Elucidating the pathophysiology of intracranial hemorrhage in a developing vertebrate

Department of Biology

Ali Taha, Dr. Thomas W. Moon, Shahram Eisa-Beygi

Introduction

Intracranial hemorrhage is a debilitating form of stroke, often associated with mortality or irreversible neurological deficits. It is the second most common form of stroke, and there are no known cures. Nonetheless, the entire suite of molecular and physiological events in the brain during cerebral hemorrhage is still unexplored. Zebrafish (*Danio rerio*) have gained immense reputation as ideal organisms to elucidate the etiology and characterization of human diseases. By using a previously established model of spontaneous hemorrhage in developing zebrafish, the ensuing impacts of vessel rupture can be thoroughly examined. The biological activity in the brain after cerebral hemorrhage can be monitored using a variety of techniques, mainly an array of immunohistochemical stains. Present studies are geared towards assessment of the state of blood vessels, apoptosis in the brain, and proliferation of immune cells.

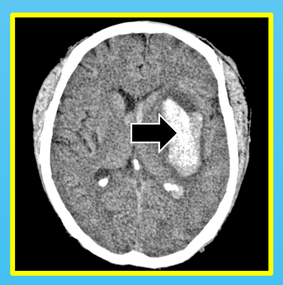

Figure 1: CT scan of a human brain showing intracranial hemorrhage

Methodology

Intracranial hemorrhage is induced by exposing the zebrafish embryos to 0.5 mg/L atorvastatin (ATV), a pharmacological inhibitor of the mevalonate pathway that works through inhibition of HMG CoA reductase enzyme. This consequently hinders endogenous cholesterol production, as well as the synthesis of compounds targeted to membranes, including those in blood vessels. Comparing the consequences of endogenous cholesterol inhibition in zebrafish exposed to 0.5 mg/L ATV to a control group not exposed to ATV will give a comparative library of the physiology of the brain post-cerebral hemorrhage

Results

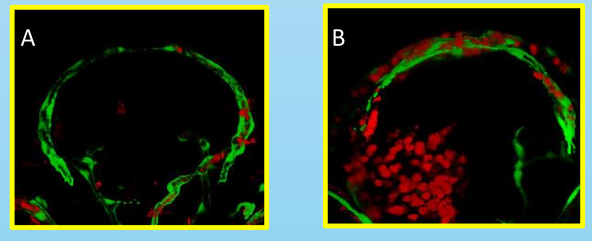

Figure 2: Hemorrhage (Gata1:dsRed) and vessel rupture (fli:eGFP) in the brain of zebrafish embryos 2 dpf in the control group (A) and those exposed to ATV (B).

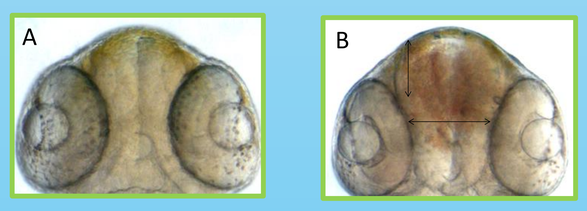

Figure 3: Volume of the brain in of zebrafish embryos 2 dpf in the control group (A) and those exposed to ATV (B).

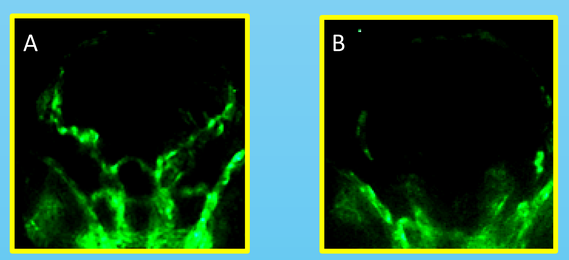

Figure 4: Blood vessel organization (fli:eGFP) in the brain of zebrafish embryos 2 dpf in the control group (A) and those exposed to ATV (B).

