



# Does Heartrate Variability Contribute to Our Understanding of Stress?

## **Interpreting the Horse and Human Response to Equine Facilitated Psychotherapy**

Dr. Laurie McDuffee, &  
Dr. William Montelpare  
Comparative Wellness Laboratory,  
University of Prince Edward Island,  
Canada

August 13, 2022.

Horses and Humans Research Foundation (HHRF)  
Mission: *Through sustained investment in rigorous research, HHRF serves as a catalyst to advance global knowledge of horse-human interactions and their impact on health and wellness. [Horsesandhumans.org](https://www.horsesandhumans.org)*



# Background

- Where did we come from?  
A brief walk through our equine research in which we applied various techniques to help us understand the different responses of both horses and humans.



# Interpreting stress in horses as a clinician

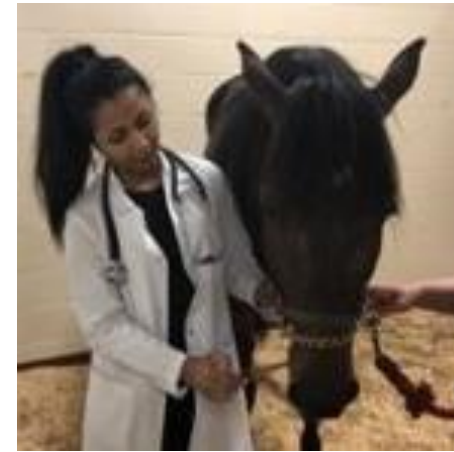


Table 1. Ethogram of Stress-Related Behaviors Counted During TR Sessions

Behavior	Behavior Description
Ears back	Both ears positioned caudally at a 45 degree angle from perpendicular
Head Raised	Head held higher than the normal carriage with the nose extended upward and neck stretched
Head turn	The movement of the head to the left or right, independent of the rider and their use of the reins
Head Toss	Head lowered below the withers, with ears back, followed by a sharp raise of the head
Head Shake	Repeated rhythmic movement of the head from left to right
Head Down	Head held below the withers, with nose extended downward and neck stretched
Biting Attempt	Bite movement directed at the rider, leader or side - walker
Kicking	Thrusting motion of one or both hind legs towards the side or back
Penile erection	Erection of the penis
Tail Swish	Tail is flicked to one side of the hindquarters without evidence of flies present
Yawning	Deep inhalation with open mouth
Swinging Hindquarters	Hindquarters moving back and forth from side to side
Licking the bit	Manipulation of the bit using the tongue, independent of the rider and their use of the reins



Common methods



Stress and THR



Stress during common VTH procedures



HRV Reliability during activities

# Progress in Learning about HRV and Measuring Stress

- Stress during positive and negative reinforcement: HR monitors (Jane)
- Reliability of the Polar HR monitor: HRV measures and intraclass correlation (Molly)
- Stress in TRH: stress ethogram, HRV measures, and cortisol (Larissa)
- Stress in equine patients during various veterinary procedures: stress ethogram, HRV measures, and cortisol (Anam)
- Pain vs stress: HRV as a measure of pain in equine patients, pain scores, cortisol (Jackie)
- EFP effects on humans and horses: Horses and Humans: HRV, cortisol, oxytocin; Humans: psych tests; Horses: stress behaviour ethogram (HHRF funded)

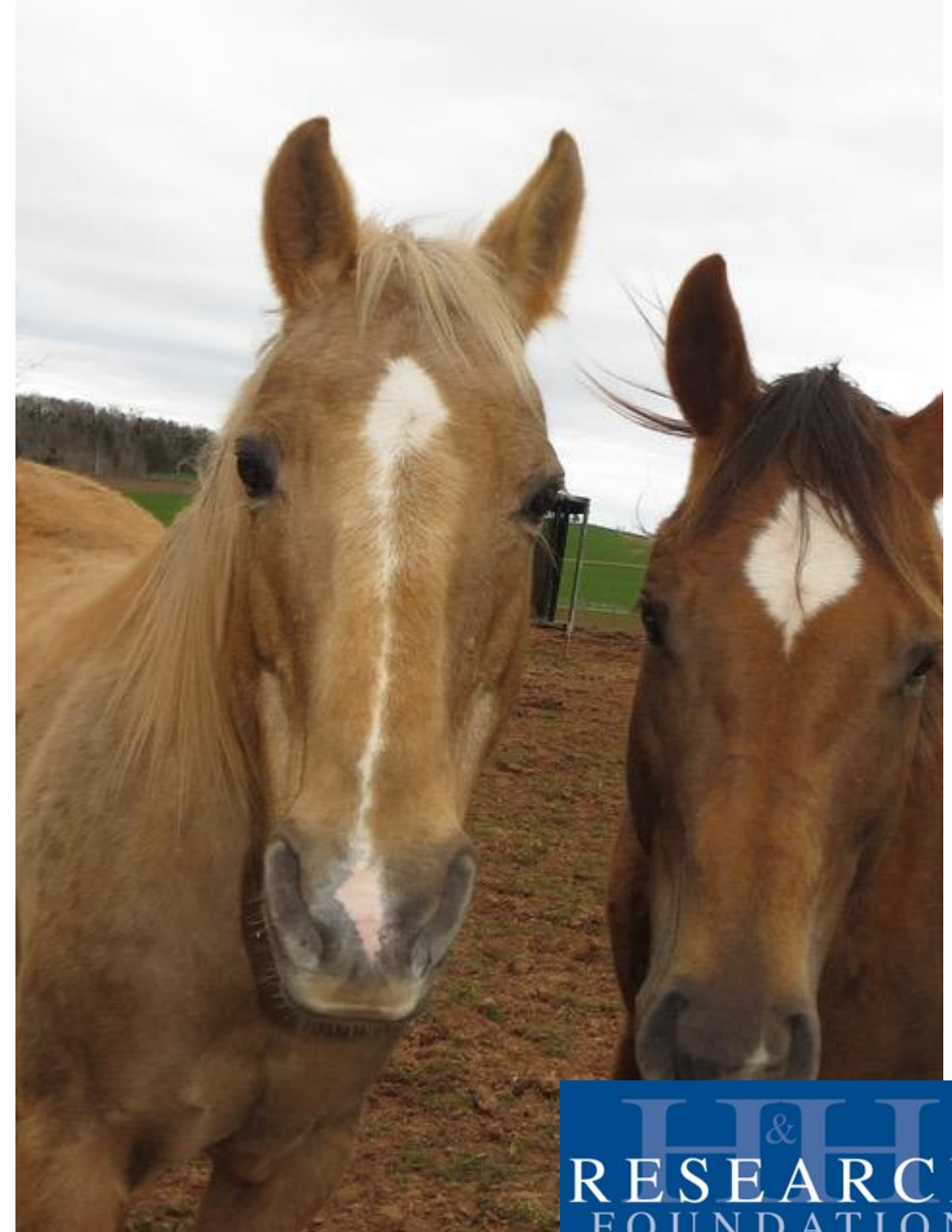
## OUR PLAN



**This presentation will draw on results from our research program with an emphasis on of the interaction between horses and humans during EFP: “Psychophysiological effects of Equine-assisted therapy on horses and in veterans diagnosed with post-traumatic stress disorder (PTSD)”.**

In particular ...

How do the measures that we collected - cortisol, oxytocin, and the derivatives of HRV (time, frequency, and Poincare plots) contribute to our understanding of stress?



---

## THREE IMPORTANT TAKE AWAYS

i) an understanding of the complexity of evaluating stress from both an objective and subjective perspective

ii) the efficacy of tools that can be used to determine stress responses

iii) the value of the various metrics used to demonstrate both individual and synchronized responses of horses and humans to stress.



## OBJECTIVES

Animal-assisted services (AAS) are used to promote the mental and physical health of humans and for our part we were interested in evaluating the efficacy of equine-assisted services for veterans with PTSD.

As part of this research, we were also interested in the welfare of the horses that were recruited to the EFP program and moreover to determine if the horses' participation in EFP led to a measurable level of stress.



## DATA COLLECTION



- Horses & Humans wore Polar Heart Rate Monitors, and RR recordings were obtained for HRV measures
- Saliva was collected for cortisol concentrations
- Saliva or blood was collected for oxytocin concentrations



*Considering the  
measurement characteristics of  
STRESS, we began by asking ...  
**What is STRESS?***



- *The answer is, stress is a construct, which we cannot measure directly.*

■ ***Because we cannot measure stress directly, in our studies we viewed STRESS as a latent variable that we measured indirectly with different approaches.***

*Horses were measured with behavior rating scales.*

*Humans, self reported psychology measures along with of mood, anxiety or well-being*



*Measured with heart rate variability: in the time domain, the frequency domain, or through non-linear estimates of entropy (disorderliness - variability)*

*Measured with a physiological response such as cortisol and oxytocin*



Objective ratings

Cortisol

Subjective Ratings

Heartrate Variability

I love oxytocin!

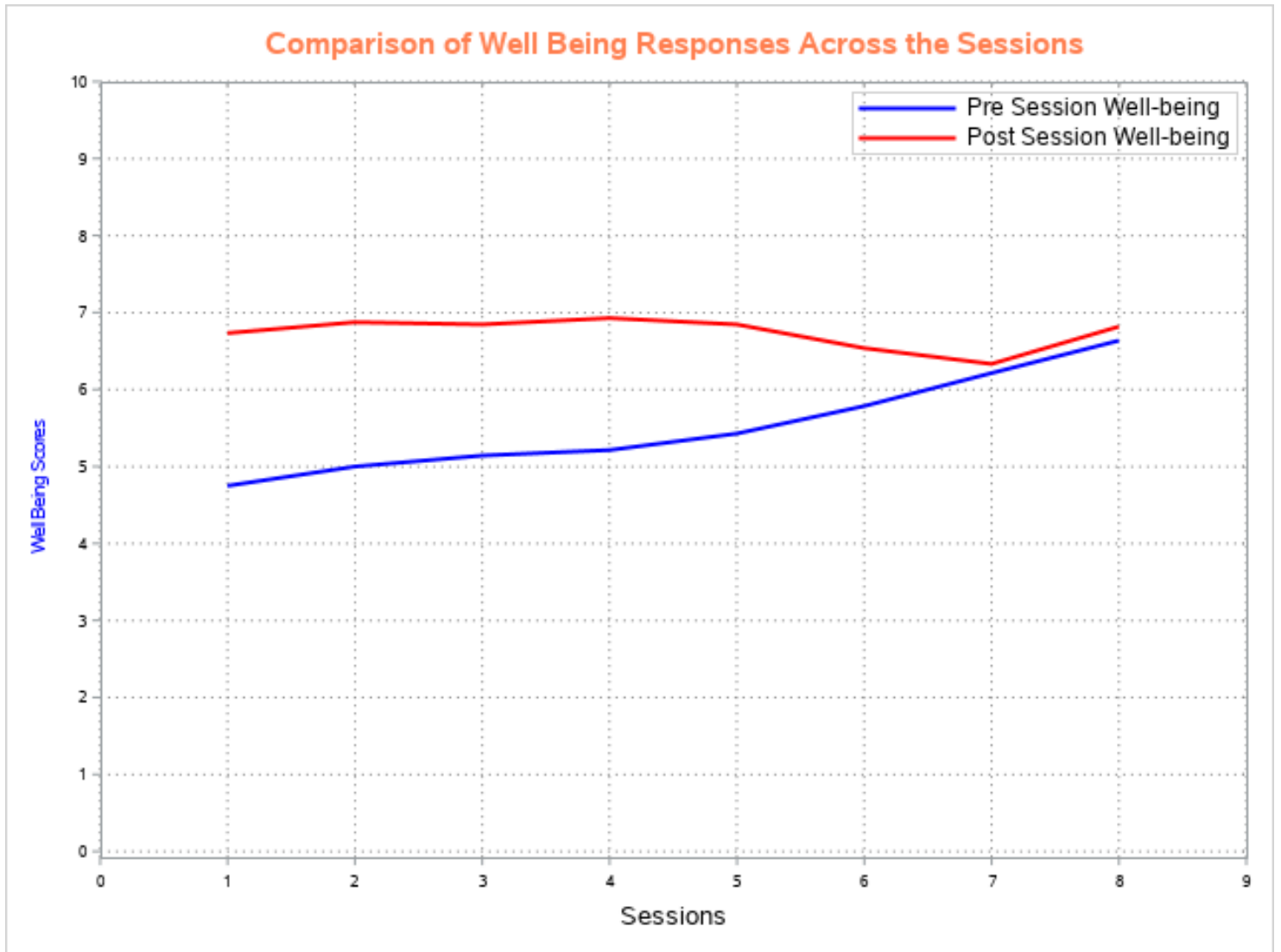
Which is best?

