

# APC-PCI ELISA Kit (KIT 040)



BioPorto Diagnostics' APC-PCI ELISA Kit measures the Activated Protein C – Protein C Inhibitor complex in human plasma. This kit is for research use only. Not for use in diagnostic procedures.

## Key features

- The first and only commercially available APC-PCI ELISA kit on the market
- Precoated Microwell strips that can be used as strips of 8 or as individual wells
- Storage at 2-8°C

## Kit components

Item	Contents	Quantity
①	12 x 8 coated Microwells + Frame	96 wells
②	Sample Diluent	1 x 30 mL
③	APC-PCI Calibrator 2 glass vials each containing 1 ng freeze-dried APC-PCI	2 x 1 ng
④	25x Wash Solution Concentrate	1 x 40 mL
⑤	Biotinylated PC Antibody	1 x 12 mL
⑥	HRP-Streptavidin	1 x 12 mL
⑦	TMB Substrate	1 x 12 mL
⑧	Stop Solution	1 x 12 mL

## Ordering information

Cat. No.	Product name
KIT 040	APC-PCI ELISA Kit

For more information, please contact BioPorto Diagnostics or your local BioPorto Diagnostics distributor

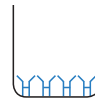


BioPorto Diagnostics A/S  
Grusbakken 8  
DK-2820 Gentofte  
Denmark

Phone (+45) 4529 0000  
Fax (+45) 4529 0001  
E-mail [info@bioporto.com](mailto:info@bioporto.com)  
Web [www.bioporto.com](http://www.bioporto.com)

## Assay Procedure

### APC-PCI antibody



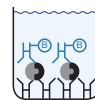
Plates are precoated with primary APC-PCI antibody. The plates are ready to use.

### APC-PCI complex



Samples and calibrators are added to each well and incubated.

### Biotinylated PC Antibody



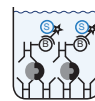
Biotinylated detection antibody is added to each well and incubated.

### HRP-Streptavidin



HRP-Streptavidin is added to each well and incubated.

### TMB Substrate



Substrate is added to each well. Develop for 10 minutes in the dark.

### Stop Solution

Stop Solution is added to each well. Quantitative results are obtained by measuring the absorbances of the wells at 450 nm.

Distributed by:

# Activated protein C-protein C inhibitor (APC-PCI) complex as a prognostic marker in sepsis

Lars Heslet<sup>1</sup>, Rikke Hald<sup>2</sup>, Camilla Recke<sup>2</sup>, Kristian Bangert<sup>2</sup> and Lars Otto Uttenthal<sup>2</sup>  
<sup>1</sup>ICU, Rigshospitalet, Copenhagen; <sup>2</sup>BioPorto Diagnostics A/S, Gentofte, Denmark

## Background

The PROWESS study and later trials of APC treatment in sepsis have shown only modest reductions in mortality. The recent Cochrane systematic review (CD004388) records doubtful efficacy and serious adverse effects. To optimize the benefit/risk ratio of APC treatment of each patient, a biomarker of protein C activation is urgently needed and the use of such a marker, APC-PCI, has been investigated in the present study.

## Methods

APC-PCI was measured in acid citrate plasma by means of a newly developed sandwich ELISA (median normal value 0.13 ng/mL, range 0.07-0.26, n = 16). The low pH of the plasma prevents *ex vivo* formation of further APC-PCI.

Levels of APC-PCI and protein C were monitored (daily to alternate days) in 135 consecutive critically ill patients, 53 of whom had sepsis during the observation period. Four patients were excluded because of incomplete data. The state of protein C activation to APC was categorized as non-activated, moderately activated or highly activated, based on maximum APC-PCI values in relation to the normal range.

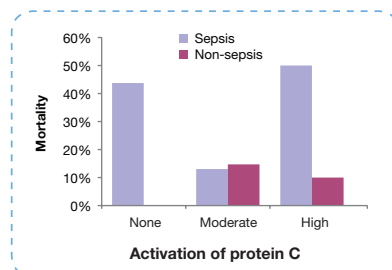
## Results

Maximum APC-PCI values ranged from 0.03 to 29 ng/mL (median 0.44 ng/mL).

Overall mortality of the sepsis patients was 32% (17/53).

