

# ECAT FOUNDATION

**External quality Control of diagnostic Assays and Tests**  
*with a focus on Thrombosis and Haemostasis*



**QUALITY PROGRAMME  
MANUAL 2011**



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## **1. GENERAL INFORMATION**

### **1.1 ECAT OFFICE**

#### **Postal address:**

ECAT FOUNDATION  
P.O. BOX 30  
2300 AA Leiden  
The Netherlands

#### **Visiting address:**

ECAT Foundation  
Wassenaarseweg 56  
Building: "De Luistervink"  
2333 AL Leiden  
The Netherlands

#### **Contact details:**

T. +31 88 866 9718  
F. +31 88 866 8965  
E. [info@ecat.nl](mailto:info@ecat.nl)  
W. [www.ecat.nl](http://www.ecat.nl)

#### **Contact details Financial Department:**

T. +31 88 866 9638  
F. +31 88 866 8965  
E. [finance@ecat.nl](mailto:finance@ecat.nl)

#### **Organisation:**

<b><u>Name</u></b>	<b><u>Position</u></b>	<b><u>Duties</u></b>
Dr. P. Meijer	Director	General managing duties Statistics Scientific affairs
Mrs. A. Veninga	Scheme Manager/Quality Manager	General information Registration Organisation surveys Website content
Mrs. M. Van der Voorn	Deputy Scheme Manager	Organisation surveys Sample management
Mrs. G. Zandbergen	Financial co-worker Survey Assistant	Financial Administration Support organisation surveys
Mrs. P. Ter Hark	Editor-in-chief	CLOT-ED

### **1.2 LEGAL ENTITY**

The ECAT is an independent and impartial organization. Its legal entity is a foundation directed by a board. The board of the ECAT Foundation serves as the Scientific Advisory Board. Members of the board are well experienced in the field of thrombosis and haemostasis.

#### **Board members:**

<b><u>Name</u></b>	<b><u>Specialism</u></b>	<b><u>Board function</u></b>
Dr. F. Haas	Clinical chemist	
Prof. Dr. C. Kluft	Biochemist	Chairman
Dr. M.P.M. de Maat	Biochemist/epidemiologists/clinical chemist	Secretary/ Deputy chairman
Dr. R. Niessen	Clinical chemist	
Dr. O. Paauwe	Director Thrombosis Foundation Netherlands	
Dr. H.W. Verbruggen	Biochemist	
Ir. D.C. van Cuilenburg	Accountant	Treasurer

The ECAT Foundations is registered at the Dutch Chamber of Commerce in Rotterdam, number 41174102



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## 1.3 SUBCONTRACTED ACTIVITIES

### ICT-related issues:

- Development and maintenance web based data submission: Biolink BV, Rotterdam
- Web and database hosting: Lindix BV, Almere
- Web design and maintenance: Goedknecht Design, Rotterdam

### Laboratory testing:

- Testing for homogeneity and stability: Erasmus Medical Centre, Rotterdam

### Sample preparation:

- Hyphen Biomed, France
- Technoclone, Austria
- Verum Diagnostics, Germany
- Recipe, Germany
- WIV, Belgium

## 1.4 SCOPE

The ECAT Foundation has been operating External Quality Assessment Programmes (EQAP) in the field of thrombosis and haemostasis since 1992.

The primary aim of the ECAT Foundation is to contribute to quality improvement of clinical laboratories operating within the field of thrombosis and haemostasis with respect to the diagnosis and treatment of patients.

The ECAT is currently in the process of accreditation according to the international guideline ISO 17043.

## 1.5 WEBSITE “www.ecat.nl”

The ECAT website gives general information about the ECAT quality assessment programme as well as information about previous and forthcoming meetings and the educational branch of ECAT, CLOT-ED (see also chapter 5).

There is a specific password-protected member section which contains the result submission facility, exercise instructions, manuals, long-term evaluation tool and questionnaires.

To contact the ECAT, a form is available in the “contact us” section. If you are a registered participants, please always add your laboratory code on the form.

## 1.6 TERMS OF DELIVERY

The terms of delivery of the ECAT Foundation can be found on our website. If you want a printed copy, please contact our office.

## 1.7 COMPLAINTS AND REQUESTS

If participants have any complaint or request, the ECAT should be contacted immediately, preferably by using the contact form on our website. The participant should provide ECAT with their laboratory code and a clear description of the complaint or request. The ECAT will respond as soon as possible to any complaint or request submitted.