# RESULTS



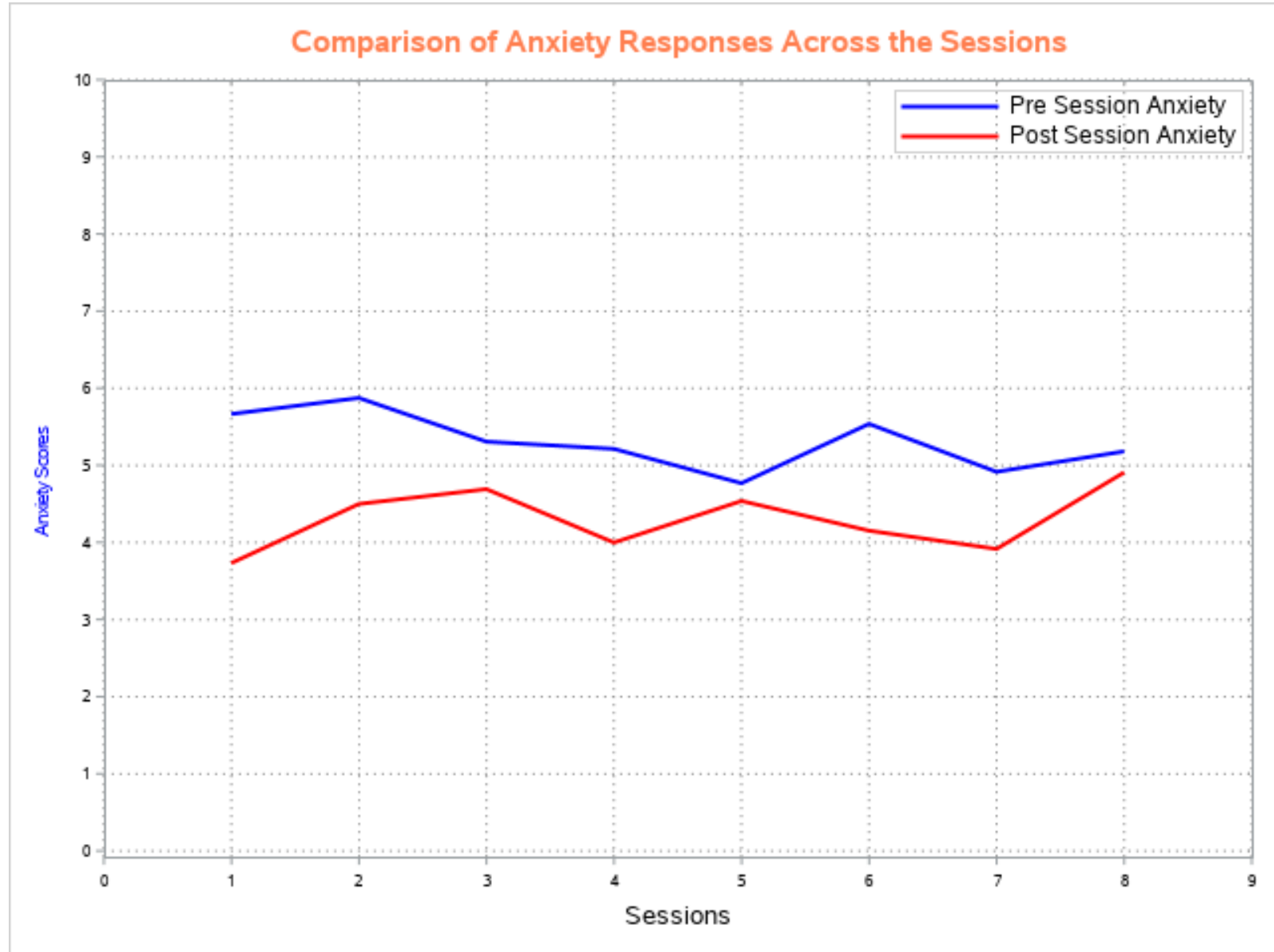
## RESPONSES FOR HUMANS

Our results showed that the measures of well-being improved across each of the sessions, when measured daily.



## RESPONSES FOR HUMANS

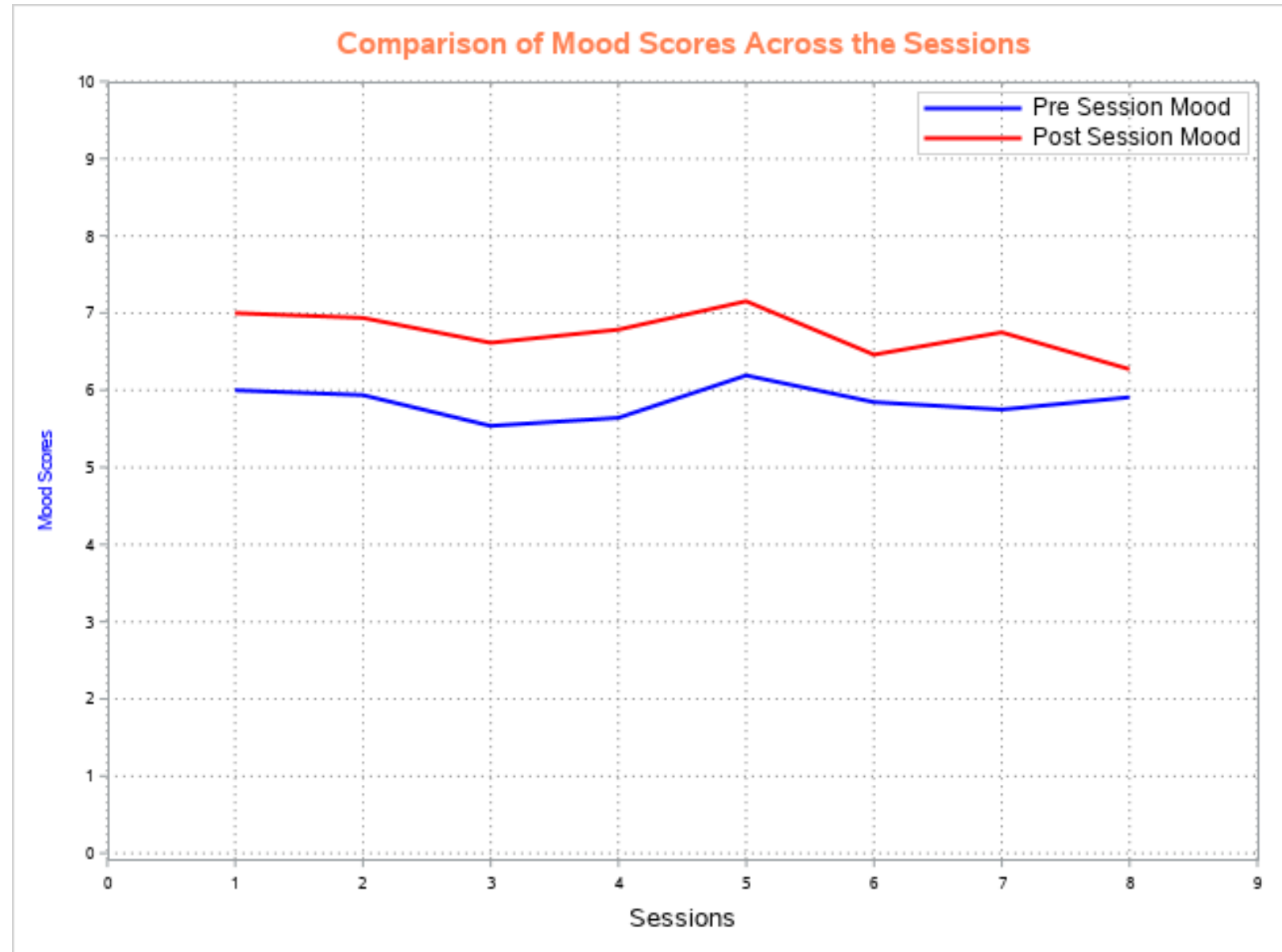
Here we see that the measures of anxiety decreased at the end of each session, when measured daily.





## RESPONSES FOR HUMANS

And the measures of mood improved at the end of each session, when measured daily.



