

# Clot busting with FXIIa

## What is the problem?

- Annually, there are 410,000 cases of heart attacks and strokes in the UK.
- Blood clotting (haemostasis) is essential to prevent blood loss during injury, but when it becomes unbalanced it is undesirable; causing clots in the bloodstream that impede oxygen delivery to vital organs.
- A blood clot is composed of fibrin, platelets and red blood cells.
- After a clot forms it is broken down by a process called fibrinolysis.
- There is a need for new clot-busting drugs to help combat cardiovascular diseases.

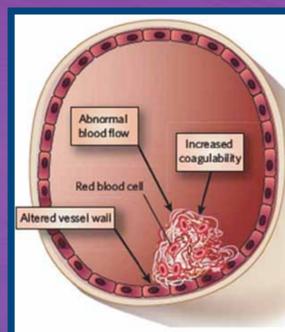


Figure 1 (left): A blood clot restricting normal blood flow (reproduced from: Mackman, N. (2008) Trigger, targets and treatments for thrombosis. *Nature*. 451, 914-918).

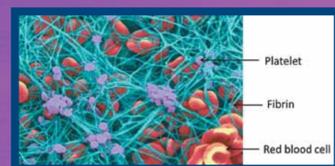


Figure 2 (above): Scanning Electron Microscopy image of a blood clot (available from: <http://www.med.upenn.edu/apps/faculty/index.php/g275/p20310> [Accessed 3rd September 2014])

## What are we interested in?

- Factor XII (FXII), an enzyme, was originally thought to promote clot formation but recent evidence suggests it also has a role in fibrinolysis
- Research in my supervisor's laboratory has found that polyphosphate (polyP), a peptide released by platelets, regulates FXII
- FXII is activated by polyP to become the enzyme FXIIa
- We set out to clarify whether FXIIa and polyP have a function in dissolving undesirable clots

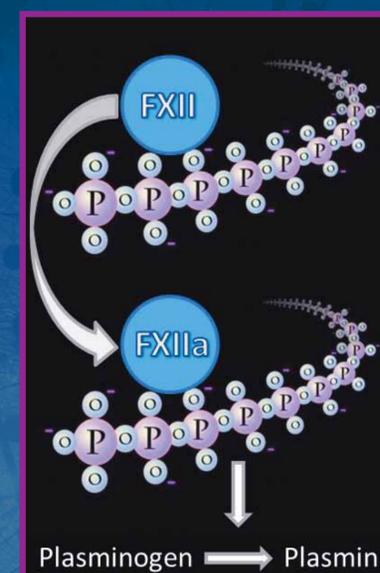


Figure 3: Involvement of FXII in Fibrinolysis

## What did we do?

The research began with a series of clot lysis (clot-dissolving) experiments which compare the efficiency of different concentrations of FXIIa and polyP to lyse clots. We also tested the ability of FXIIa and plasminogen to bind to polymethacrylate beads labelled with polyP. A method called Western Blotting was used to determine this. This involves sorting of proteins according to their molecular weight then using a specific antibody to detect the protein of interest.

## What did we find?

FXIIa alone was very inefficient at dissolving clots but their breakdown was dramatically enhanced by polyP (see Figure 4). This suggests that polyP improves the ability of FXIIa to dissolve clots.

Western blots showed that both FXIIa and plasminogen bound to polymethacrylate beads labelled with polyP (see Figure 5). Some protein could be detected in the flow-through (FT) fraction but some remained on the beads. Low salt (LS) washes did not remove the protein, but high salt (HS) washes disrupted the interaction. This suggests that the interaction of polyP with FXIIa and plasminogen is electrostatic.

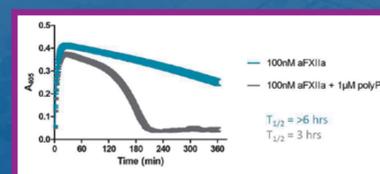


Figure 4



Figure 5

## What does this mean?

These data suggest that polyP acts as a bridge between FXIIa and plasminogen allowing it to become activated into plasmin which dissolves clots. This interaction could help direct new clot-busting treatments.

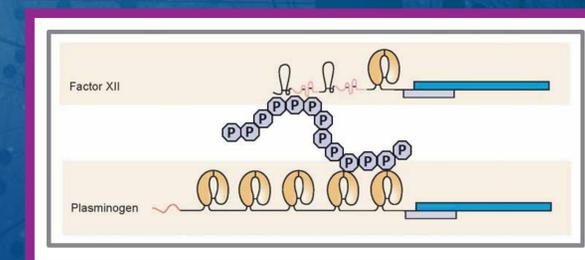


Figure 6: polyP as a bridge (reproduced from: Mutch NJ, Booth NA. Plasminogen activation & regulation of fibrinolysis (Chapter 20; p314-334); In *Haemostasis & Thrombosis. Basic Principles and Clinical Practice*, Sixth edition, 2012)

## Who am I?

I am in my 4th year in Biomedical Science (Physiology) at the University of Aberdeen. Afterwards, I am interested in pursuing a PhD.

## Acknowledgements

I would like to thank Medical Research Scotland for their funding and Dr Nicola J Mutch and Austra Lionikiene for their continuous training and support throughout my studentship.