High Fat and Sugar Diet Induces Gut Leukocyte Disruptions Exacerbated by SARS-CoV-2

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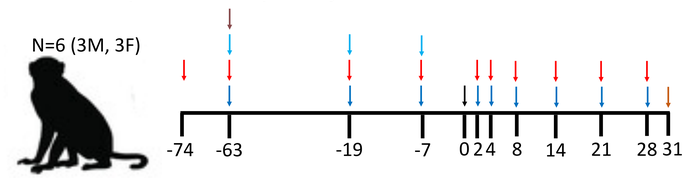
BACKGROUND

COVID-19 morbidity and mortality have not been uniform across global regions, with developed nations in Europe and North America experiencing particularly high numbers of infections and fatalities per capita. As SARS-CoV-2 infections can cause gastrointestinal damage that contributes to pathology, dietary impacts on gut susceptibility to infection have not been thoroughly explored. We investigate the impact of a Western-like diet that is high in fat and sugar on gut tissue and explore outcomes of infection in a non-human primate model.

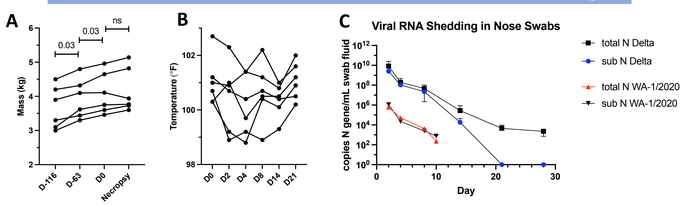
METHODS

- Pigtail macaques were fed a standard monkey chow, then a high fat and sugar diet prior to SARS-CoV-2 infection.
- Sampling occurred according to the schedule below in conjunction with measurements of clinical parameters such as mass, temperature, respiratory rate, etc.



RESULTS

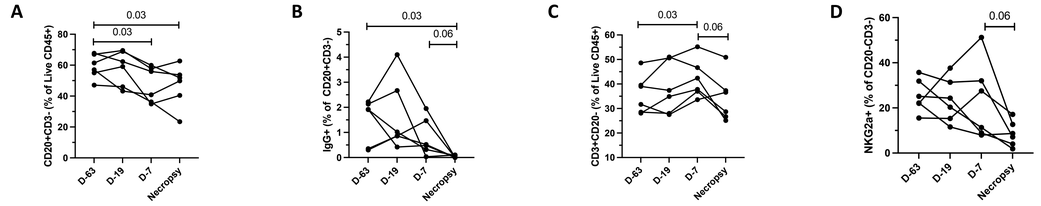
Mild SARS-CoV-2 Infection with Viral Shedding



Animals experienced significant mass increase from prior to high fat and sugar diet (HFD) initiation to HFD start and from HFD start to infection (**A**, Wilcoxon matched-pairs tests). SARS-CoV-2 infection was mild, with animals not having lost significant mass at necropsy (**A**) or experienced fever (temperature >103°F, **B**). However, mean total viral RNA and subgenomic RNA indicating viral replication shed in nose swabs was approximately 3.5 and 3 logs higher, respectively, than RNA shed in WA-1/2020 strain SARS-CoV-2 infected macaques fed a standard diet (**C**).