There was no correlation between maximum APC-PCI and minimum protein C levels in individual patients (Spearman coefficient 0.02, P 0.89).

## Activation of protein C and mortality in ICU patients

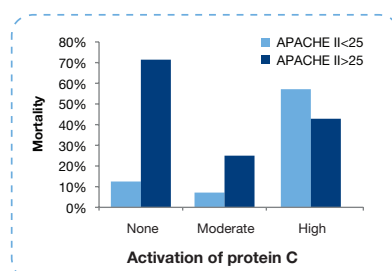


Patients with sepsis showed a bell-shaped mortality relationship with protein C activation, high mortality occurring in both the non-activated and highly activated groups. This relationship was not observed in the patients without sepsis, where no deaths occurred in the non-activated group.

Relative mortality (calculated separately for sepsis and non-sepsis patients) = (activation group mortality)/(overall mortality)

Protein C activation group	Non-activated APC-PCI <0.25 ng/mL		Moderately activated APC-PCI 0.25-0.72 ng/mL		Highly activated APC-PCI >0.72 ng/mL		P ( $\chi^2$ )
	Mortality	Relative mortality	Mortality	Relative mortality	Mortality	Relative mortality	
Sepsis	43.8% (7/16)	136.4%	13.0% (3/23)	40.7%	50.0% (7/14)	155.9%	0.032
Non-sepsis	0.0% (0/14)	0.0%	14.7% (5/34)	143.4%	10.0% (3/30)	97.5%	0.311

## Activation of protein C, APACHE II score and mortality in sepsis patients

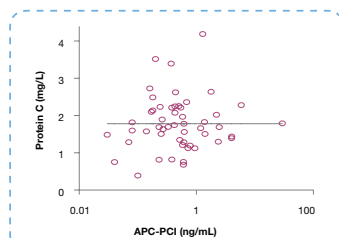


Patients with sepsis were subgrouped according to their APACHE II score (not available for 1 survivor and 1 non-survivor). Both absolute and relative mortalities were highest in the subgroup without protein C activation and APACHE II  $\geq 25$ .

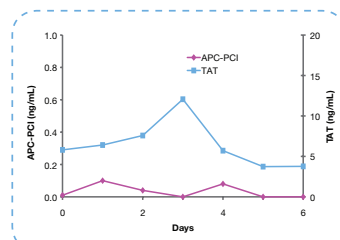
Relative mortality = (subgroup mortality)/(overall mortality for the sepsis patients)

Protein C activation group	Non-activated APC-PCI <0.25 ng/mL		Moderately activated APC-PCI 0.25-0.72 ng/mL		Highly activated APC-PCI >0.72 ng/mL		P ( $\chi^2$ )
	Mortality	Relative mortality	Mortality	Relative mortality	Mortality	Relative mortality	
APACHE II $\geq 25$	71.4% (5/7)	227.7%	25.0% (2/8)	79.7%	42.9% (3/7)	136.6%	0.1946
APACHE II < 25	12.5% (1/8)	39.8%	7.1% (1/14)	22.8%	57.1% (4/7)	208.2%	0.311

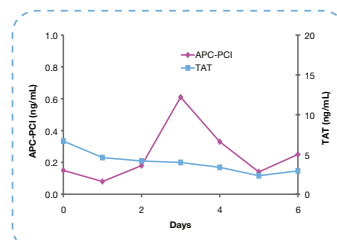
## APC-PCI and TAT levels in individual sepsis patients



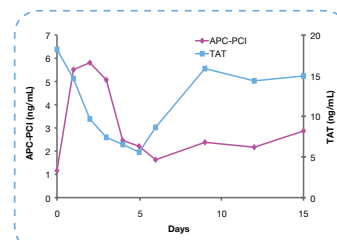
Scatter plot of minimum protein C values against maximum APC-PCI values



Low protein C activation  
APACHE II: 29  
Non-survivor



Moderate protein C activation  
APACHE II: 29  
Survivor



High protein C activation  
APACHE II: 26  
Non-survivor

## Conclusion

The relationship between mortality and protein C activation differs in critically ill patients with and without sepsis. In sepsis patients, failure of protein C activation and high protein C activation are both associated with high mortality. Sepsis patients without protein C activation and an APACHE II score  $\geq 25$  have a particularly high mortality. Patients in this subgroup may be those who are most likely to benefit from APC treatment.