ABSTRACT

SARS-CoV-2 is the virus responsible for a global pandemic due to its infectivity and severe symptoms. In addition to the typical respiratory symptoms, infection with COVID-19 has been shown to promote microclots within the blood vessels, and these clots are predicted to contribute to a number of the uniquely COVID-associated symptoms. Our formation is a natural response to cellular damage within the vascular system and is facilitated by the release of a series of fibrin-like structures known as Von Willebrand Factor (VWF). To prevent unnecessary clots from forming during vascular inflammation, an enzyme called ADAMTS13 prevents the accumulation of VWF, in the absence of arraics cell damage.

Objectives: The goal of our research is to determine if the SARS-CoV-2 3c protease, used in viral replication, promotes clotting during infection by targeting ADAMTS13.

Methods: The amino acid sequence of human ADAMTS13 was evaluated for potential cut sites using published cleavage data from the SARS 3c protease. Western blotting was used to directly evaluate interactions between ADAMTS13 and purified COVID-19 3c protease.

Results: In silico data identified 3 potential 3c protease cleavage sites in the ADAMTS13 amino acid sequence. Western blotting confirmed cleavage of ADAMTS13 in the presence of the SARS-CoV-2 3c protease.

Conclusions: Our current data demonstrate that a viral protein capable of cleaving ADAMTS13 may be responsible for the reduced levels of ADAMTS13 noted in COVID-19 patients that is linked to increased clot formation during infection. Additionally, this interaction may provide insights into the rebound symptoms observed with the anti-COVID drug Pasvold, a 3c protease inhibitor, and may provide clues to the nature of symptoms associated with long COVID.

KEY POINTS

- SARS-CoV-2 is a respiratory virus.
- SARS-CoV-2 is associated with the formation of blood clots.
- Microtubules are associated with COVID toes.
- Microtubules are associated with strokes, cardiovascular, and respiratory issues.
- Blood clot formation is associated with the Von Willebrand Factor (VWF).
- VWF is a blood glycoprotein.
- VWF is released during vascular inflammation.
- VWF expression levels are controlled by ADAMTS13.
- ADAMTS13 is a VWF-cleaving protease responsible for regulating blood clotting.
- Loss of ADAMTS13 leads to increased clot formation.
- Observed in Thrombotic Thrombocytopenic Purpura which is a rare disorder causing blood clots to form in blood vessels.
- Imbalance of platelet levels could lead to coagulation.
- Early investigations indicated that the severity of SARS-CoV-2 infection had an association with blood type.
- VWF is a blood protein that has blood group A antigens.

REFERENCES


ACKNOWLEDGEMENTS

We would like to thank the East Carolina University Department of Biology.

CONCLUSIONS

- Clots are strongly associated with the symptoms of SARS-CoV-2.
- SARS-CoV-2 3c protease cleaves ADAMTS13.
- Loss of ADAMTS13 results in increased clot formation.
- This is the first evidence of any COVID-19 protein having a direct interaction resulting in clot formation.
- These findings may provide insights into long COVID as well as the rebound symptoms associated with antiviral drug Pasvold.
- Long COVID may result from an immune response to the fragmented ADAMTS13.
- Pasvold is a 3c protease inhibitor. If the protease remains active and is responsible for some symptoms, symptoms may return when the inhibitors are removed.

FUTURE DIRECTIONS

- Identify the exact cleavage sequence for the viral 3c-protease.
- Demonstrate that this cleavage activity occurs in vivo.
- Determine how long the viral 3c protease persists after infection.
- Determine the recovery time of ADAMTS13 levels after depletion.
- Determine if autoantibodies are reacting to the ADAMTS-13 fragments.

Figure 1: Model of Von Willebrand Factors interaction blood platelets.
- Von Willebrand Factor is a fibrous protein that collects red blood platelets.
- VWF collects and aggregates red blood cell platelets.
- Hypercoagulation can begin to result within the blood.

Figure 2: Model of ADAMTS-13 cleaving and regulating Von Willebrand Factors in blood vessels.
- ADAMTS-13 is a disintegrin protein that regulates VWF by cleaving VWF multimers.
- Regulation of Ultra Von Willebrand Factors (ULVWF) breaks apart aggregates of red blood platelets and VWF lowering chances of embolism and thrombotic risk.

Figure 3: Model of VWF and ADAMTS-13 homeostasis and different types of patients.
- Part A: Model of ADAMTS-13/VWF homeostasis levels in normal individuals. There is a homeostatic balance for a normal healthy individual.
- Part B: Model of ADAMTS-13/VWF homeostasis of patients with Von Willebrand Disease (VWD). Excess amounts of ADAMTS-13 leads to excessive bleeding due to inability of VWF to create clots.
- Part C: Model of ADAMTS-13/VWF homeostasis of COVID-19 patients. Excess amounts of VWF leads to micro-clotting. Low levels of ADAMTS-13 lead to unregulated VWF leading to thrombotic risks.
- Part D: Model of ADAMTS-13/VWF homeostasis of patients with Thrombotic Thrombocytopenic Purpura. Excess amounts of VWF leads to excessive clotting due to low amounts of ADAMTS-13 to regulate VWF.