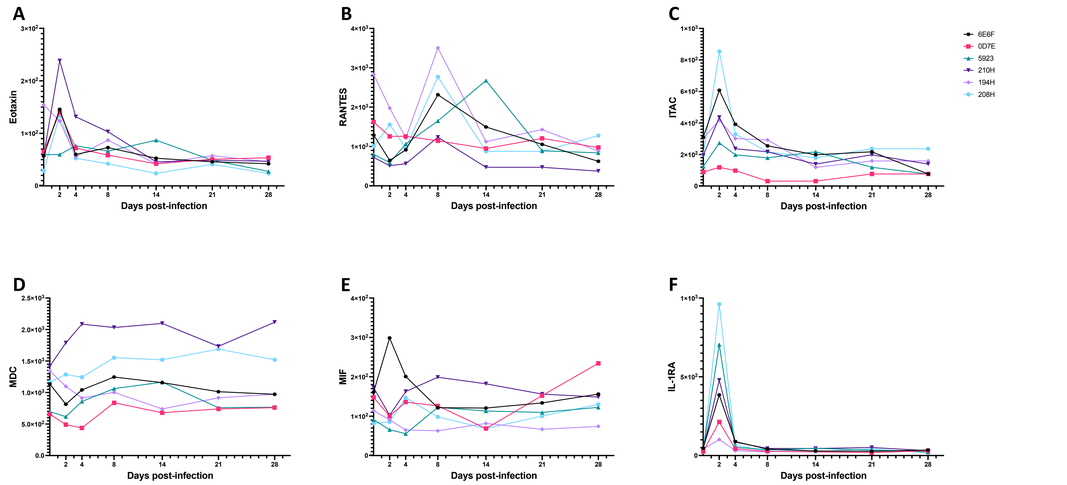
RESULTS

Immune Cells in Rectal Tissue Impacted by Diet and Infection

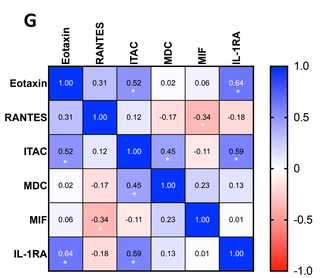


High fat and sugar diet (HFD) impacted the B cell compartment in rectal tissue, with significantly lower B cell frequencies after approximately two months of HFD administration (**A**, Wilcoxon matched-pairs tests). B cell frequencies remained significantly lower than pre-HFD levels at necropsy (**A**). Although not statistically significant, rectal IgG+ B cell frequency declined from one week pre-infection to approximately one month post-infection, with mean frequency at necropsy only 0.3% of B cells; necropsy frequencies were significantly lower than pre-HFD levels (**B**, Wilcoxon matched-pairs tests). While B cell frequency was decreasing during HFD administration, T cell frequency in rectal tissue significantly increased after approximately two months of HFD, and was modestly reduced at necropsy (**C**, Wilcoxon matched-pairs tests). NK cell frequencies were also modestly reduced from one week pre-infection to necropsy at approximately one month post-infection (**D**, Wilcoxon matched-pairs test).

Rapid Inflammatory Response to Infection

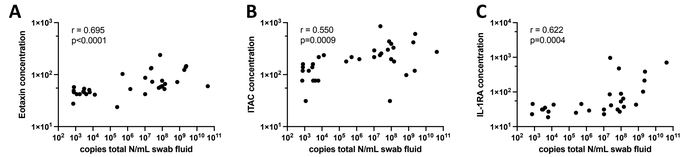


Systemic responses to SARS-CoV-2 infection were measured with Luminex of plasma cytokines. Noticeable increases following infection were observed in eotaxin/CCL11 (**A**), RANTES/CCL5 (**B**), ITAC/CXCL11 (**C**), and IL-1RA (**F**), with peaks at day 2 post-infection for all but RANTES, which peaked at day 8. Significant increases from day 0 to day 2 were observed for ITAC and IL-1RA (Wilcoxon matched-pairs tests), while MDC/CCL22 (**D**) and MIF (**E**) were comparatively stable throughout infection. Increases in both pro-inflammatory chemokines such as ITAC and anti-inflammatory cytokines such as IL-1RA at day 2 suggest a rapid inflammatory response to viral replication with a coordinated dampening response. Indeed IL-RA significantly correlated to ITAC and eotaxin (**G**, Spearman r shown and p<0.05 indicated with asterisk). Although the highest response were largely at day 2, the later peak of RANTES indicates an ongoing immune response at day 8.



RESULTS

Correlation of Viral RNA with Cytokine Responses



Spearman correlations of eotaxin (**A**), ITAC (**B**), and IL-1RA (**C**) with total viral RNA copies in nose swabs across all time points and animals. Correlations suggest the magnitude of the immune response is proportional to the quantity of viral antigen present.

CONCLUSIONS

High fat and sugar diet in macaques induces B cell loss and T cell expansion in rectal tissue, with T cells and NK cells modestly reduced one month following SARS-CoV-2 infection. Diet may influence gut barrier immunity and predispose the tissue to SARS-CoV-2 pathology.

- IgG+ B cell frequency was also modestly reduced with infection; mean frequency <0.5% at necropsy
- Macaques fed a high fat and sugar diet and infected with Delta SARS-CoV-2 showed enhanced viral shedding over WA-1/2020 infected animals fed a standard diet
- Pro- and anti-inflammatory cytokines increased rapidly in response to infection, with correlations to viral RNA burden

FUTURE DIRECTIONS

- Feed pigtail macaques a standard diet and infect with SARS-CoV-2 Delta strain
- Explore impact of diet and infection on:
  - gut microbiome composition
  - biomarkers of intestinal integrity
  - microbial translocation across gut

ACKNOWLEDGEMENTS

We would like to thank the animal care teams at BIOQUAL, Inc. for their management of study animals. Funding is provided in part by the National Institute of Allergy and Infectious Diseases Division of Intramural Research; the views and conclusions expressed here do not represent NIAID, NIH, or DHHS.

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