Introduction:
Clinicians in the emergency setting use a wide range of haemostatic markers to diagnose and monitor disease and treatment. Current methods rely on the anticoagulant effect of citrate on whole blood prior to laboratory analysis. Despite the well-recognised modulatory effects of citrate on haemostasis, the use of anticoagulated blood has clear analytical advantages, including repeat sampling and storage. However by altering the physiological state of the blood reproducibility and accuracy of the test is affected. Recent studies have shown the potential of a novel functional biomarker of clot formation: Fractal Dimension ($d_f$), that may give an improved diagnostic accuracy. In this study we assessed the potential of this new biomarker in scientifically measuring the effects of recalcification of citrated samples.

Methods:
35 healthy volunteers were included. Unadulterated and sodium citrate samples of blood were taken from each volunteer. Citrated samples were recalcified using (1M CaCl$_2$). In the study we compared unadulterated whole blood $d_f$ results to citrated $d_f$ results and repeated the citrated $d_f$ experiments 5 times for each sample over a 2 hour period to ascertain reproducibility.

Results:
The $d_f$ of citrated blood was significantly lower than that of unadulterated blood (1.57±0.04 vs 1.69±0.04, p<0.001). The results of the citrate samples when tested 5 times over 2 hrs gave a Coefficient of Variation of 1.16%.

Conclusion:
For the first time we show that a functional biomarker of clot microstructure, $d_f$, can precisely quantify and measure accurately the direct effect that the addition of the anticoagulant sodium citrate has on whole blood clot microstructure. The study also shows that the test is reproducible and has potential utility as a biomarker of acute disease in the emergency setting in citrated blood. This procedure now needs to be evaluated in a group of acute disease states.