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## 2. PARTICIPATION

### 2.1 PROGRAMME INFORMATION

The ECAT is based in The Netherlands but provides EQAP for assays and tests in the field of thrombosis and haemostasis on an international scale. The programme is open for every laboratory providing services in the mentioned discipline.

The EQAP of the ECAT is a modular programme. The main programme of the ECAT consists of the following modules:

<u>Description</u>	Number of exercises per year	Number of different samples per exercise	Sample material (lyophilized)	Measuring range (approx.)	Number of participants	Annual price in € excl VAT
<b>Screen assays:</b> APTT, PT/INR, Fibrinogen	4	2	plasma	normal – prolonged normal – prolonged 100 – 400 mg/dL	82	85
<b>Thrombophilia module:</b> Anti-thrombin (activity and antigen), Protein C (activity [chromogenic and clotting] and antigen), Protein S activity, Protein S antigen (total and free), APC Resistance	4	2	plasma	20 – 120% normal / FV Leiden	317	400
<b>Protein C Pathway Test</b>	4	2	plasma	normal / abnormal	16	55
<b>Lupus Anticoagulant / Antiphospholipid Antibodies</b>	4	1	plasma	negative - positive	454	110
<b>D-Dimer</b>	4	2	plasma	normal - elevated	660	105
<b>Coagulation Factor module I</b> (Factor VIII, IX, XI and XII)	4	2	plasma	0 – 200%	223	110
<b>Coagulation Factor module II</b> (Factor II, V, VII and X)	4	2	plasma	0 – 200%	194	110
<b>Von Willebrand Factor module</b> (antigen, activity, collagen binding, multimers, Factor VIII)	4	1	plasma	0 – 125%	245	110
<b>Factor VIII inhibitor</b>	2	2	plasma	0 – 5 BU/mL	228	110
<b>Thrombin Generation Test</b>	2	3	plasma	normal / abnormal	43	110
<b>HIT – Immunological assays</b>	2	2	plasma/ serum	negative / positive	240	125
<b>HIT – Functional assays</b>	2	2	plasma/ serum	negative / positive	58	150
<b>Factor XIII</b>	4	2	plasma	0 – 120%	100	110
<b>Fibrinolysis parameters I</b> (Plasminogen, Antiplasmin)	4	2	plasma	0 – 120%	88	110
<b>Fibrinolysis parameters II</b> (t-PA, PAI-1)	4	2	plasma	0 – 50 ng/mL	42	110
<b>Unfractionated Heparin Monitoring</b> (anti-Xa)	4	2	plasma	0 – 1.25 IU/mL	76	100
<b>Low-Molecular Weight Heparin Monitoring</b> (anti-Xa)	4	2	plasma	0 – 1.25 IU/mL	124	100
<b>Homocysteine</b>	4	2	serum	0 – 100 µmol/L	72	85



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The samples used in the exercises are plasma and serum based. Part of the samples is from commercial source. For abnormal samples real patient plasma is used when appropriate. Samples are provided as lyophilized material.

After receipt, the unreconstituted lyophilised plasma should immediately be stored at 2 - 8°C. Reconstituted plasma should preferably be used within 1 hour after reconstitution. Plasma should be stored at room temperature after reconstitution. For immunological methods the reconstituted plasma can be stored for 1 months at -20°C.

The ECAT samples should be treated as regular patient plasmas and included in the normal daily analytical process in the laboratory. Results should be reported similar as a result of a patient is reported.

## **Electronic post-analytical platelet function survey:**

In co-operation with the NASCOLA in the United States twice a year a post-analytical survey for platelet function testing is provided.

The annual price of this survey is:

€ 110 excl VAT

## **Molecular Biology:**

In co-operation with the DGKL in Germany twice a year an EQA programme for Molecular Diagnostic Testing is provided. This programme exists of the following modules:

Description	Number of surveys per year	Sample material	Annual price in € excl VAT
<b>Molecular Biology Set A:</b> FV-Leiden, Prothrombin, MTHFR (C677T, A1298C), PAI-1 4g5g	2	DNA preparation, lyophilized	**
<b>Molecular Biology Set B:</b> FXIII V34L, GPIIIa, βFib g-455a, VKORC1 (g-1639a/c1173t), FXII c46t, FV H1299R	2	„	**
<b>Molecular Biology Set C:</b> a1 PI, Apo E, Apo B100, ACE, CETP	2	„	**
<b>Molecular Biology Set D:</b> TPMT, Cyp2C19 *1/*2/*17, Cyp2D6, Cyp2C8 (K399R), Cyp2C9 *2/*3, UGT1a1 (*28), DPD Exon 14 skipping, BCHE A/K	2	„	**
<b>Molecular Biology Set E:</b> ALDO B (149/174/334), HFE (H63D, C282Y, S65C), LCT c-13910t, NOD2 (R702W, G908R, L1007fins C)	2	„	**
<b>Molecular Biology Set F:</b> M. Wilson ATP7B-C3207 A, FSAP (Marburg-I), ITGA2 Gplalla C807T	2	„	**
<b>Molecular Biology Set G:</b> K-Ras: Codon 12/13/61	2	„	**
<b>DNA Sequencing</b> (Sequencing and corresponding diagnostic interpretation)	2	„	200
<b>DNA Isolation</b> (DNA isolation and FV genotyping)	2	Full blood, K-EDTA	90

\*\* One set : € 86,=; Two sets: € 122,=; Three sets: € 158,=; Four sets: € 194,=; Five sets: € 230,=; Six sets: € 266,=; Seven sets € 302,=.

The ECAT Foundation is not responsible for either the content or the evaluation of the test results of the Electronic survey and the Molecular Biology part of the programme.



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## **2.2 REGISTRATION**

The available modules and corresponding prices are updated annually. Either combination of modules can be selected. It is possible to start any time during the year. The registration will start the first exercise scheduled after the registration is received and will continue until the end of the year.

To register for our programme new participants can return the "Registration Form". The "Registration Form" is available on our website or can be ordered at our office. Registration forms are only accepted when signed to confirm that the new participants agrees with our "terms of delivery".

After registration new participants receive a welcome letter with information about their registration, an unique laboratory code and the website login codes. Together with this letter the Exercise Instruction Manual is supplied.

## **2.3 PAYMENT**

The invoice is sent to new participants after registration. Payment should be done by bank transfer. Cheques are not accepted. When the invoice is not paid in due time the registration will be cancelled.

For participants of one of the EMU countries we ask for a VAT number. If the VAT number of your organisation is not available at our Financial Department, the ECAT is legally obliged to add 19% VAT to the invoice.

## **2.4 ANNUAL SUBSCRIPTION**

Annually participants receive a subscription form to confirm their profile for the next year. This ensures that all participants are informed about added or deleted modules in the ECAT programme. The subscription form has to be completed and returned to the ECAT office before the end of the year.

## **2.5 CANCELLATION POLICY**

Cancellation is only accepted at the end of a year by a written confirmation.

## **3. EXERCISES**

### **3.1 EXERCISE SCHEDULE**

Every year the ECAT Foundation prepares the annual Exercise Schedule. In this schedule information is given about:

- Exercise: code number of the exercise
- Dispatch date: date that we send the samples to all participants
- Closing date: date that result submission closes (fax/postal service and website)
- Closing date D-Dimer: date that D-Dimer result submission closes (shorter turn-over time)

All participants receive a copy of this schedule together with their subscription form, so they know in advance when to expect the samples.



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## The Exercise Schedule of 2011:

### GENERAL PROGRAMME

Exercise	Dispatch Date	Closing Date	Closing Date D-Dimer
2011-1	8 March	13 April	23 March
2011-2	24 May	29 June	8 June
2011-3	30 August	5 October	14 September
2011-4	8 November	14 December	23 November

### MOLECULAR BIOLOGY

Exercise	Dispatch Date	Closing Date
FV1/11	23 March	16 April
FV2/11	28 September	22 October

### DNA SEQUENCING

Exercise	Dispatch Date	Closing Date
SQ1/11	2 February	26 February
SQ2/11	7 September	1 October

### DNA ISOLATION

Exercise	Dispatch Date	Closing Date
DI1/11	16 March	9 April
DI2/11	7 September	1 October

## **3.2 EXERCISE INSTRUCTION MANUAL**

Every year participants receive an updated Exercise Instruction Manual. This Manual gives instructions how to perform the exercises. This includes:

- the exercise schedule
- information about the reconstitution and measurement of samples
- instructions how to report results with the report forms or via the website
- explanation of the reports
- code lists of methods and equipments



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## **3.3 DISTRIBUTION OF SAMPLES**

Samples are sent to participants according to the exercise schedule. This can be two or four times a year. The frequency is clearly indicated on the subscription and registration forms.

