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Production and Functional Analysis of *Echis coloratus* Disintegrins in Platelet Aggregation

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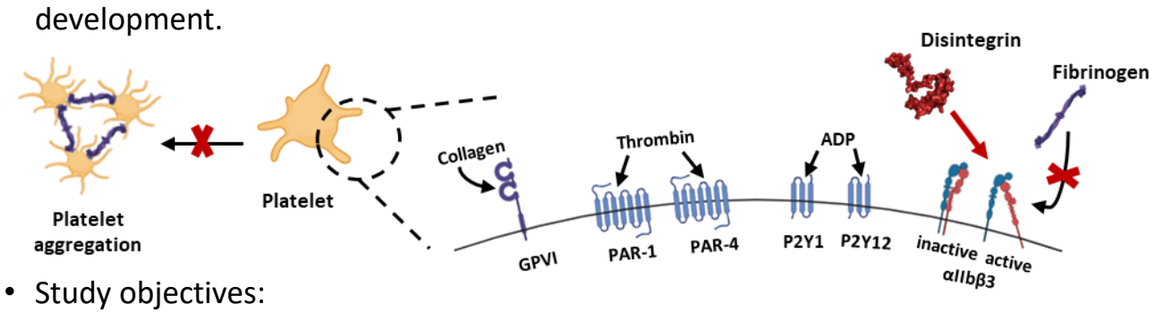
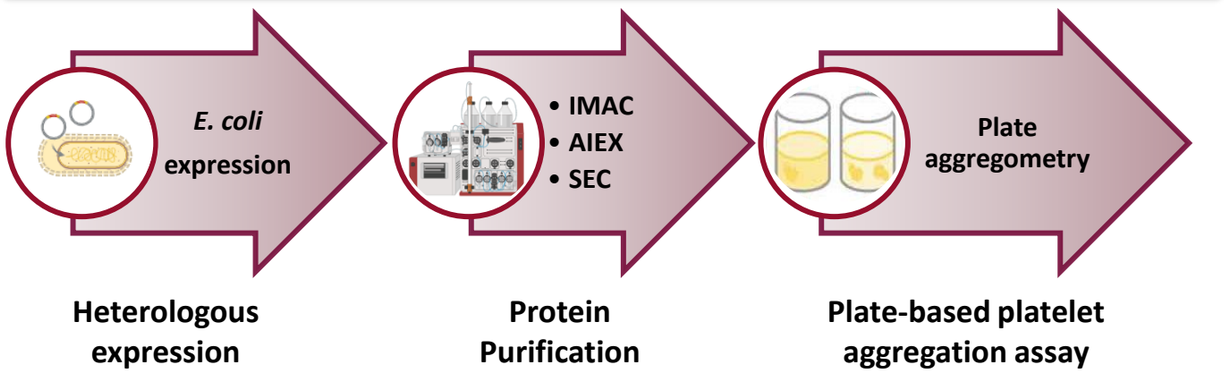
INTRODUCTION & AIM

- Snake venom disintegrins are small, non-enzymatic proteins that bind integrins with high specificity.
- *Echis coloratus* venom gland transcriptome includes a disintegrin cluster comprising ~5% of toxin transcripts.
- Disintegrins are valuable templates for antiplatelet and antithrombotic drug development.



- Study objectives:
  - ➢ produce recombinant disintegrins from *E. coloratus* (EcDis_VGD, EcDis_KGD, EcDis_RGD);
  - ➢ evaluate their effects on platelet aggregation.

