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**The results of
fibrinolytic surveys**



Pilot survey 2009

PAI-1 activity and antigen

t-PA antigen

Plasmin Inhibitor (alpha-2-antiplasmin)

Plasminogen

PAI-1 and t-PA: broad range of between laboratory CV

Need for standardisation of PAI-1 and t-PA methods
and improvement of the between laboratory variation



Fibrinolytic surveys in EQA-programme 2010

EQA-programme was extended with two fibrinolytic modules

One containing the parameters plasminogen and plasmin inhibitor and the other PAI-1 and t-PA.

50 participants signed up for these new fibrinolytic modules



Number of responders and participation rate

	2010.1		2010.2	
	responders	Part. Rate (%)	responders	Part. Rate (%)
Plasminogen act.	35	71	38	76
Plasminogen ag.	6	12	5	10
Plasmin inhibitor	27	55	30	60
PAI-1 antigen	17	35	16	32
PAI-1 activity	14	29	17	34
t-PA antigen	14	29	13	26



Samples used in the fibrinolytic surveys

In each survey 2 lyophilized control samples were used from different manufacturers

2010.1: normal and abnormal control plasma (Siemens)

2010.2: coagulation control abnormal and borderline
(Technoclone)



Plasminogen activity (%)

2010.1

<u>Plasminogen act. (sample 10.20)</u>	n	mean	CV	range
Total group	34	95.5	8.0%	81 - 112
Biophen Plasminogen (Hyphen)	1	88.0		
Coamatic Plasminogen (Chromogenix)	3	89.3		86 - 92
HemosIL Plasminogen (I.L.)	5	91.1		87 - 97
Plasminogen (Siemens)	19	98.9	7.1%	86 - 112
Plasminogen (Stago / Roche)	6	92.8		81 - 105

<u>Plasminogen act. (sample 10.21)</u>	n	mean	CV	Range
Total group	33	32.2	9.8%	24 - 40
Biophen Plasminogen (Hyphen)	1	28.0		
Coamatic Plasminogen (Chromogenix)	3	30.7		28 - 34
HemosIL Plasminogen (I.L.)	5	31.5		29 - 34
Plasminogen (Siemens)	18	32.8	11.4%	24 - 40
Plasminogen (Stago / Roche)	6	32.7		31 - 34

2010.2

<u>Plasminogen act. (sample 10.48)</u>	n	mean	CV	range
Total group	37	47.9	9.2%	39 - 58
Biophen Plasminogen (Hyphen)	1	46.0		
Coamatic Plasminogen (Chromogenix)	5	46.8		43 - 53
HemosIL Plasminogen (I.L.)	7	46.4		44 - 49
Plasminogen (Siemens)	17	49.7	11.0%	39 - 58
Plasminogen (Stago / Roche)	7	46.0		43 - 49

<u>Plasminogen act. (sample 10.49)</u>	n	mean	CV	Range
Total group	37	49.7	11.0%	42 - 60
Biophen Plasminogen (Hyphen)	1	46.0		
Coamatic Plasminogen (Chromogenix)	5	46.4		42 - 55
HemosIL Plasminogen (I.L.)	7	45.9		44 - 50
Plasminogen (Siemens)	17	53.3	10.2%	44 - 60
Plasminogen (Stago / Roche)	7	47.3		42 - 51



Plasmin Inhibitor (%)

2010.1

<u>Plasmin Inhibitor (sample 10.20)</u>	n	mean	CV	range
Total group	27	99.6	9.3%	86 – 120
Coamatic Plasmin Inhibitor (Chromogenix)	4	101.8		86 – 109
HemosIL Plasmin Inhibitor (I.L.)	4	92.7		89 – 95
Antiplasmin (Siemens)	14	101.2	8.0%	89 – 120
Antiplasmin (Stago / Roche)	5	98.8		88 - 119

<u>Plasmin Inhibitor (sample 10.21)</u>	n	mean	CV	range
Total group	27	36.7	12.9%	28 – 47
Coamatic Plasmin Inhibitor (Chromogenix)	4	33.8		28 – 40
HemosIL Plasmin Inhibitor (I.L.)	4	35.2		30 – 38
Antiplasmin (Siemens)	14	37.8	13.3%	29 – 47
Antiplasmin (Stago / Roche)	5	37.0		31 – 43

2010.2

<u>Plasmin Inhibitor (sample 10.48)</u>	n	mean	CV	range
Total group	29	56.5	9.4%	44 - 67
Coamatic Plasmin Inhibitor (Chromogenix)	4	54.3		44 – 64
HemosIL Plasmin Inhibitor (I.L.)	5	57.4		54 - 67
Antiplasmin (Siemens)	14	57.6	7.9%	49 – 66
Antiplasmin (Stago / Roche)	6	54.7		44 - 58

