

12-2014

Altered Cell-surface Receptor Levels Result from Fructose Advanced Glycation End Product-Induced Inflammation

Matthew Stryker

St. John Fisher College, mds05927@students.sjfc.edu

Regina Blackley

St. John Fisher College, rcb00745@students.sjfc.edu

Melinda E. Lull

St. John Fisher College, mlull@sjfc.edu

[How has open access to Fisher Digital Publications benefited you?](#)

Follow this and additional works at: http://fisherpub.sjfc.edu/doctoral_ext_pub

 Part of the [Pharmacy and Pharmaceutical Sciences Commons](#)

Publication Information

Stryker, Matthew; Blackley, Regina; and Lull, Melinda E., "Altered Cell-surface Receptor Levels Result from Fructose Advanced Glycation End Product-Induced Inflammation" (2014). *Doctoral External Publications*. Paper 3.

http://fisherpub.sjfc.edu/doctoral_ext_pub/3

Please note that the Publication Information provides general citation information and may not be appropriate for your discipline. To receive help in creating a citation based on your discipline, please visit <http://libguides.sjfc.edu/citations>.

Altered Cell-surface Receptor Levels Result from Fructose Advanced Glycation End Product-Induced Inflammation

Abstract

Objective: As a result of the heightened reactivity fructose demonstrates compared to glucose and our current knowledge of glucose advanced glycation end-products, the aim of this research was to further elucidate the proinflammatory pathways involved in the response to fru-AGE exposure, including the effects of fru-AGEs on cell-surface receptor expression. We hypothesized that once microglia were activated in response to fru-AGE exposure, there would be an increase in the expression of RAGE and TLR4 to facilitate the proinflammatory cascade.

Disciplines

Pharmacy and Pharmaceutical Sciences

Comments

Poster presented at at American Society of Health Systems Pharmacy Midyear Clinical Meeting in Anaheim, California, in December 2014.

Introduction

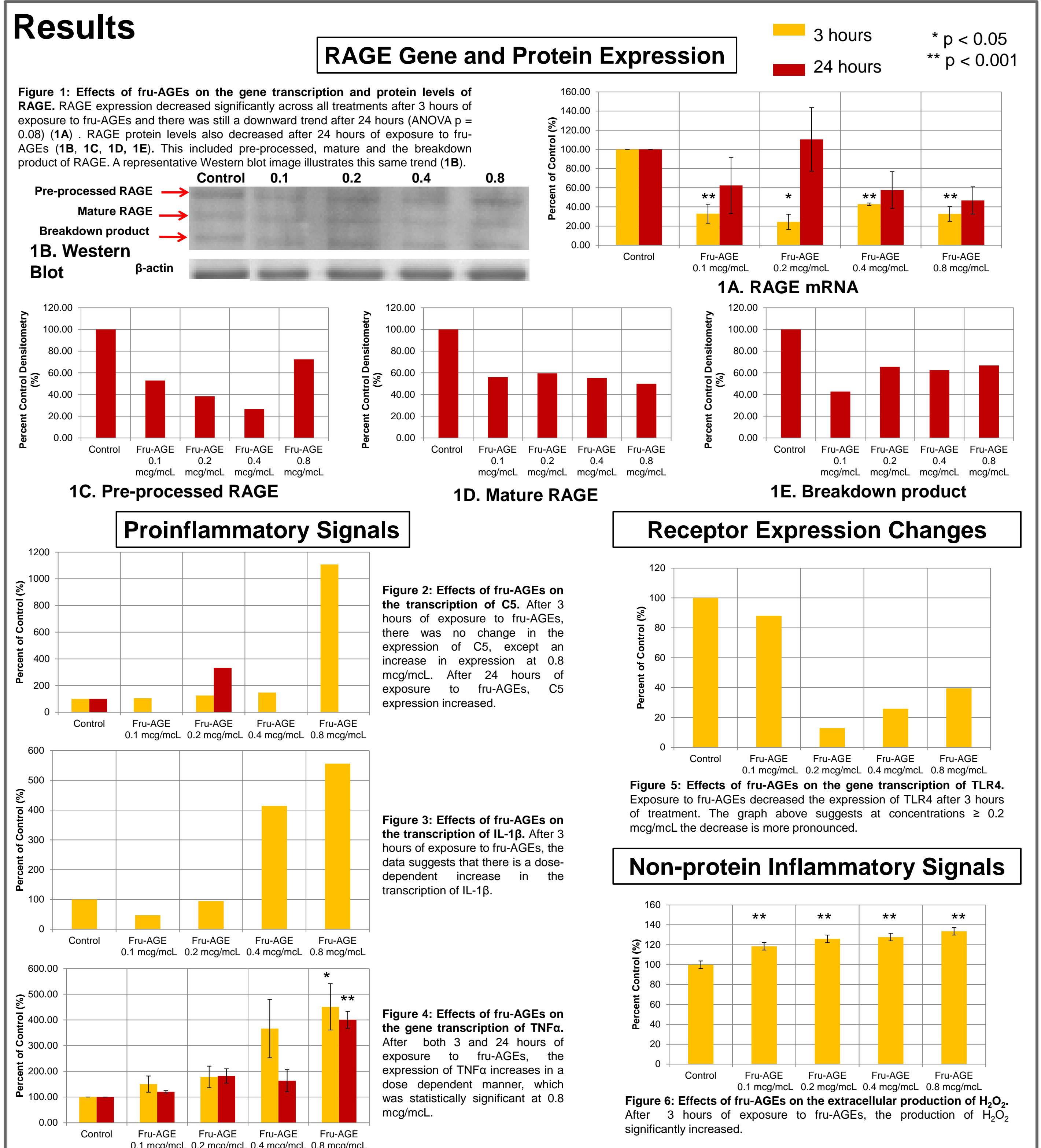
Fructose, a naturally occurring simple sugar, is also an industrial sweetener that has become a mainstay in the American diet, notably in sugar drinks.¹ A consequence of excess fructose consumption is the non-enzymatic generation of fructose advanced glycation end-products (fru-AGE[s]). The non-enzymatic glycation with fructose is ten times greater compared to glucose.² Within the central nervous system, microglia, residential macrophages surveying their microenvironment, become activated when exposed to fru-AGEs. The activation of microglia initiates a cascade of proinflammatory processes. Some of the proinflammatory mediators released include: IL-1 β , toll-like receptor 4 (TLR4), tumor necrosis factor α (TNF α) and reactive oxygen species (e.g., H₂O₂). Current evidence suggests that activation of microglia is mediated through the receptor for advanced glycation end-products (RAGE). Although AGE formation is a natural process, the excess exposure to AGEs has been implicated in the pathology of certain diseases, including: diabetic microvascular complications³ (e.g., retinopathy, nephropathy) and age-related diseases, such as Alzheimer's disease.