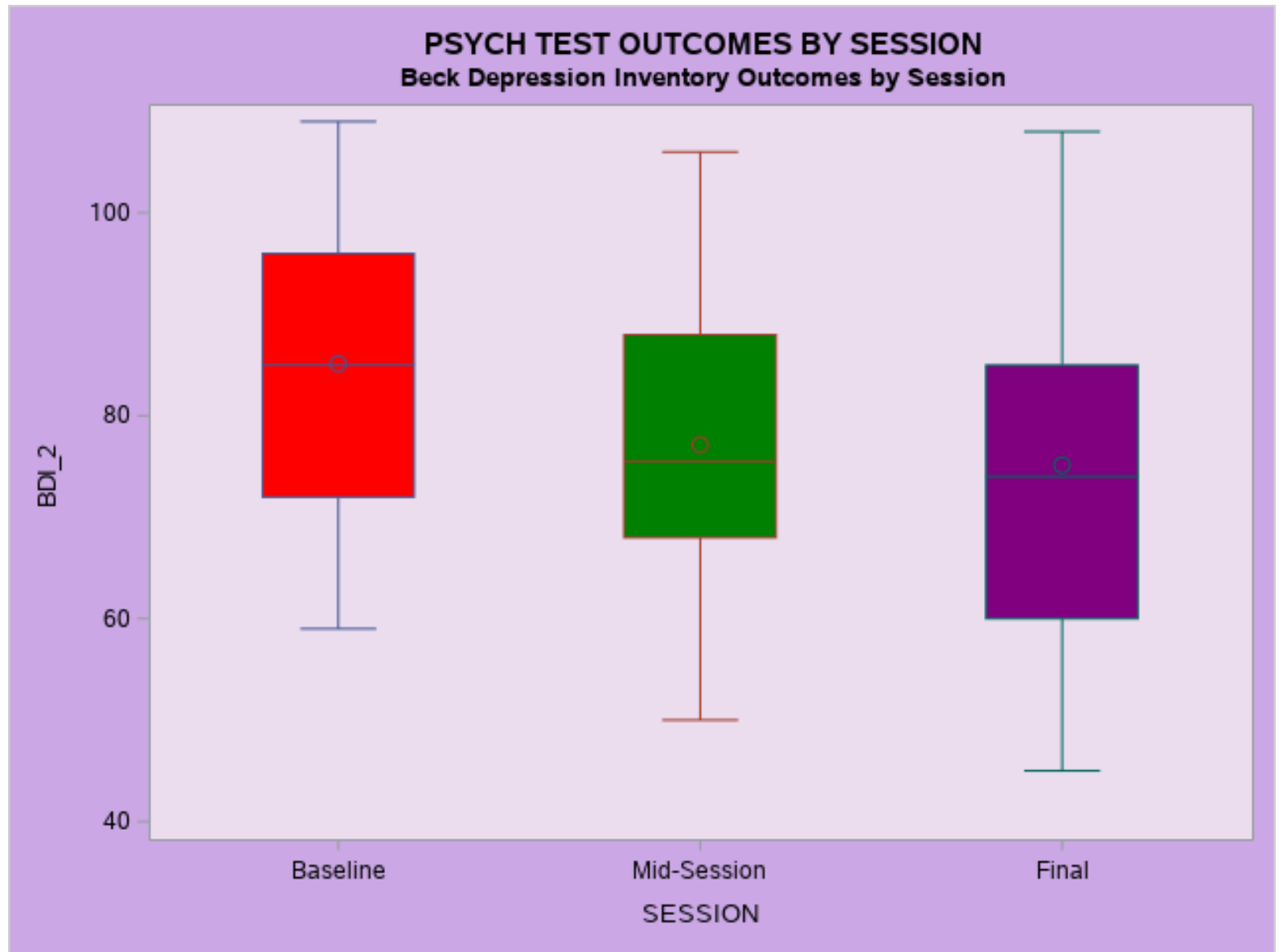
## SELF REPORTED SURVEY RESPONSES FOR OUR HUMAN PARTICIPANTS



- *In addition, we measured changes in the participant's overall psychological responses, at the start of the cohort sessions, mid-way through the cohort sessions, and at the end of the cohort sessions using the*
  - *the Beck Depression Inventory*
  - *the MAQ- global assessment anxiety symptoms*
  - *the RAND SF-36 symptom*
  - *State-trait anger expression scale*
  - *PTSD checklist*

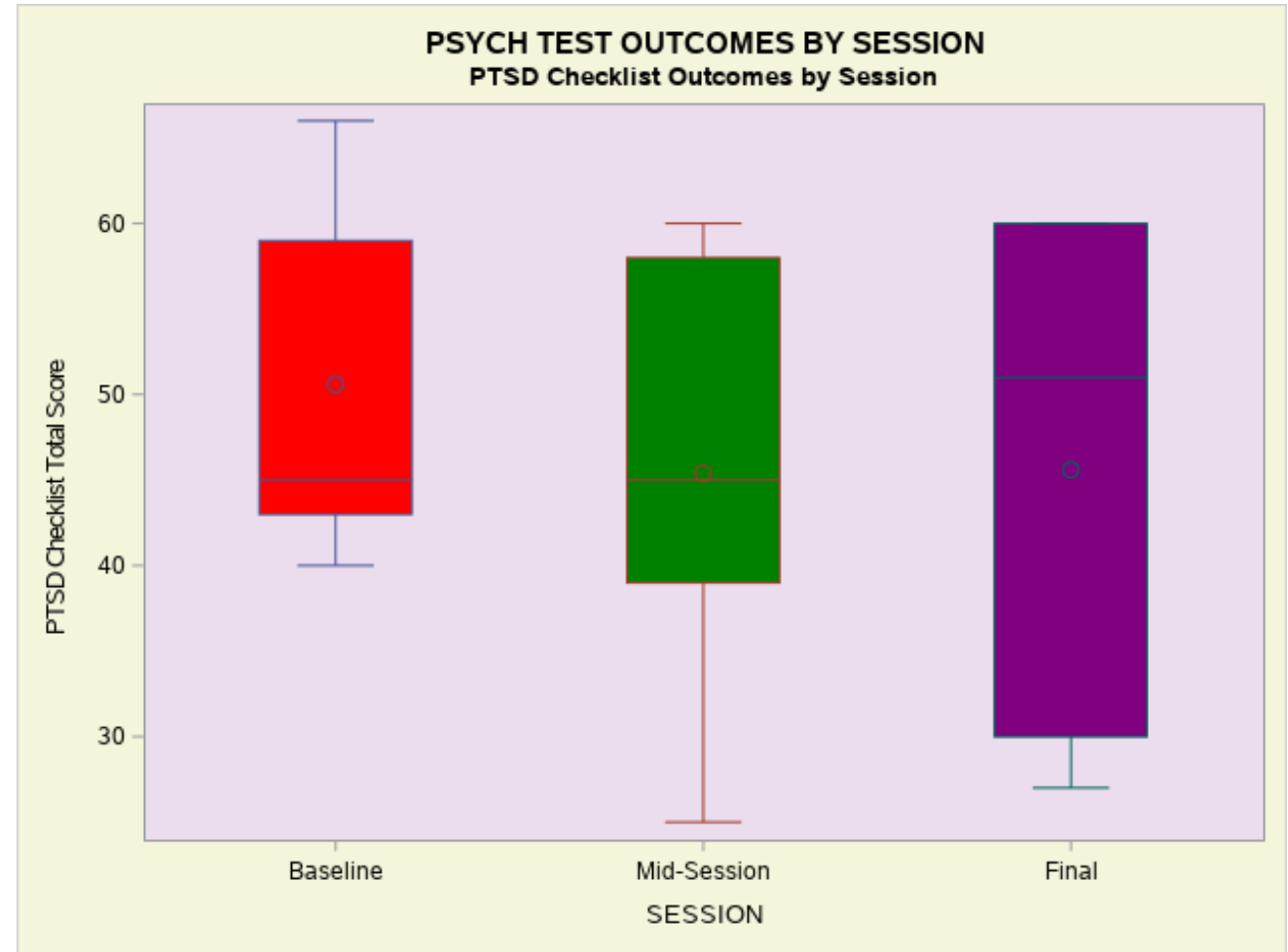
## INTERMITTENT PSYCHOLOGICAL ASSESSMENT RESPONSES FOR HUMANS

Here we notice that there was a decrease in the Beck Depression scores for the total group across the duration of the program, even though it was not statistically significant as anticipated because of the sample size.



# INTERMITTENT PSYCHOLOGICAL ASSESSMENT RESPONSES FOR HUMANS

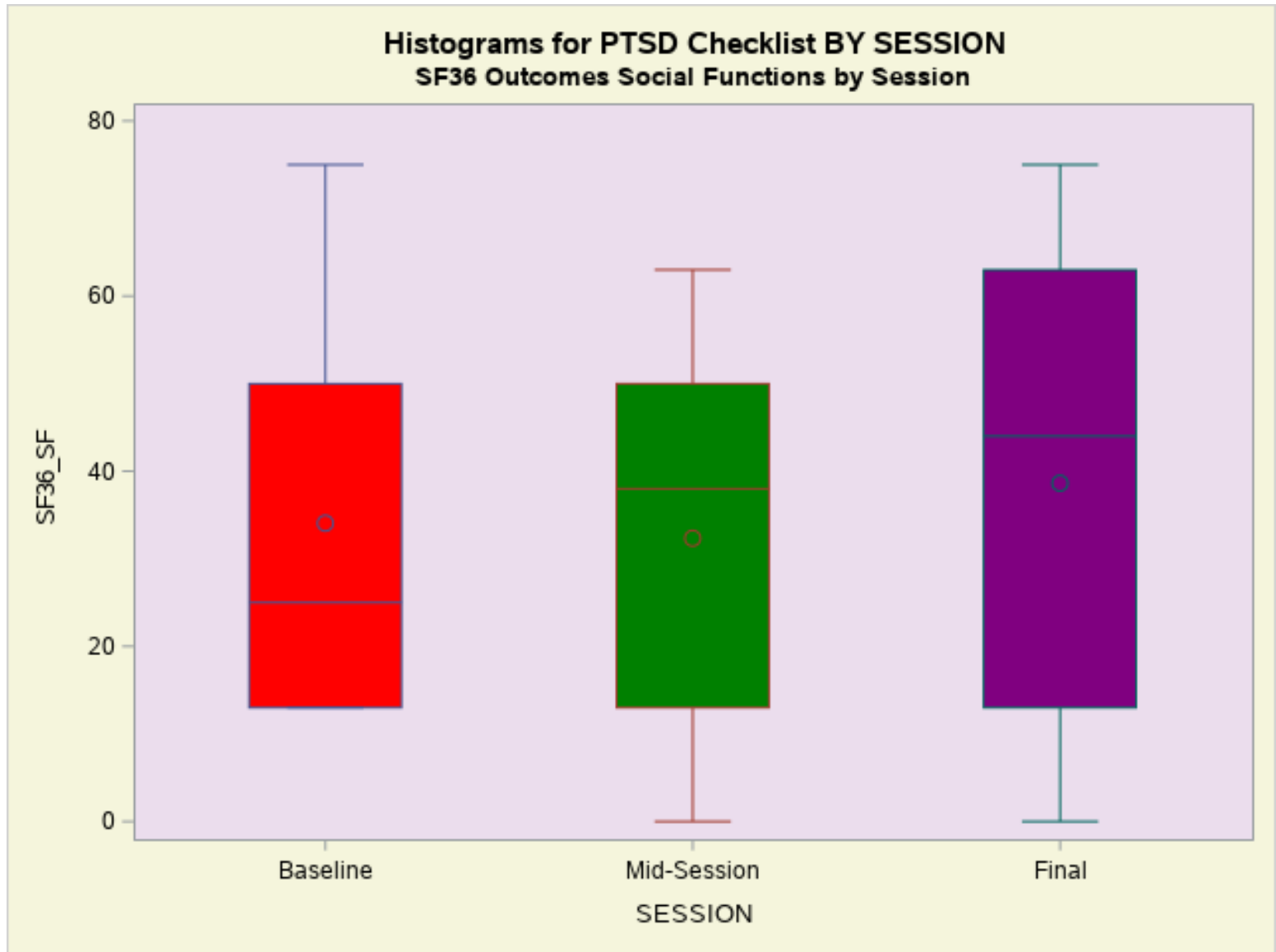
Here we show the responses to the PTSD Checklist based on reporting at the start-middle- and end of sessions.



