Introduction

Cortisol is the primary glucocorticoid hormone secreted in response to many adverse conditions, such as chemical, physical, and emotional stress. It is well-known for its anti-inflammatory and immunosuppressive actions at high levels.

Hypothesis

- Epinephrine will play a role in sensitization of immune cells to dexamethasone in blood from healthy dogs.
- The concentration of dexamethasone that inhibits 50% of cytokine response (IC50) will be measured. Shifts in dexamethasone sensitivity (IC50) will predict patient outcomes.

Objective

To develop a whole-blood based assay that enables a functional quantification of glucocorticoid sensitivity.

Methods

- Whole blood culture
- ELISA and MFI for cytokines

Each well contained the following:

- Whole blood diluted 1:2 with RPMI medium
- Lipopolysaccharide (100 ng/ml) or PBS
- 5 µM of Epinephrine or PBS
- Variable Dexamethasone
- Control (Phosphate buffered saline or PBS)

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Conclusion

Can the effect of stress on glucocorticoid sensitivity of immune cells in blood be modeled in vitro?

- Epinephrine recapitulates the effects of acute stress in vitro.
- Epinephrine plays a role in sensitizing the cells in whole blood of a healthy dog to dexamethasone.
- In a healthy dog, it takes less [dexamethasone] for the cells in whole blood to suppress IL-6 secretion in the presence of stressor.
- The cell response to epinephrine is more prominent in the presence of an active immunological stimulant, LPS.

Ongoing & Further Studies

Can we inform prognoses of critical patients with this assay by measuring their glucocorticoid sensitivity?

- Whole blood culture of a variety of sick dogs will be compared with cultures from healthy dogs to determine whether results from this assay correlate with the prognoses of dogs with critical illness.
- Patients with poorer prognoses are expected to have an increased IC50 or become insensitive to dexamethasone.
- Interestingly, we observed a decrease in cell sensitivity to dexamethasone with respect to suppression of basal IL-6 secretion in the presence of epinephrine. We will explore whether this result is repeatable in additional healthy animals. This suggests a possible mechanism by which the stress of being in the hospital could affect patient outcomes.

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