Objective

As a result of the heightened reactivity fructose demonstrates compared to glucose and our current knowledge of glucose advanced glycation end-products, the aim of this research was to further elucidate the proinflammatory pathways involved in the response to fru-AGE exposure, including the effects of fru-AGEs on cell-surface receptor expression. We hypothesized that once microglia were activated in response to fru-AGE exposure, there would be an increase in the expression of RAGE and TLR4 to facilitate the proinflammatory cascade.

Methods

Fru-AGEs were generated by incubating fructose with bovine serum albumin at 37°C for 8 weeks.⁴ An immortalized rat microglial cell line (HAPI) was treated with control media or fru-AGEs *in vitro* for 3 or 24 hours in the following concentrations: 0.1, 0.2, 0.4 or 0.8 mcg/mL. Gene expression of pro-inflammatory markers and cell-surface receptors was measured using quantitative real-time polymerase chain reaction (qRT-PCR). Genes investigated included the following: complement 5 (C5), interleukin-1 β (IL-1 β), tumor necrosis factor α (TNF α), receptor for AGE (RAGE) and toll-like receptor 4 (TLR4). Levels of RAGE protein were measured after 24 hours of exposure to fru-AGEs via Western Blot.⁵ To further characterize the proinflammatory response, extracellular hydrogen peroxide (H₂O₂) was quantified using the ROS-Glo H₂O₂ chemiluminescent assay (Promega) following a 3 hour incubation with the fru-AGE treatments listed above. Statistically significant changes for each measure were determined using a one-way ANOVA with a Tukey post-hoc test; a p-value < 0.05 was considered significant. All data are expressed as the mean \pm SEM.



Conclusions

- This evidence suggests that exposure to fru-AGEs increases the expression of known mediators of inflammation, including the following: C5, IL-1 β , TNF α (statistically significant, dose-dependent acute and chronic effects) and extracellular H₂O₂ (statistically significant dose-dependent effects)
- Cell-surface receptors including RAGE and TLR4 also demonstrate changes in their gene expression when exposed to fru-AGEs. RAGE expression was acute and chronically suppressed after exposure to fru-AGEs. After 3 hours, TLR4 was also suppressed but more replicates are needed.
- Analysis of protein levels support the gene expression trend observed with RAGE. Chronic exposure to fru-AGEs leads to a diminished level of mature RAGE protein.
- The decrease in receptor expression observed with both RAGE and TLR suggests that microglia can become desensitized to noxious stimuli. This observation is concerning because immune cells need to remain sensitized to noxious stimuli to help protect and defend the host.
- This research continues to illustrate and support the hypothesis that excess exposure to fru-AGEs induces an inflammatory state mediated by microglia within the brain and alters cell-surface receptors.
- In addition to their neurodegenerative effects in disease states such as Alzheimer's disease, further investigation about the endogenous effects of fru-AGEs should be considered, especially as a result of its accessibility from its use as an industrial sweetener.
- Further areas of research include additional replicates of C5, IL-1 β and TLR gene expression and extracellular H₂O₂ production to further validate this hypothesis.

References

- Consumption of Sugar Drinks in the United States, 2005 – 2008. Centers for Disease Control and Prevention. Available at: <http://www.cdc.gov/nchs/data/databriefs/db71.htm#summary>. Accessed Nov 7 2014.
- Suarez G, Rajaram R, Oronsky AL. Nonenzymatic glycation of bovine serum albumin by fructose (fructation). Comparison with the Maillard reaction initiated by glucose. J Biol Chem. 1989 Mar 5;264(7):3674-9.
- Goldin A, Beckman JA, Schmidt AM, et al. Advanced glycation end products: sparking the development of diabetic vascular injury. Circulation. 2006;114(6):597-605.
- Chandler D, Woldu A, Rahmadi A, et al. Effects of plant-derived polyphenols on TNF α and nitric oxide production induced by advanced glycation endproducts. Molecular Nutritional Food Research. 2010;54:141-50.
- Lull ME, Levesque S, Block ML. Chronic apocynin treatment attenuates beta-amyloid plaque deposition and microglia in hAPP_{SL} TG mice. In revision for PLOS One, March 2011.

Disclosure

The authors of this presentation have no personal or financial interest in the subject matter of this presentation.

Matthew Stryker: Nothing to disclose
Regina Blackley: Nothing to disclose
Melinda Lull: Nothing to disclose