# INTERMITTENT PSYCHOLOGICAL ASSESSMENT RESPONSES FOR HUMANS

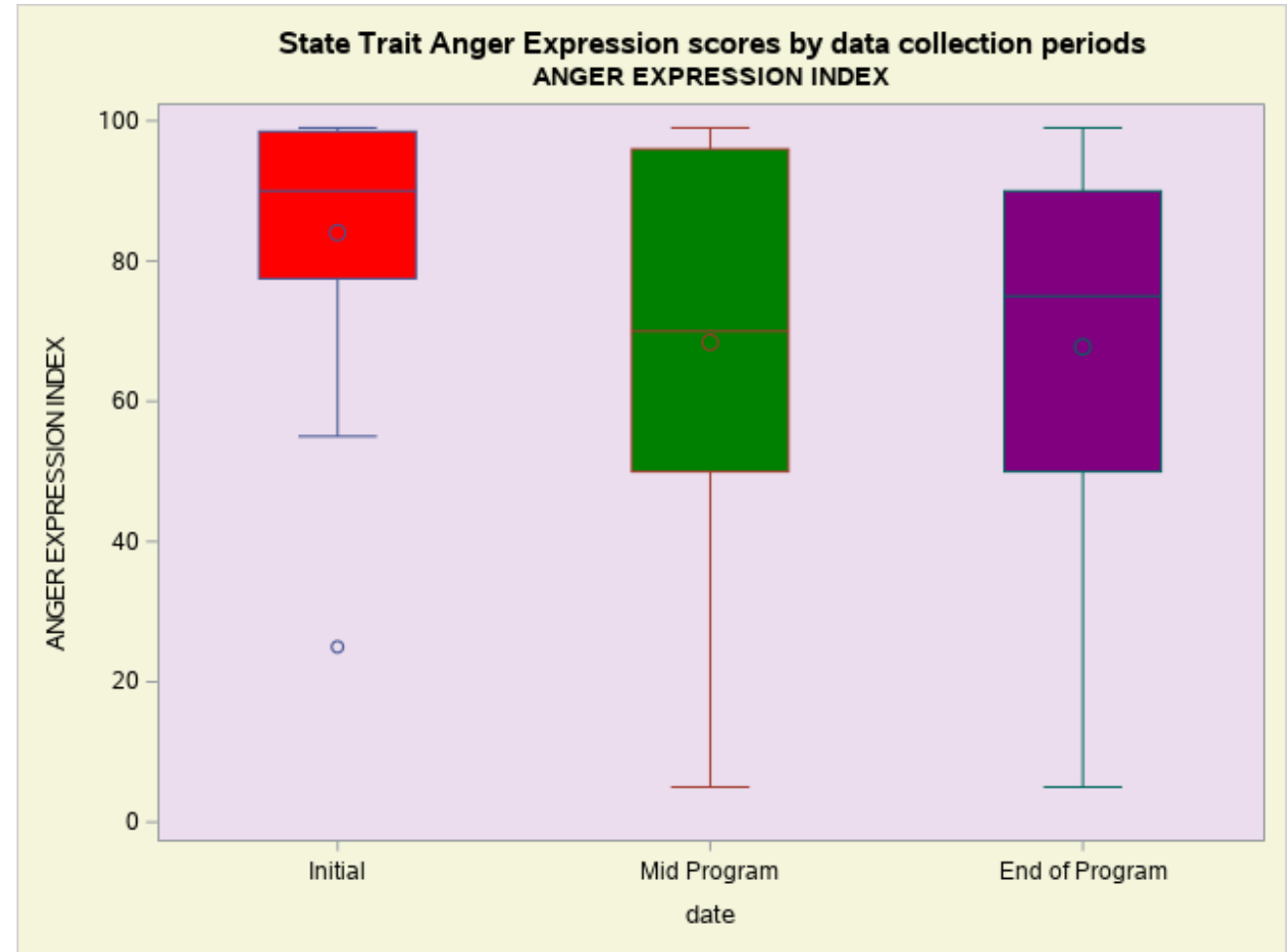
Here we show the responses to the SF-36 Outcomes for the Social Functioning construct based on reporting at the start- middle- and end of sessions.

The scores represent % of social functioning among respondents over time.



And finally here we show one of the outcomes for the State - Trait Anger Expression Scores based on reporting at the start- middle- and end of sessions.

In all these intermittent survey responses, we did not see a statistically significant change in the group overall. However, we did see changes which suggest that we need a larger sample size with a program of a longer duration, and possibly more exposures to influence these typical TRAIT estimates.



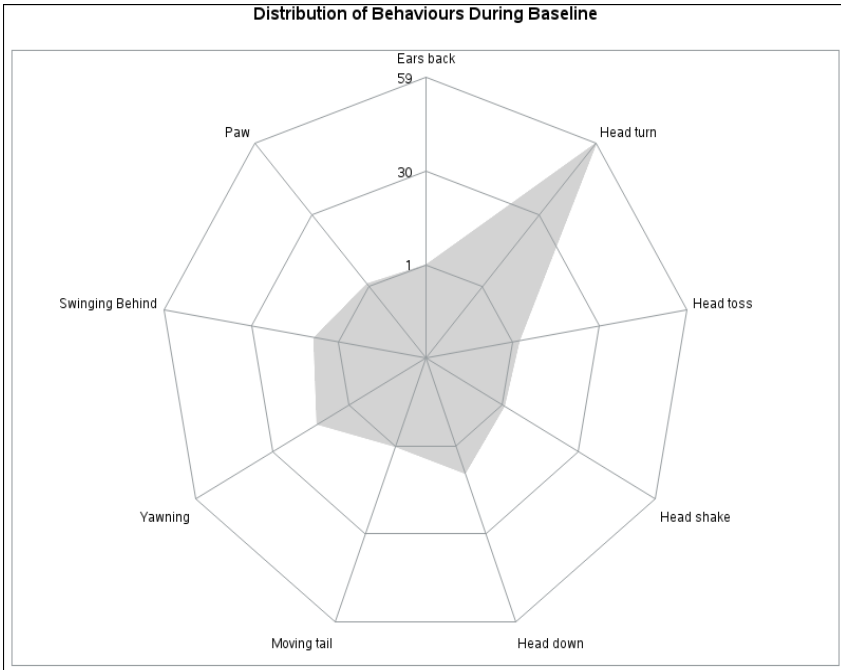
WE MEASURED  
BEHAVIORS  
AMONG OUR  
HORSE  
PARTICIPANTS  
USING THIS  
ETHOGRAM



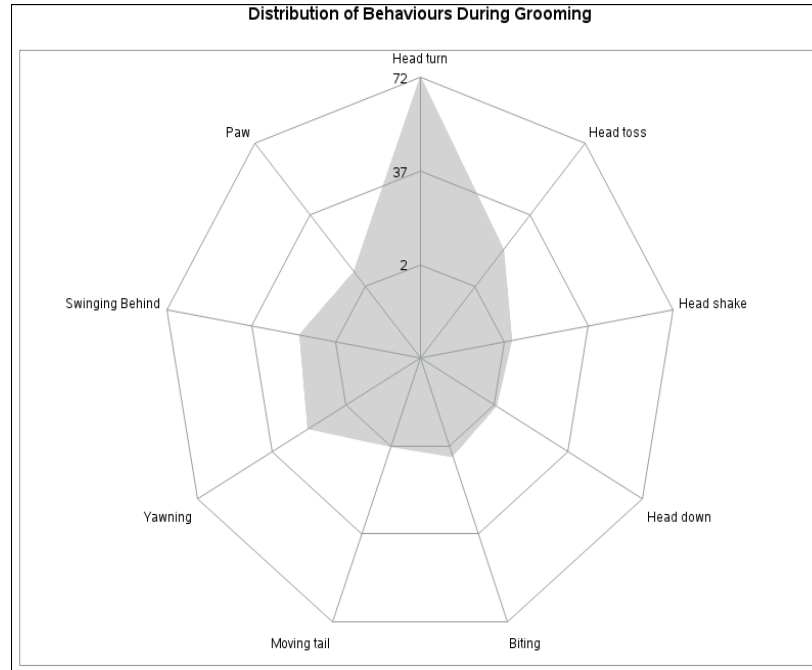
LAM

Ears pinned back	Ears pointed caudally at 45 degree angle
Turning head	Movement of the head away from handler independent of the handler's commands
Tossing the head	Head lowered below the withers, with ears back, followed by a sharp raise of the head
Shaking the head	Repeated movement of the head from left to right, flipping the head
Holding the head down	Head held below the withers, with nose extended downward and neck stretched
Biting at the handler	Bite movement directed at the handler
Kicking at the handler	Thrusting motion of one or both hind legs towards the side or back, directed toward the handler
Moving the tail	Excessive movements of the tail, characterized by a swinging motion from the left to the right
Yawning	Deep inhalation with open mouth
Swinging hindquarters	Swinging motion of the hindquarters, from side to side, independent of handler commands
Rearing	Shifting of weight-bearing to hind legs, with at least both front legs leaving the ground
Pawing	Scraping of air or ground with front hoof

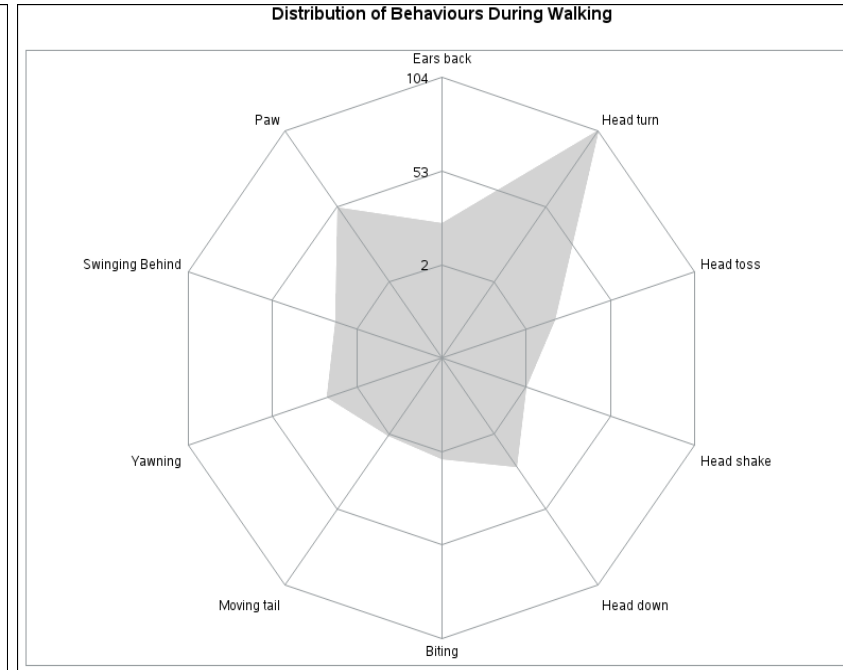
# RADAR GRAPHS FOR THE FREQUENCY OF HORSE BEHAVIORS ACROSS ACTIVITIES



Baseline



Grooming



Post walk (end of session)