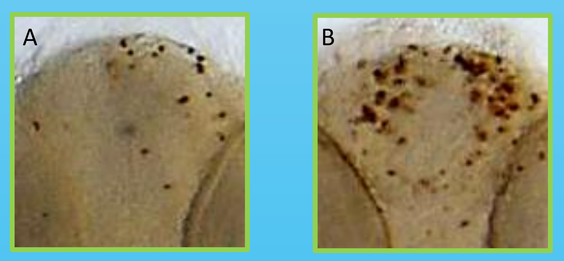

Figure 5: Apoptosis (TUNEL) in the brain of zebrafish embryos 2 dpf in the control group (A) and those exposed to ATV (B).

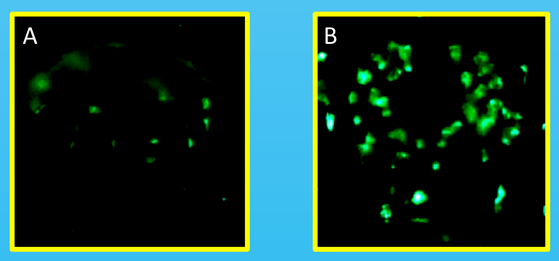

Figure 6: Immune cells (coro1α: eGFP) in the brain of zebrafish embryos 2 dpf in the control group (A) and those exposed to ATV (B).

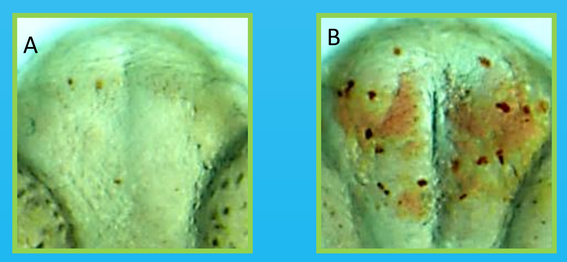

Figure 7: Macrophage recruitment (Neutral red) in the brain of zebrafish embryos 2 dpf in the control group (A) and those exposed to ATV (B).

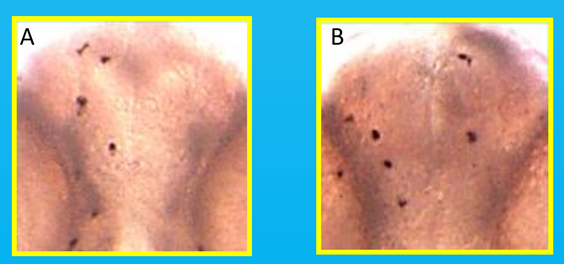

Figure 8: Neutrophils (Sudan black) in the brain of zebrafish embryos 2 dpf in the control group (A) and those exposed to ATV (B).

Conclusion

Zebrafish that experienced intracranial hemorrhage showed the following characteristics compared to the control group:

- Increased hemorrhage and vessel rupture.
- Degeneration of the organization of blood vessels in the brain.
- Increased apoptosis, which can be attributed to the cells composing the vessel walls.
- Increase in the volume of the brain.
- Increase in immune cell proliferation.
    - Macrophages are recruited in a large number.
    - Neutrophil levels are relatively equal in the ATV group and control group.

Future research

Ongoing research seeks to study the recovery period associated with intracranial hemorrhage in zebrafish embryos. This is done by comparing the intracranial physiology of 2 dph zebrafish exposed to ATV to 3 dpf zebrafish also exposed to ATV. By looking at the same physiology (vessel rupture, hemorrhage, immune cells, etc.) conclusions can be made concerning the progress of zebrafish embryos in recovery. Ultimately, the goal will be to employ the understanding of this library of physiology in designing therapies to treat intracranial hemorrhage.

References

Skrifvars, M. B., & Parr, M. J. (2012). Incidence, predisposing factors, management and survival following cardiac arrest due to subarachnoid haemorrhage: a review of the literature. *Scandinavian Journal of Trauma, Resuscitation and Emergency Medicine*, *20*, 75.

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