METHOD



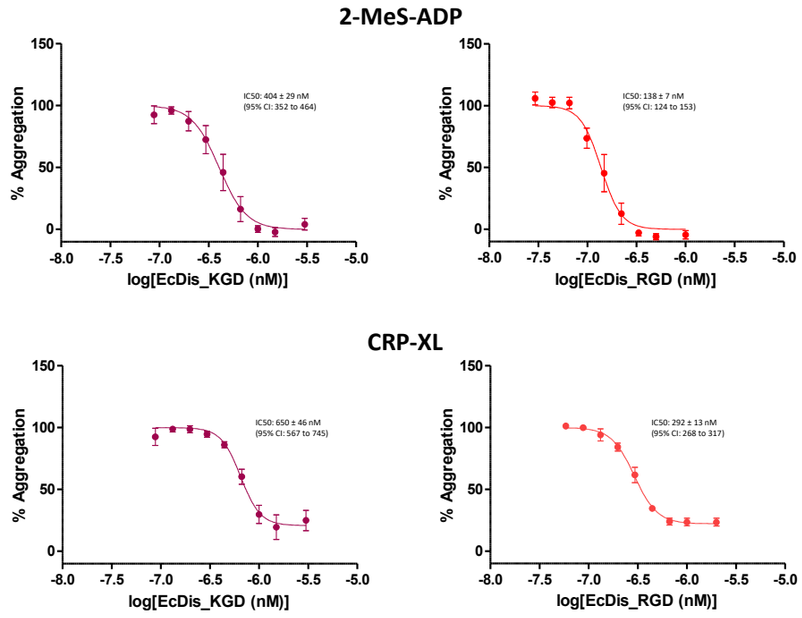


Figure 3. Dose-response curves for IC50 determination of EcDis_KGD and EcDis_RGD against 2-MeS-ADP- and CRP-XL-induced platelet aggregation. EcDis_KGD is shown in magenta, and EcDis_RGD is shown in red. Data are presented as the mean of % aggregation ± SEM versus disintegrin concentration.

RESULTS & DISCUSSION

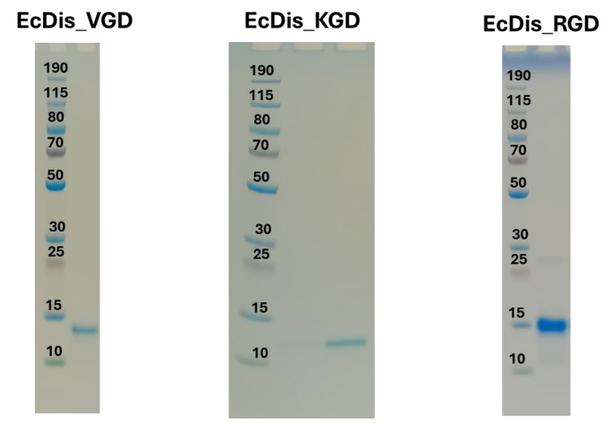


Figure 1. Reducing SDS-PAGE analysis of purified EcDis_VGD, EcDis_KGD, and EcDis_RGD following size-exclusion chromatography (SEC), demonstrating the purity and estimated molecular weights of each protein.