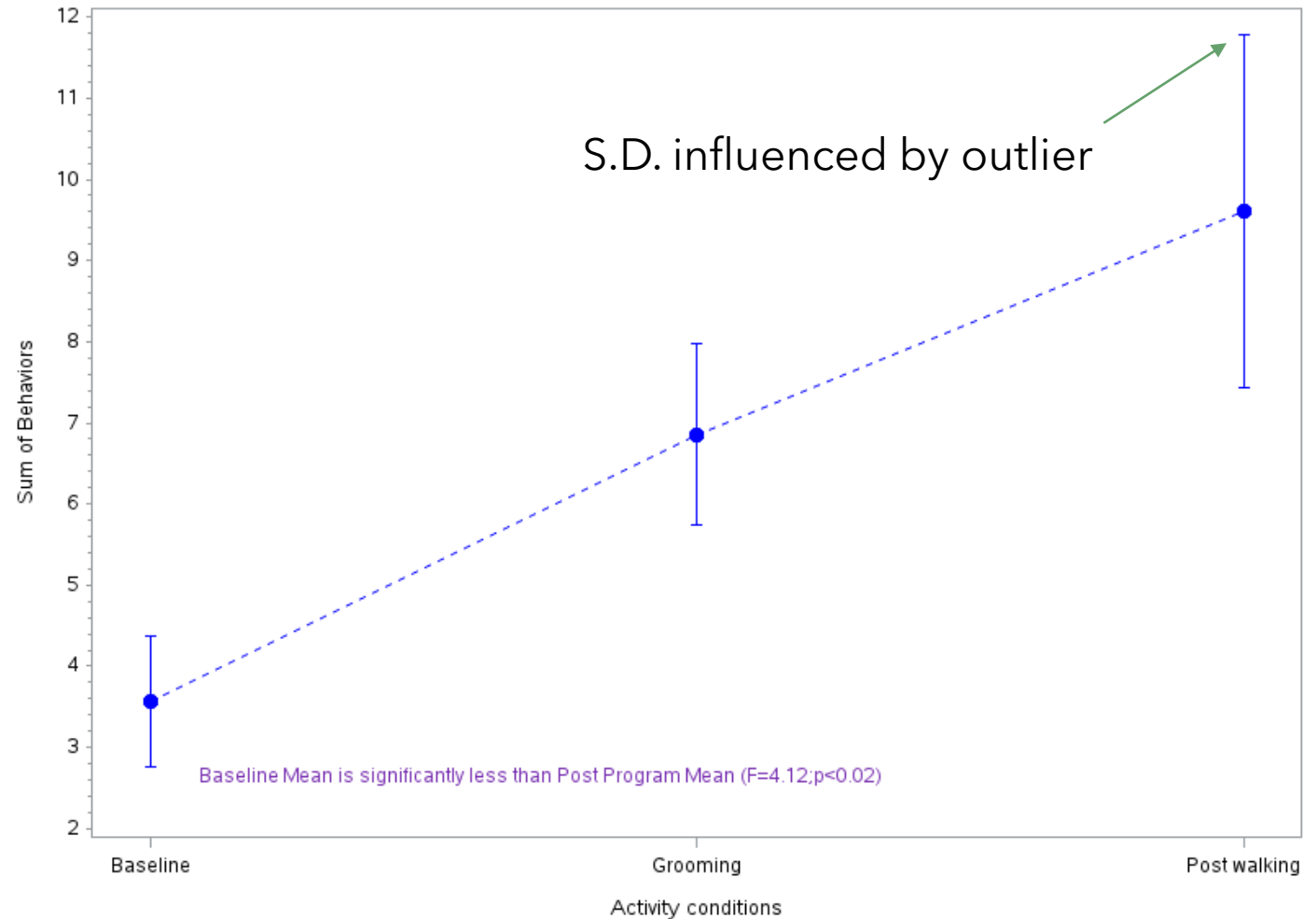
- Notice which behaviors showed the higher frequencies across the activities



# OBSERVATIONS ON HORSE PARTICIPANTS: BEHAVIOR SCORES

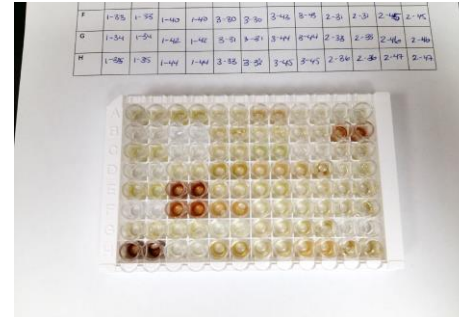
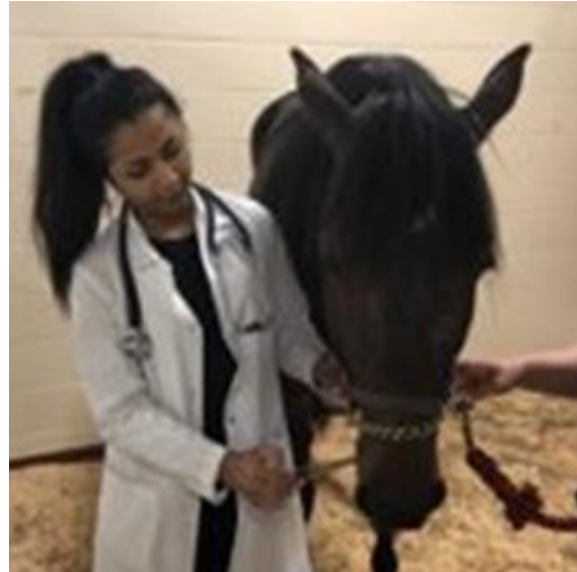
- Comparison of average number of behaviors across activities  
Average @ Baseline =  $3.57 \pm 4.25$   
Range=0 to 15
- Average @ Grooming =  $6.86 \pm 5.95$  Range=0 to 19
- Average @ Post Program =  $9.60 \pm 11.51$  Range=0 to 59

Plot of Means with Standard Error Bars - Behaviour by Activity Group



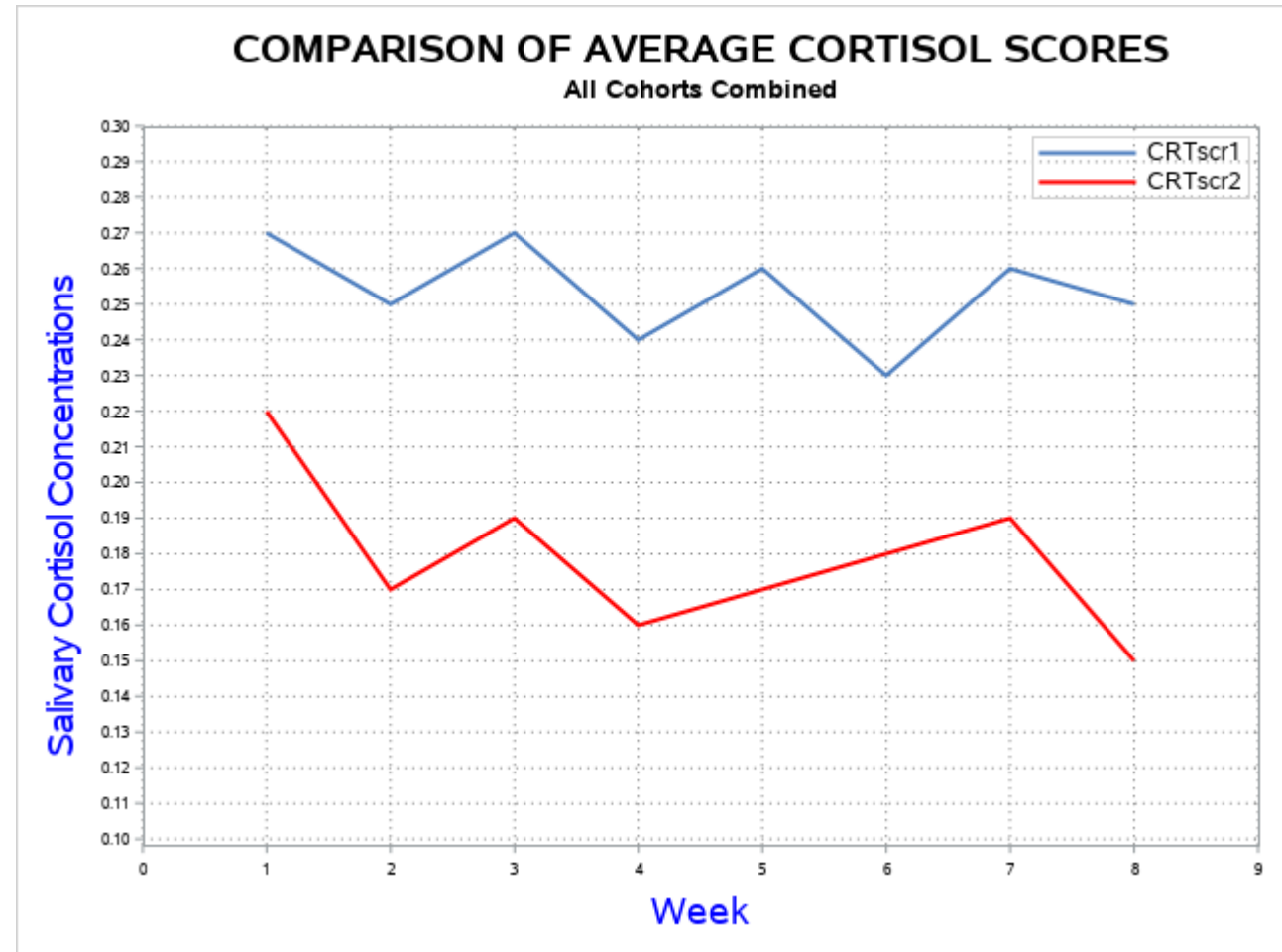
# OBJECTIVE DATA

Our assessments of stress also included measures of salivary cortisol and salivary oxytocin in humans and salivary cortisol and blood oxytocin for our horse participants