The samples are packed in plastic bubble bags and carton boxes to prevent damage during transport. After receipt the samples should be stored at 2-8 degrees until use.

Each vial has a label with the ECAT logo and a sample code. This code corresponds with a code in the sample list on the exercise instruction. This sample code is also the identification code when results are reported.

## **3.4 EXERCISE INSTRUCTIONS**

Together with the samples the detailed Exercise Instructions are provided. These instructions include:

- information about the samples of each module
- the volume of reconstitution
- safety matters

Participants can also download the Exercise Instructions as well as the Exercise Instruction Manual from the member section at the website.

## **4. RESULTS**

### **4.1 RESULT SUBMISSION**

Exercise results can be reported either by fax/postal service or via internet. The ECAT prefers to have results reported via internet. For the use of fax/postal service an extra annual subscription fee will be charged (details can be found at our website).

To report results via post/fax pre-printed report forms are used. Each report form is identified with the appropriate laboratory code. In the Exercise Instruction Manual detailed instructions are given how to complete these report forms.

Reporting via internet can be done via our result submission facility in the member section of our website. This facility is password-protected. The password is provided to the participant during the registration procedure. In the Exercise Instruction Manual detailed instructions are given how to use this web-based result submission facility.

Besides the test results on the ECAT samples and the unit in which the result is expressed, information should be given on the assay principle, methodology and equipment used. For most of the parameters also a clinical classification of the samples is asked.

Inappropriate completion of the report forms may lead to exclusion of the results from the statistical evaluation.

Results returned after the exercise closing date will not be included in the statistical evaluation.



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## 4.2 STATISTICAL EVALUATION

For the external quality assessment programme of the ECAT the robust average of the results reported by all participants in the exercise is used as the assigned value (= consensus value). In accordance with ISO guideline 17043 and ISO guideline 13528 Algorithm A is used as a robust statistical algorithm for the calculation of the consensus value and the standard deviation.

The standard procedure for the evaluation of quantitative test results is as follows:

- Results are harmonised to the same unit (% / U/dL).
- The consensus value and standard deviation (SD) using Algorithm A.
- Based on this consensus value and SD the between-laboratory variation is calculated.

Algorithm A is applied on the total group and the level of assay type and method if there are at least 10 participants included in the same group (for the screening assays and homocysteine a minimum of 5 participants is used).

## 4.3 PERFORMANCE EVALUATION

As an individual performance indicator the Z-score is used. The Z-score indicates the distance between the participants' result and the consensus value expressed as a ratio of the standard deviation. The Z-score can be either positive or negative depending whether the participants' results is higher or lower than the consensus value.

The z-score is calculated as follows:

$$\frac{[(\text{laboratory result}) - (\text{mean result of all laboratories})]}{(\text{standard deviation of all results})}$$

The Z-score is also calculated for groups on the level of assay type and method with at least 10 participants. (for the screening assays and homocysteine a minimum of 5 participants is used).

### Acceptance criteria

Each participants should carefully evaluate the Z-scores given in the report. In accordance with ISO guideline 17043 and ISO guideline 15328 the following acceptance criteria are used:

-2 < Z-score < 2	:	The result is acceptable
-3 < Z-score < -2 or 2 < Z-score < 3	:	The results is questionable (warning signal)
Z-score < -3 or Z-score > 3	:	The result is unacceptable (action signal)

A single action signal or two warning signals in consecutive exercises shall be taken as evidence that a anomaly has occurred that requires investigation by the laboratory.

## 4.4 REPORTS

From each exercise the participant receive an evaluation report of the modules they have registered for including the results of all participants. These reports are sent by postal service to all participants. The position of the participants' own results in relation to all results are clearly presented both in the statistical tables as well as in histograms.



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The participants' performance is presented by the Z-score (see above) both in the statistical tables as well as in Z-score plots (only when two sample are distributed per exercise) and Z-score history plots.

The following reports are produced:

- Screen assays
- D-Dimer
- Lupus Anticoagulant
- Thrombin Generation Test
- HIT
- Main (including all modules not mentioned above)

## Report set-up

For each analyte a report sheet is given. The report sheet consists of three parts:

1. The header
2. The graph
3. The table

- **The header**

The header of each report sheet consist of two parts. At the top of each page the exercise number, number of pages of the report, the date the report is issued and the labcode is indicated. Also the name of the module and the analyte is indicated.