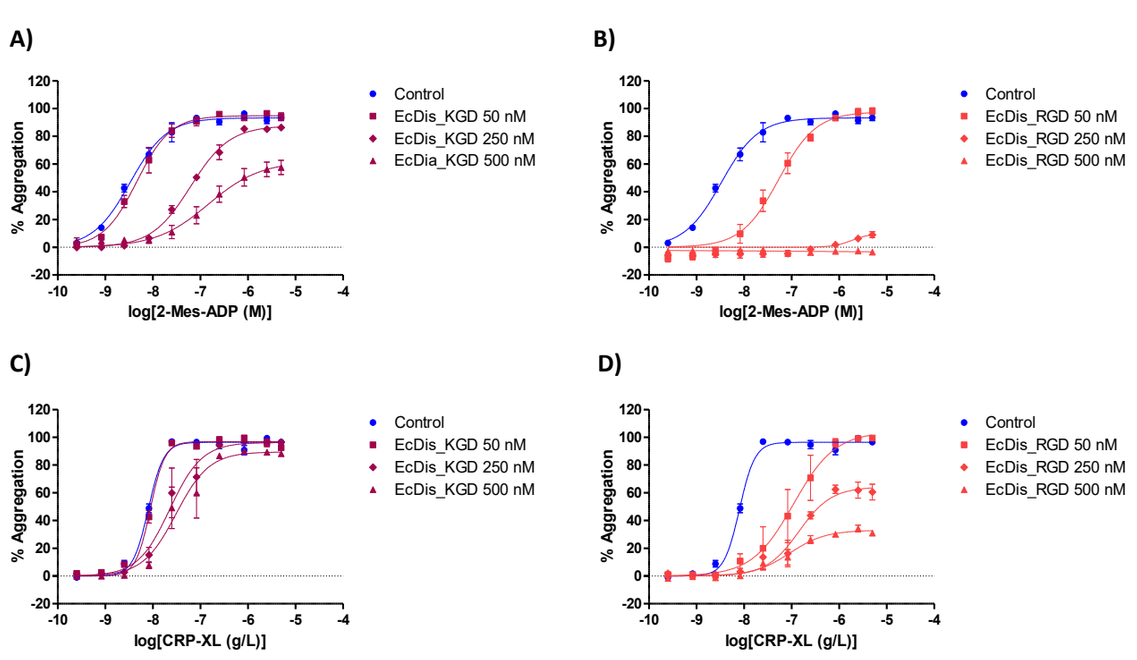


Figure 2. EcDis_KGD and EcDis_RGD inhibit 2-MeS-ADP- and CRP-XL-induced platelet aggregation. **A)** and **C)** EcDis_KGD (magenta curves) and **B)** and **D)** EcDis_RGD (red curves). Data are presented as the mean of % aggregation ± SEM versus agonist concentration.

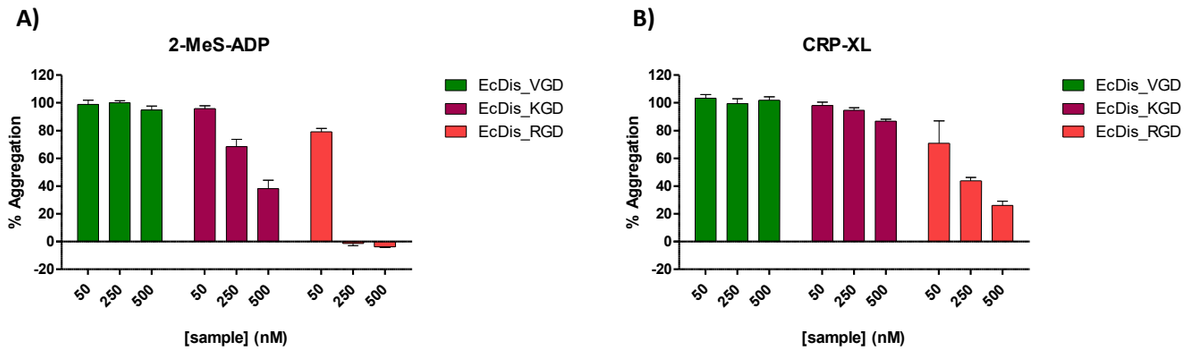


Figure 4. Effect of disintegrins on 2-MeS-ADP- and CRP-XL-induced platelet aggregation. The bar graph shows the percentage of aggregation at **A)** 2.5 µM 2-MeS-ADP and **B)** 2.5 µg/mL CRP-XL. EcDis_VGD is shown in green, EcDis_KGD in magenta and EcDis_RGD in red. Data are presented as the mean of % aggregation ± SEM.

CONCLUSION

- Recombinant *E. coloratus* disintegrins were successfully expressed and purified.
- Functional studies revealed that EcDis_RGD is the most potent inhibitor of platelet aggregation, followed by EcDis_KGD, while EcDis_VGD was inactive.
- Our work highlights the functional diversity of *E. coloratus* disintegrins and supports their exploration as leads for novel therapeutics.

ACKNOWLEDGMENTS

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