# OBSERVATIONS ON HUMAN PARTICIPANTS: CORTISOL

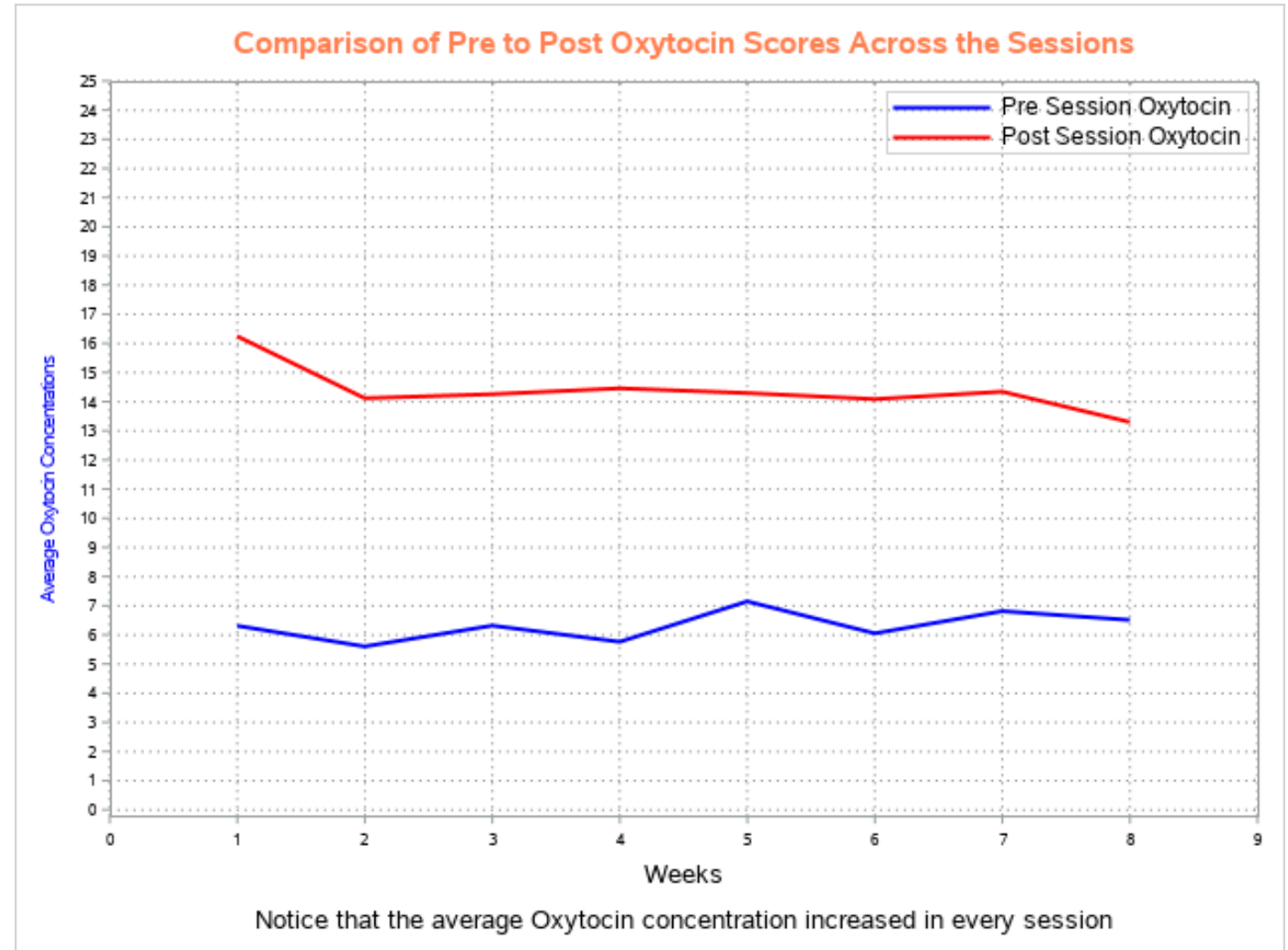
- A comparison of the average pre versus post session cortisol for human participants across the weekly sessions showed that there was a significant difference in average cortisol concentrations for the entire sample.



Method	Variances	DF	t Value	Pr >  t
Pooled	Equal	209	4.30	<.0001
Satterthwaite	Unequal	200.48	4.30	<.0001

# OBSERVATIONS ON HUMAN PARTICIPANTS: OXYTOCIN

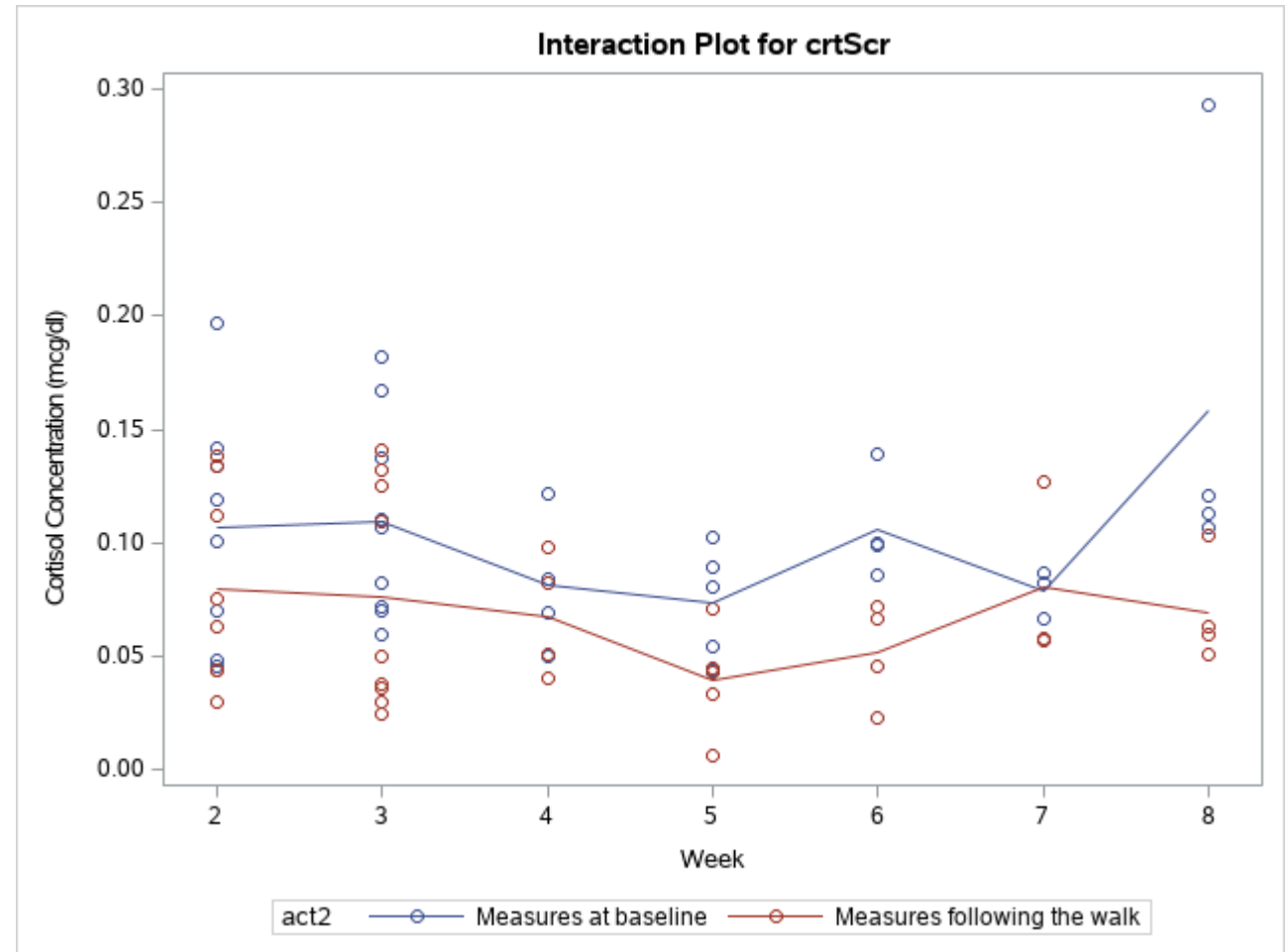
- A comparison of the average pre versus post session oxytocin concentrations for human participants across the weekly sessions showed that there was a significant difference in average oxytocin concentrations for the entire sample.



Tests for Location: $\mu_0=0$				
Test	Statistic		p Value	
Student's t	t	13.45	Pr >  t	<.0001
Sign	M	34	Pr >=  M	<.0001

# OBSERVATIONS ON HORSE PARTICIPANTS: CORTISOL

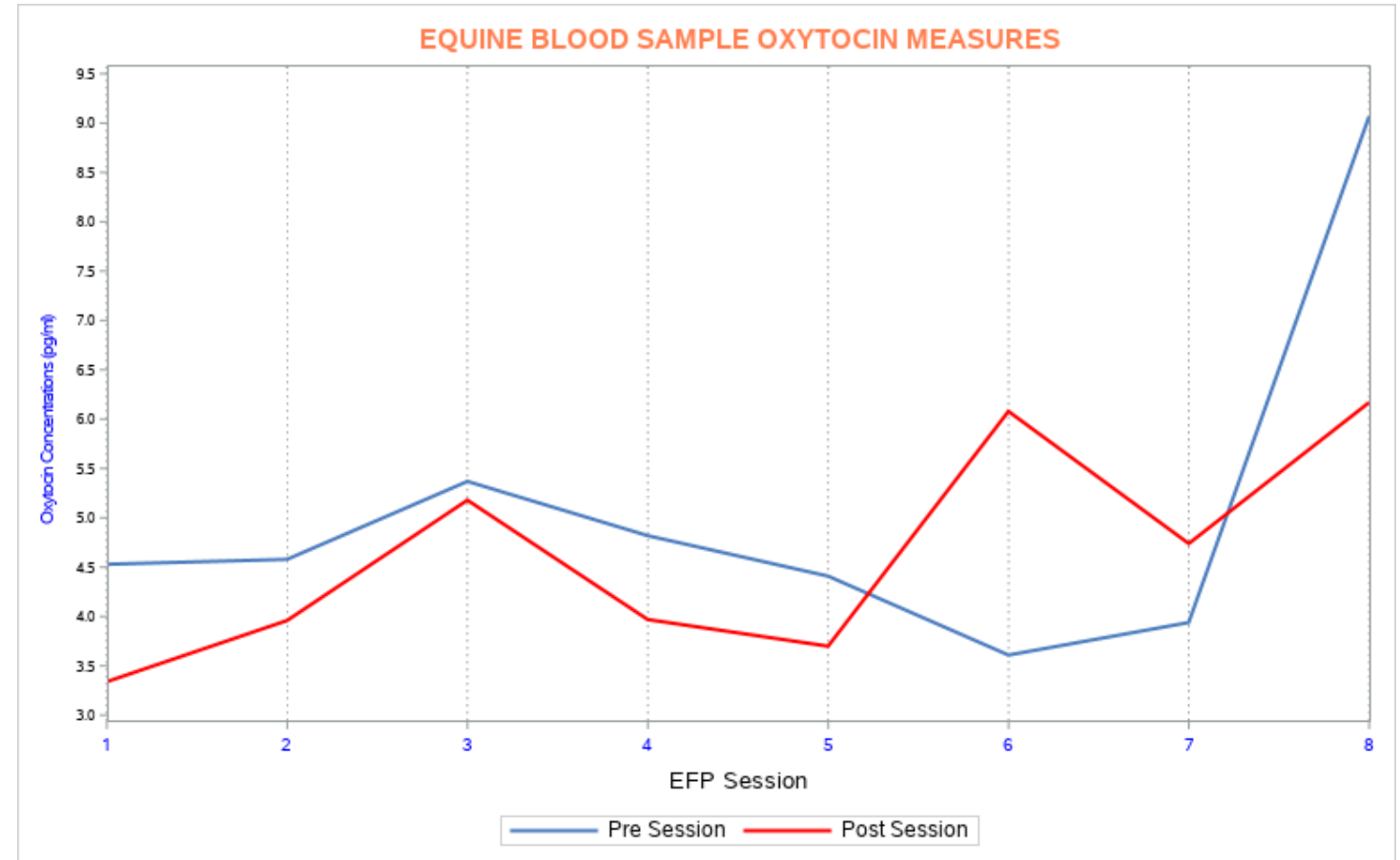
- Here we see cortisol responses of the horses across 7 weeks of the program.
- The results showed a significant decline in the **overall mean** from pre to post levels of salivary cortisol in horses across the sessions ( $F=2.19, p<0.01, df=13,60$ ).



