock: Paratope Structural Optimization during Antibody-Antigen Docking Compensates for Errors in Antibody Homology Models. *PLoS Comput. Biol.*, **6**(1), 1-13.
- Stashenko, P.; Peros, W. J.; Gibbons, R. J. ; Dearborn, S. M. (1986). Effect of monoclonal antibodies against lipoteichoic acid from the oral bacterium *Streptococcus mutans* on its adhesion and plaque-accumulation *in vitro*. *Oral Biol.*, **31**(7), 455-461.
- Theilacker, C.; Kaczynski, Z.; Kropec, A.; Fabretti, F.; Sange, T.; Holst, O. ; Huebner, J. (2006). Opsonic Antibodies to *Enterococcus faecalis* Strain 12030 are directed against Lipoteichoic acid. *Infect. Immun.*, **74**(10), 5703-5712.
- Toma, I.; Theilacker, C.; Sava, I.; Kropec, A.; Hammer, F. ; Huebner, J. (2007). Antibodies against LTA isolated from *E. faecalis* 12030 recognize LTA from heterologous enterococcal strains but mediate opsonophagocytic killing only to CPS-A and CPS-B strains. *Eur. Society Clin. Microbiol. Infect. Dis.*, **29**, 512-518.
- Willey, J. M.; Sherwood, L. M. ; Woolverton, C. J. (2008). " Prescott, Harely and Klein Microbiology ". 7th edn., Mc Graw-Hill book companies Inc., New York.
- Yuji, Y. ; Yasuaki, H. (2002) . Intranasal immunization with lipoteichoic acid and cholera toxin evokes specific pharyngeal IgA and systemic IgG responses and inhibits streptococcal adherence to pharyngeal epithelial cells in mice. *J. pediatric otorhinolaryngol.*, **63**(3), 235-241.
- Zamora, A., Bojalh, E. E. ; Basearrachha, F. (1963). Immunologically active polysaccharide from *Nocardia asteroides* and *Nocardia brasiliensis*. *J. Bacteriol.*, **85**, 549-555.