<u>Plasmin Inhibitor (sample 10.49)</u>	n	mean	CV	range
Total group	29	53.0	13.7%	37 – 79
Coamatic Plasmin Inhibitor (Chromogenix)	4	51.8		46 – 56
HemosIL Plasmin Inhibitor (I.L.)	5	51.4		47 – 58
Antiplasmin (Siemens)	14	52.3	9.4%	44 – 59
Antiplasmin (Stago / Roche)	6	56.7		37 - 79



PAI-1 antigen (ng/ml)

2010.1

<u>PAI-1 antigen (sample 10.20)</u>	n	mean	CV	range
Total group	17	14.2	37.3%	6.0 – 26.0
Immubind PAI-1 (American Diagnostica)	1	15.2		
PAI-1 antigen (Haemochrom)	1	21.0		
Zymutest PAI-1 (Hyphen-Biomed)	5	13.4		6.0 – 26.0
Asserachrom PAI-1 (Stago/Roche)	4	12.4		10.0 – 14.7
TintElize PAI-1 (Trinity Biotech)	6	14.7		12.0 – 23.3

<u>PAI-1 antigen (sample 10.21)</u>	n	mean	CV	range
Total group	17	2.6	53%	1.0 – 6.0
Immubind PAI-1 (American Diagnostica)	1	2.6		
PAI-1 antigen (Haemochrom)	1	2.0		
Zymutest PAI-1 (Hyphen-Biomed)	5	2.2		1.0 – 5.0
Asserachrom PAI-1 (Stago/Roche)	4	3.2		1.0 – 6.0
TintElize PAI-1 (Trinity Biotech)	6	2.7		2.0 – 4.0

2010.2

<u>PAI-1 antigen (sample 10.48)</u>	n	mean	CV	range
Total group	16	6.3	67.2%	1.9 – 19.0
Immubind PAI-1 (American Diagnostica)	1	2.9		
PAI-1 antigen (Haemochrom)				
Zymutest PAI-1 (Hyphen-Biomed)	5	6.7		1.9 – 19.0
Asserachrom PAI-1 (Stago/Roche)	4	5.2		4.0 – 6.0
TintElize PAI-1 (Trinity Biotech)	5	6.8		5.0 – 12.8
Unknown	1	10.0		

<u>PAI-1 antigen (sample 10.49)</u>	n	mean	CV	range
Total group	15	24.1	78.8%	4.6 – 67.0
Immubind PAI-1 (American Diagnostica)	1	4.6		
PAI-1 antigen (Haemochrom)				
Zymutest PAI-1 (Hyphen-Biomed)	4	23.3		4.9 – 67.0
Asserachrom PAI-1 (Stago/Roche)	4	20.2		18.0 – 23.0
TintElize PAI-1 (Trinity Biotech)	5	27.6		17.9 – 62.4
Unknown	1	45.0		



PAI-1 activity (IU/ml)

<u>PAI-1 activity (sample 10.20)</u>	n	mean	CV	range
Total group	14	3.4	60.5%	1.0 – 6.9
Berichrom PAI-1 (Siemens)	3	1.8		1.0 – 2.9
Stachrom PAI-1 (Stago/Roche)	2	6.3		5.7 – 6.9
Technozyme PAI-1 activity (Technoclone)	2	1.4		1.0 – 1.8
Spectrolyse PAI-1 (Trinity Biotech)	1	6.0		
Chromolyze PAI-1 (Trinity Biotech)	6	3.4		1.9 – 6.0

<u>PAI-1 activity (sample 10.21)</u>	n	mean	CV	range
Total group	13	1.6	111%	0.0 – 6.0
Berichrom PAI-1 (Siemens)	3	0.0		0.0 – 0.0
Stachrom PAI-1 (Stago/Roche)	1	13.5		
Technozyme PAI-1 activity (Technoclone)	2	0.6		0.1 – 1.0
Spectrolyse PAI-1 (Trinity Biotech)	1	3.0		
Chromolyze PAI-1 (Trinity Biotech)	6	1.7		0.5 – 6.0

<u>PAI-1 activity (sample 10.48)</u>	n	mean	CV	range
Total group	14	4.1	92.7%	1.0 – 11.2
Berichrom PAI-1 (Siemens)	1	0.0		
Zymutest PAI-1 activity (Hyphen-Biomed)	1	1.0		
Stachrom PAI-1 (Stago/Roche)	2	10.5		9.8 – 11.2
Technozyme PAI-1 activity (Technoclone)	2	6.0		1.0 – 11.0
Spectrolyse PAI-1 (Trinity Biotech)				
Chromolyze PAI-1 (Trinity Biotech)	6	3.1		1.7 – 6.0
TriniELize PAI-1 activity (Trinity Biotech)	2	2.0		2.0 – 2.0