Source	DF	Type III SS	Mean Square	F Value	Pr > F
Week	6	0.01821522	0.00303587	1.68	0.1405
act2	1	0.02052059	0.02052059	11.38	0.0013
Week*act2	6	0.00984252	0.00164042	0.91	0.4940

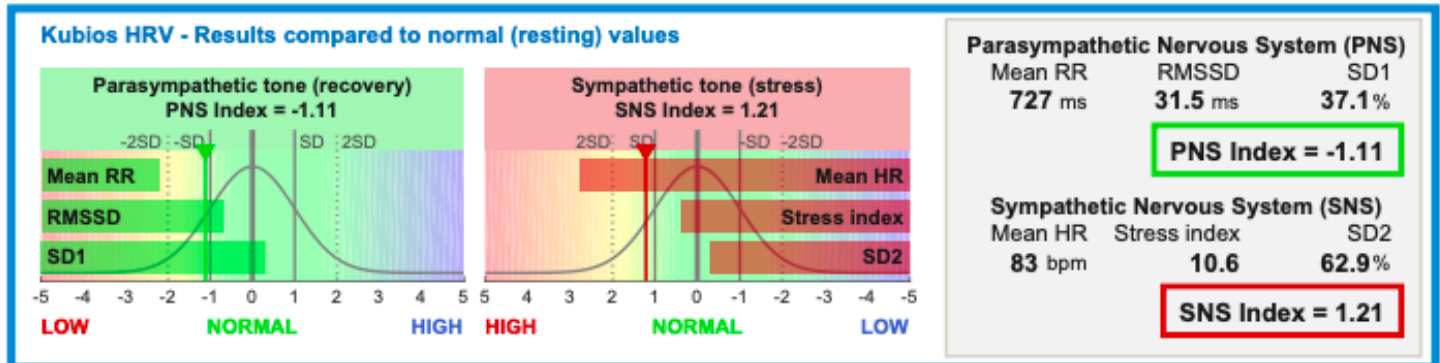
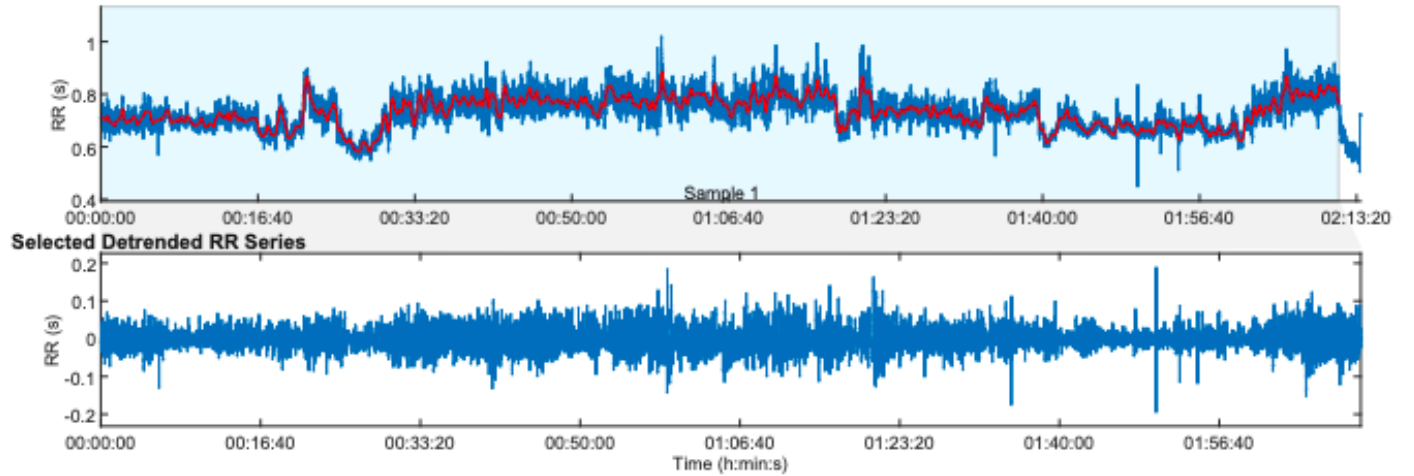
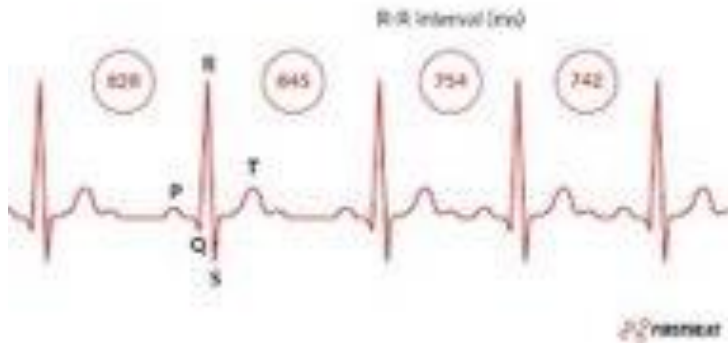
## OBSERVATIONS ON HORSES: OXYTOCIN

- Graph of the average pre to post blood oxytocin measures for the horses across the weekly EFP sessions.
- The results showed that there was **no significant** difference in the pre to post blood oxytocin measures across the sessions.



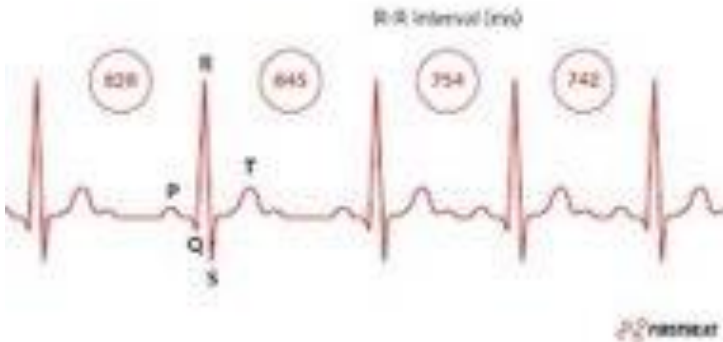
# HEART RATE VARIABILITY MEASURES

## Heart rate variability interpretation

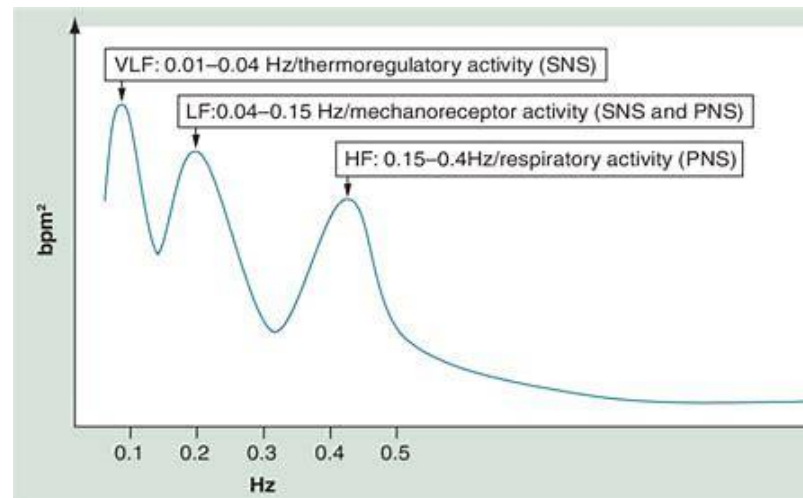


# THREE WAYS WE MEASURED HEART RATE VARIABILITY

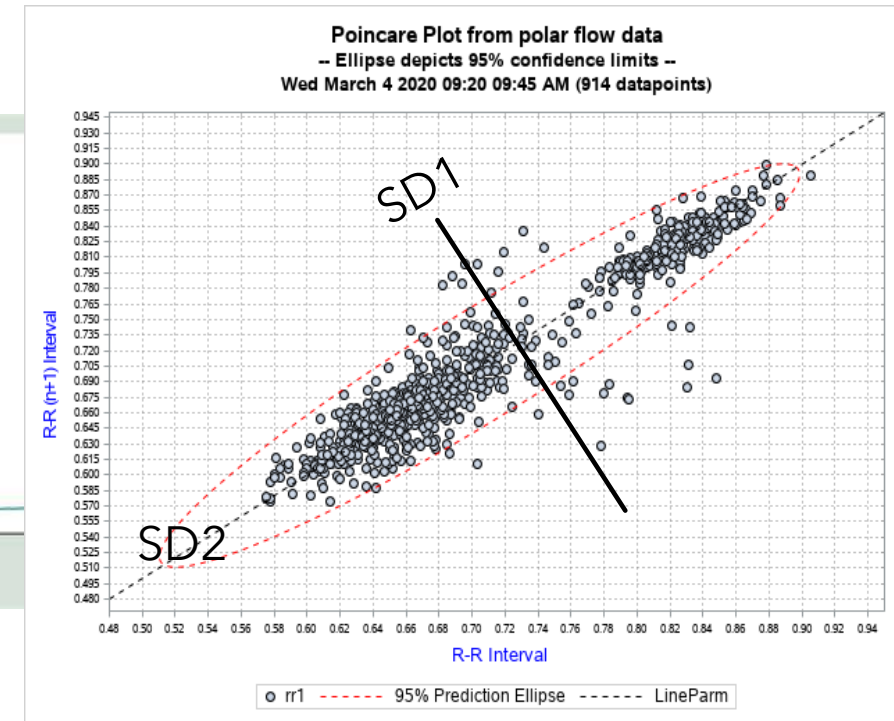
- Time Domain - refers to the linear scale of time. Here our measures are of the beat-to-beat rhythms and our estimates are derived from the distance between each heartbeat