	<b>ECAT Foundation</b>	2011-1
	<b>External Quality Control for Assays and Test</b> <i>With a focus on Thrombosis and Haemostasis</i>	Page 1 of 82 06-July-2011 100
<b>Thrombophilia</b>		<b>Antithrombin activity</b>

In addition, information about the number of participants, the sample used and the units in which the results are reported are given.

When appropriate the clinical classification is given in a separate table.

<b>No. of Responders</b> 260					
<b>Sample No</b>	11.03				
<b>Sample Details</b>	Normal Coagulation Control Units: % or IU/dL Prior Use: None				
<b>Classification</b>	Normal	BorderLine normal	Borderline abnormal	Abnormal	No classification
<b>Total</b>	251	4	1	1	3

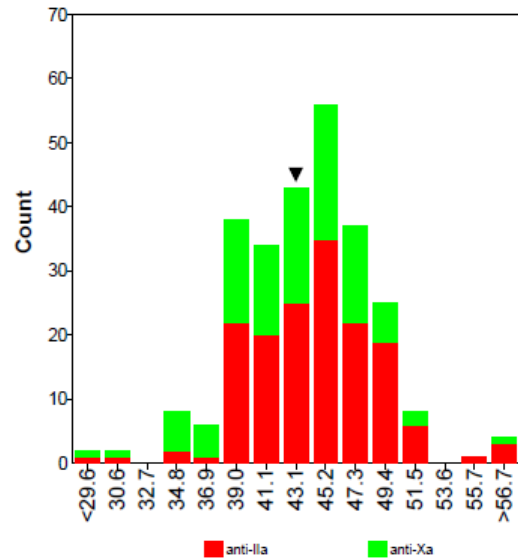


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- The graph**

The distribution of the results is represented in a histogram. Depending of the analyte the results are grouped based on the assay principle or the method used. The position of your own result within the distribution is indicated by a black arrow on top of the respective column in the histogram.



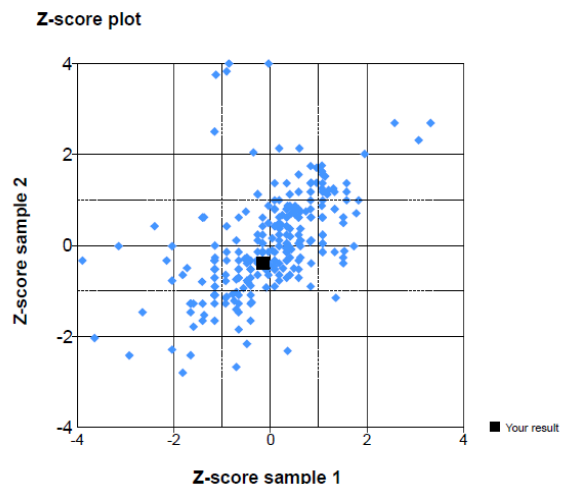
- The table**

The table given in the report show the descriptive statistics for all results and for each assay principle and method. The between-laboratory variation (CV) and Z-score are only given when at least 10 participants belong to the same group. The group(s) to which your results belong are highlighted in grey.

	n	mean	CV (%)	range	your result	z-score
<b>Total Group</b>	264	44.1	9.5	29 - 86	42.5	-0.38
<b>Chromogenix, anti-IIa</b>	158	44.6	9.0	29 - 86		
Beckman Coulter Electrachrom AT	1	40.0				
Homemade	1	39.0				
Hyphen Biophen AT (anti-IIa)	1	47.0				
Other	1	48.0				
Siemens Antithrombin III	59	45.3	8.2	29 - 86		
Stago/Roche Stachrom/Antithrombin	94	44.3	9.4	30 - 58		
Tcoag TriniCHROM Antithrombin IIa	1	42.0				
<b>Chromogenix, anti-Xa</b>	106	43.2	10.5	29 - 64	42.5	-0.15
Chromogenix Coamatic Antithrombin	27	41.4	13.8	30 - 64		
Chromogenix Coamatic LR Antithrombin	2	43.0		35 - 51		
DG-Chromat ATIII	1	50.0				
Hyphen Biophen Antithrombin (anti-Xa)	6	44.5		35 - 48		
Hyphen Biophen AT (LRT)	1	44.0				
I.L. HemosIL Antithrombin	4	47.0		37 - 50		
<b>I.L. HemosIL liquid Antithrombin</b>	41	42.8	9.6	29 - 50	42.5	-0.08
Other	1	38.0				
Siemens Innovance AT	23	44.8	3.5	42 - 48		

### The Z-score plots

In the case two different samples are distributed a Z-score plot is presented. In this Z-score plot the obtained Z-scores for both samples are presented in a Youden-plot. This provides the participant with insight whether the deviations from the consensus values are mostly related to random or to systematic errors.