<u>PAI-1 activity (sample 10.49)</u>	n	mean	CV	range
Total group	15	5.8	47.4%	0.1 – 9.5
Berichrom PAI-1 (Siemens)	2	0.8		0.1 – 1.5
Zymutest PAI-1 activity (Hyphen-Biomed)	1	3.2		
Stachrom PAI-1 (Stago/Roche)	2	5.4		5.3 – 5.6
Technozyme PAI-1 activity (Technoclone)	2	5.4		5.0 – 5.8
Spectrolyse PAI-1 (Trinity Biotech)				
Chromolyze PAI-1 (Trinity Biotech)	6	8.5		7.6 – 9.5
TriniELize PAI-1 activity (Trinity Biotech)	2	5.0		4.0 – 6.0



t-PA antigen (ng/ml)

<u>t-PA antigen (sample 10.20)</u>	n	mean	CV	range
Total group	14	3.8	38.7%	1.0 – 5.9
Imubind t-PA (American Diagnostica)	1	3.8		
Elitest t-PA (Aniara)	1	4.2		
Zymutest t-PA (Hyphen-Biomed)	4	2.8		1.0 – 4.5
Asserachrom t-PA (Stago/Roche)	2	5.5		5.0 – 5.9
Technozyme t-PA (Technoclone)	2	2.2		2.1 – 2.3
TintElize t-PA (Trinity Biotech)	4	4.7		4.3 – 5.1

<u>t-PA antigen (sample 10.21)</u>	n	mean	CV	range
Total group	14	1.9	48.1%	0.5 – 4.0
Imubind t-PA (American Diagnostica)	1	2.4		
Elitest t-PA (Aniara)	1	1.3		
Zymutest t-PA (Hyphen-Biomed)	4	1.6		0.5 – 4.0
Asserachrom t-PA (Stago/Roche)	2	1.9		1.7 – 2.0
Technozyme t-PA (Technoclone)	2	1.7		1.1 – 2.3
TintElize t-PA (Trinity Biotech)	4	2.2		1.8 – 2.9

<u>t-PA antigen (sample 10.48)</u>	n	mean	CV	range
Total group	13	2.9	67.8%	0.4 – 7.6
Imubind t-PA (American Diagnostica)	1	7.6		
Elitest t-PA (Aniara)				
Zymutest t-PA (Hyphen-Biomed)	5	1.6		0.4 – 2.9
Asserachrom t-PA (Stago/Roche)	2	3.0		2.0 – 4.0
Technozyme t-PA (Technoclone)	2	1.7		1.2 – 2.3
TintElize t-PA (Trinity Biotech)	3	4.0		2.9 – 4.6

<u>t-PA antigen (sample 10.49)</u>	n	mean	CV	range
Total group	11	3.8	55.1%	0.8 – 6.9
Imubind t-PA (American Diagnostica)				
Elitest t-PA (Aniara)				
Zymutest t-PA (Hyphen-Biomed)	4	2.5		0.8 – 4.3
Asserachrom t-PA (Stago/Roche)	2	5.0		4.0 – 6.0
Technozyme t-PA (Technoclone)	2	1.7		1.2 – 2.3
TintElize t-PA (Trinity Biotech)	3	6.1		5.1 – 6.9



Between laboratory variation and assay type

	Assay-type	Range total group CV (%)	Concentration range
Plasminogen act. (%)	chromogenic	8 - 11	32 - 96
Plasmin inhibitor (%)	chromogenic	9 - 14	37 - 100
PAI-1 antigen (ng/ml)	ELISA	37 - 79	3 - 24
PAI-1 activity (IU/ml)	BIA/ELISA chromogenic	47 - 111	2 - 6
t-PA antigen (ng/ml)	ELISA	39 - 68	2 - 4



For PAI-1 antigen and activity, and t-PA antigen a broad range for the total group CV is observed.

This is also reflected in the different classification of the control samples; illustrated with the abnormal control plasma in survey 2010.1



Classification abnormal control sample 2010.1

	Normal	Borderline normal	Borderline abnormal	Abnormal	No classification
PAI-1 antigen	8	2	0	6	1
PAI-1 activity	7	1	1	5	0
t-PA antigen	9	1	1	3	0



In conclusion

For plasmin inhibitor activity and plasminogen activity, in which chromogenic methods are used, results of different methods are fairly good comparable.

The much higher between laboratory variation for PAI-1 activity and antigen, and t-PA antigen show the lack of standardisation in these measurements, mainly due to the heterogeneity of the antibodies used and their specificity.



In conclusion

The results of the first two surveys on fibrinolytic parameters of the ECAT foundation showed the need for standardisation for PAI-1 and t-PA methods and improvement of the between laboratory variation.

