



Aalborg Universitet

AALBORG UNIVERSITY
DENMARK

Biophysical Characterization of a dysfunctional antithrombin variant "Antithrombin III Aalborg

Pedersen, Shona; Thomsen, Marie; Kristensen, Søren Risom; Stensballe, Allan; Otzen, Daniel

Publication date:
2009

Document Version
Publisher's PDF, also known as Version of record

[Link to publication from Aalborg University](#)

Citation for published version (APA):

Pedersen, S., Thomsen, M., Kristensen, S. R., Stensballe, A., & Otzen, D. (2009). *Biophysical Characterization of a dysfunctional antithrombin variant "Antithrombin III Aalborg*. Poster presented at XXII Congress International Society on Thrombosis and Haemostasis, Boston, United States.

General rights

Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

- ? Users may download and print one copy of any publication from the public portal for the purpose of private study or research.
- ? You may not further distribute the material or use it for any profit-making activity or commercial gain
- ? You may freely distribute the URL identifying the publication in the public portal ?

Take down policy

If you believe that this document breaches copyright please contact us at vbn@aub.aau.dk providing details, and we will remove access to the work immediately and investigate your claim.

Characterization of a dysfunctional antithrombin variant “Antithrombin III Aalborg”

S Pedersen^{*}, M Thomsen[†], SR Kristensen^{*}, A Stenballe[†] and DE Otzen[†]

^{*}Department of Clinical Biochemistry, Cardiovascular Research Centre, Aalborg University Hospital, Aalborg, Denmark; and [†]Department of Life Sciences, Aalborg University, Aalborg, Denmark

Introduction. An AT variant denoted AT-III Aalborg has been identified in patients suffering from venous thromboembolic disease. The variant is a type II AT deficiency with normal antigen and decreased activity levels, but only when measured with an anti-IIa method whereas anti-Xa activity is normal. The aim of this study was to elucidate and characterize AT-III Aalborg through a biophysical and proteomic approach.

Method. To understand the cause of the dysfunction of this AT-variant several biophysical experiments and mass spectroscopic analyses were performed:

- Identification of the disease-related modification by mass spectrometry (MS) and DNA-diagnostics
- Estimation of the rate constants for inhibition of thrombin and factor Xa
- Estimation of the heparin binding affinity with Fluorescence spectroscopy
- Investigation of the conformational stability of AT-III Aalborg by circular dichroism
- Detection of Polymers by Dynamic light scattering.
- Conformation and structural analysis by electrophoresis.

Results. DNA-analysis and MS showed a serine to leucine conversion at position 394, i.e. in the reactive loop. MS did not indicate any other changes. The inhibition rate constant of AT-III Aalborg for IIa was 1/5th of the normal whereas for Xa it was half of the normal. Heparin binding affinity was normal. Biophysical investigations did not indicate a changed conformational stability

Conclusion. AT-III Aalborg has a change in the reactive loop which lowers the inhibition rate of IIa much more than expected. If the mutated AT had no effect at all, a rate constant of half the normal would be expected. This indicates that the mutated AT not only does not inactivate thrombin, but it inhibits the effect of the wt AT, further indicating that a type II AT deficiency may be more severe than a type I deficiency which may explain the severe phenotype of AT-III Aalborg.