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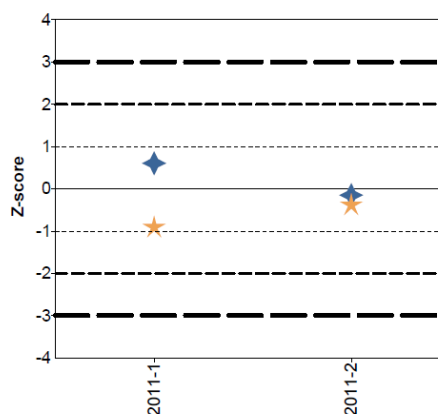
The table below shows for each of the parameters which Z-scores are used for the Z-score plot.

Description	Category of Z-score
Screen assays	reagent
<u>Thrombophilia module</u> Antithrombin, Protein C, Protein S APC Resistance	assay type method
Protein C Pathway Test	method
Lupus Anticoagulant / Antiphospholipid Antibodies	assay type
D-Dimer	method
Coagulation Factor module I (Factor VIII, IX, XI and XII)	assay type
Coagulation Factor module II (Factor II, V, VII and X)	assay type
Von Willebrand Factor module (antigen, activity, collagen binding, multimers, Factor VIII)	not applicable
Factor VIII inhibitor	assay type
Thrombin Generation Test	not applicable
HIT – Immunological assays	not applicable
HIT – Functional assays	not applicable
Factor XIII	assay type
Fibrinolysis parameters I (Plasminogen, Antiplasmin)	assay type
Fibrinolysis parameters II (t-PA, PAI-1)	assay type
Unfractionated Heparin Monitoring (anti-Xa)	assay type
Low-Molecular Weight Heparin Monitoring (anti-Xa)	assay type
Homocysteine	assay type

## The Z-score history plot

To provide the participant with insight in the history of their performance the Z-scores of the last 4 exercises are presented as a Z-score history plot. For this plot the same Z-scores are used as for the Z-core plots (see table above). For the Von Willebrand Factor module the Z-scores of the assay types are used.

Z-score history



## 4.5 ADDITIONAL USE OF RESULTS

Exercise results may be used for scientific purposes. In this case anonymous use of results will be guaranteed.

Individual exercise results will never be provided to commercial parties without permission of the participant.



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## **5. OTHER ACTIVITIES**

### **5.1 CLOT-ED (EDUCATIONAL BRANCH)**

The name, CLOT-ED, is an acronym. **CLOT** means **C**oagulation, (**f**ibrino)**L**ysis, **O**r **T**hrombosis. **ED** means Education.

The mission of CLOT-ED is to support and educate laboratory professionals with an interest in haemostasis (coagulation & fibrinolysis) and thrombosis by providing practical and concise information in order to improve the quality of laboratory testing related to these areas. CLOT-ED is a specific part at the ECAT website. There is an open-access part, containing for instance an international meeting calendar, terminology used in the field of thrombosis and haemostasis and a corporate corner which is a portal to the websites of diagnostic companies. The password-protected area contains a lot of educational resources. This part of CLOT-ED is accessible for participants in the ECAT external quality assessment programme. Separate registration for CLOT-ED is also possible.

Every year two issue of The Clotting Times are provided with a lot of background information on quality and laboratory testing related issues in the field of thrombosis and haemostasis. Previous issues of The Clotting Times can be found in the open-access part of CLOT-ED at the ECAT website.

### **5.2 WORKSHOPS**

On a regular basis the ECAT organises workshops on topics related to our programme. For example, workshops were organised on thrombin generation testing, inhibitor testing and platelet function testing.

### **5.3 BIENNIAL PARTICIPANTS' MEETING**

Every even year the ECAT organises a participants' meeting in Leiden, The Netherlands. The programme of this meeting focuses on laboratory-related topics in the field of thrombosis and haemostasis. Further information can be found at our website.

The next ECAT Participants' Meeting will be in November 2012. All participants are informed in advance about the details of the programme as well as registration procedure.