Evaluating the effectiveness of a baicalin treatment for inhibition of the cGAS synapse in lowering inflammatory rates of drosophila melanogaster with Huntington's Disease

Academies of Sciences, Academies of Loudoun

AOS Senior Scientific Research

Mr. Andrew Allshouse

Pranav Vishnubhatla

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The National Center for Health Statistics (NCHS) stated that every year, roughly 2.2 million people die from effects related to Huntington's disease (Lanska et al., 1988). Huntington's disease (HD) is a hereditary neurodegenerative disorder that affects cognitive and locomotive parts of the brain, such as the cerebellum and basal ganglia. HD leads to the gradual degeneration and death of neurons within the prefrontal cortex and the peripheral (PNS) and central nervous systems (CNS), leading to many observed cognitive disordersg. (National Institute of Health, 2023). Though HD is seen in all ages, it is most prevalent in the 30- to 40-year-old age group. HD is formed by a mutation in the gene that contains the coding for the huntingtin gene. The huntingtin gene, also known as the HTT gene, is expressed in every single person. The HTT mutation leads to the three main pillars of DNA, cytosine, adenine, and guanine (CAG), repeating at an extremely rapid pace as compared to CAG repeats without the HTT mutation (Faquih et al., 2023). Patients with around 36 or more CAG repeats are in the target zone for developing HD. To put this into perspective, healthy individuals have no more than 26 CAG repeats. HD is passed on from offspring through just a single gene variation that leads to mass CAG repeats.The genetic mutation responsible for HD is inherited in an autosomal dominant pattern, which means that an affected person has a 50% chance of passing this mutation down to their offspring.

One major factor that contributes to the severity of HD is the Cyclic GMP-AMP Synthase (cGAS Synthase). The cGAS Synthase is the cellular response that triggers the production of various molecules. In this case, the cGAS Synthase is used in a certain way. HD starts with an htt mutation. The htt mutation within the body causes abnormal DNA creation that is then detected by an enzyme called the cGAS enzyme. Once detection occurs, the cGAS enzyme is activated and produces Cyclic Guanosine Monophosphate-Adenosine Monophosphate (cGAMP) (Bai et

al., 2019). Once the cGAMP molecule is produced, it reacts with the STING receptor. The STING receptor is located within the endoplasmic reticulum (ER) and is used for pathogen defense. Once the STING receptor is activated, an inflammatory response will be released where harmful interferons will enter the brain and lead to neural inflammation and degradation. With HD, these effects are more severe and cause further damage on neural health (Ferecsko et al., 2023).

Baicalin, is a flavonoid compound that has been used for over many centuries for its medicinal benefits. First discovered in Chinese medicine, it has evolved into a major compound that is known to have medicinal benefits. It is often found in plants such as *Scutellaria baicalensis* (Chinese skullcap). Baicalin has been the subject of many research papers and some findings have indicated that baicalin can be used as an efficient treatment method for various neurodegenerative diseases. Along with this, other research studies have shown that baicalin can provide low level treatment for other diseases as well such as COVID-19 and SARS (NIH, n.d.).

Currently, though there is a large amount of research within the HD field, no cure has been found. Along with this, there is little research which studies the possibilities that can come from targeting the cGAS synthase. One study though, targets both these fields. A research conducted by Jiameng Li et al. in August 2023, worked to study the effect baicalin had in decreasing cGAS - STING pathway's activity. By using a pyrophosphatase (PPiase) coupling assay, scientists were able to conclude that baicalin and other compounds were able to significantly reduce cGAS enzymes within the cell. (Li et al., 2023).

Drosophila melanogaster are fruit flies that have been part of scientific research for many years now. Their short lifespan and easy maintainability make the drosophila the model organism

for research. Along with this, drosophila have many different neuronic and genetic similarities to human beings (Jeibmann et al., 2009). For example, the drosophila brain is very similar to the human one. Another advantage of using Drosophila to study HD is that their genetic structure allows for simple gene modification. This genetic toolkit allows UAS, GAL-4, and other things to be easily added to drosophila.

Homogenization is the process of breaking down cellular tissue of organisms to isolate important cellular DNA of these organisms. Centrifugation is the process of separating a mixture created by homogenization into soluble and insoluble molecules. Centrifugation is vital for research as it helps researchers get access to the specific molecules and specific tissues that they need from an organism. In this study, the goal is to homogenize drosophila and, through centrifugation, isolate the cGAMP molecule. The cGAMP molecule is released when the cGAS synthase is triggered, and these cGAMP molecule levels can be detected within an organism, in this case, drosophila. With the use of an assay called the ELISA quantification assay (a specified assay that evaluates the content of certain molecules within a solution), the cGAMP values can be determined to understand whether or not the treatment method is inhibiting inflammation within drosophilae with Huntington's.

This research plays a role in identifying a possible treatment for HD and opens the pathway for research that can utilize this flavonoid in HD treatment. The goal of this research is to evaluate the effectiveness of a Baicalin-based food treatment for drosophilae that suffer from early stage Huntington's disease. Once drosophilae are treated with baicalin, through the process of centrifugation and homogenization, samples will be collected and they will be studied with the ELISA assay. The cGAMP levels will then be evaluated in order to determine whether or not the treatment works in lowering HD within drosophilae.

Resources

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