Frequency Domain - refers to the pattern of the responses and shows the number of times a pattern of n-bpm occurs - this illustrates the pattern of oscillations in the heart rate rhythms

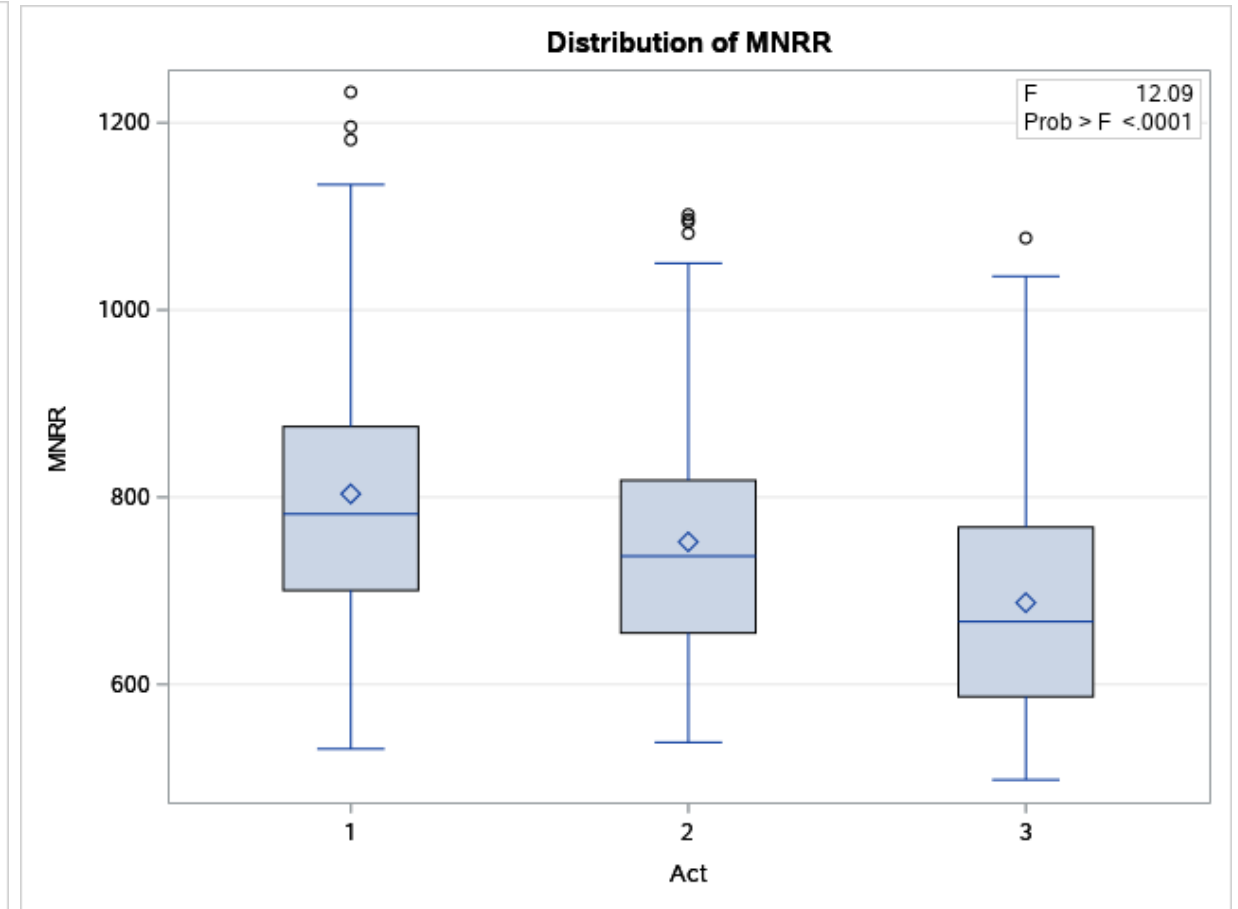
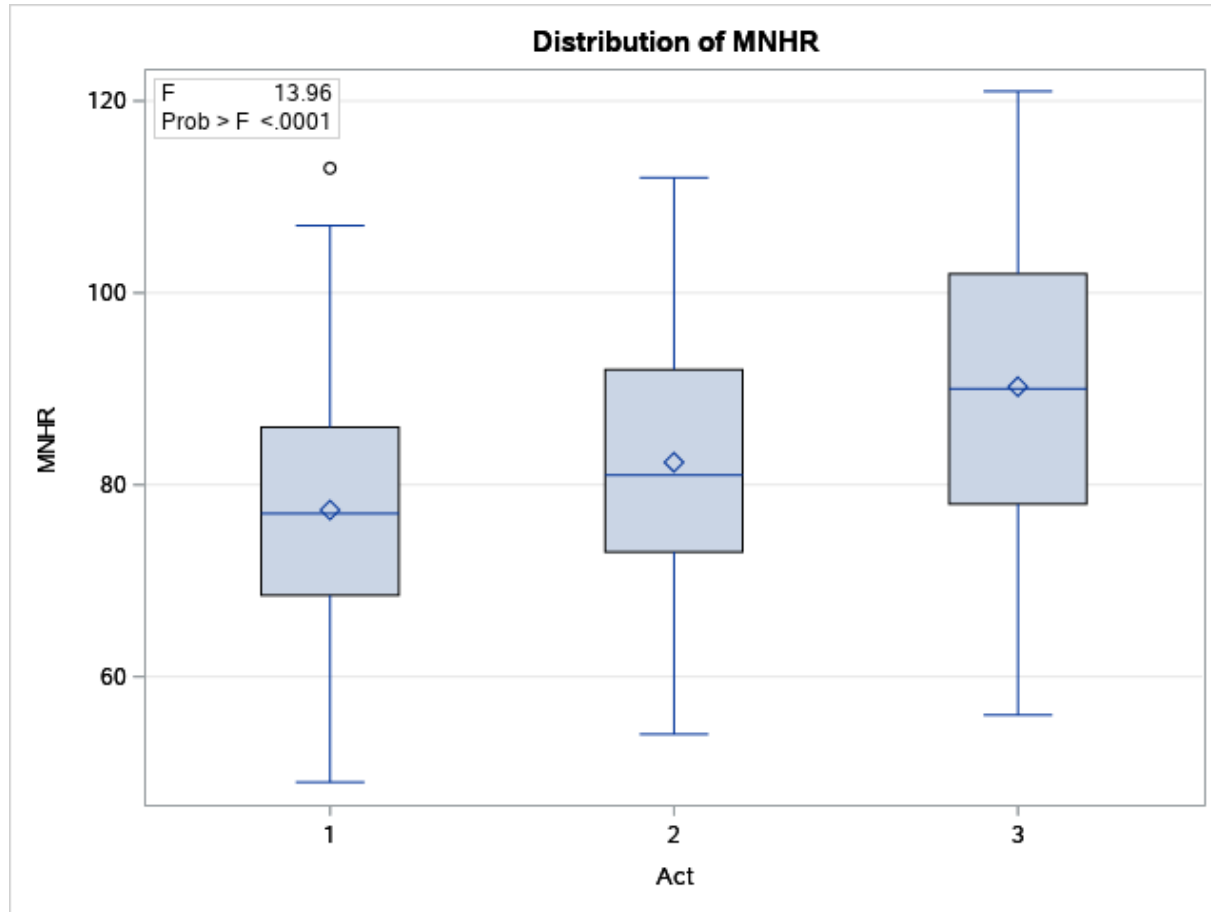


- Non-linear estimates - referring to the non-time scaled measures of beats, such as the Poincare Plots, SD1 and SD2 and the SNS and PNS indices





**FOR HUMANS** -- A comparison of time domain measures showed that the average heart rate increased across the activities, while the average r-r interval decreased

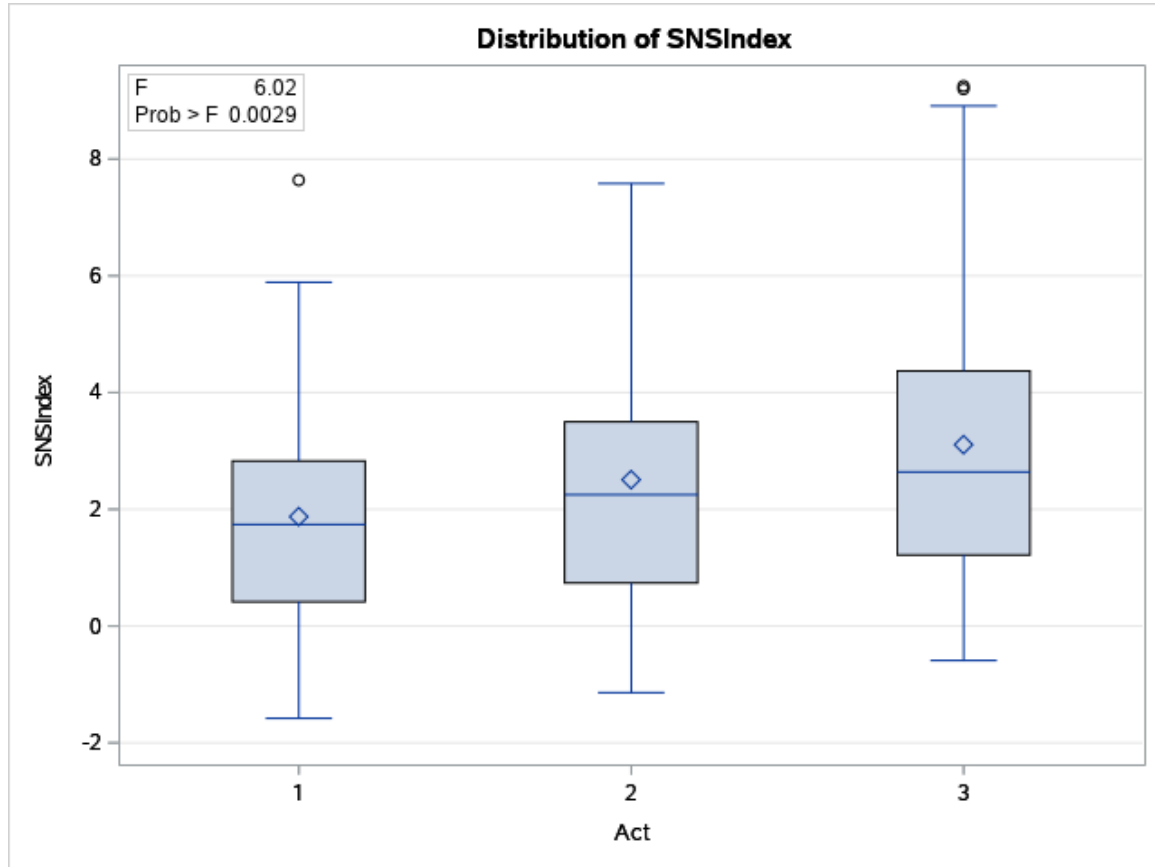


WJM

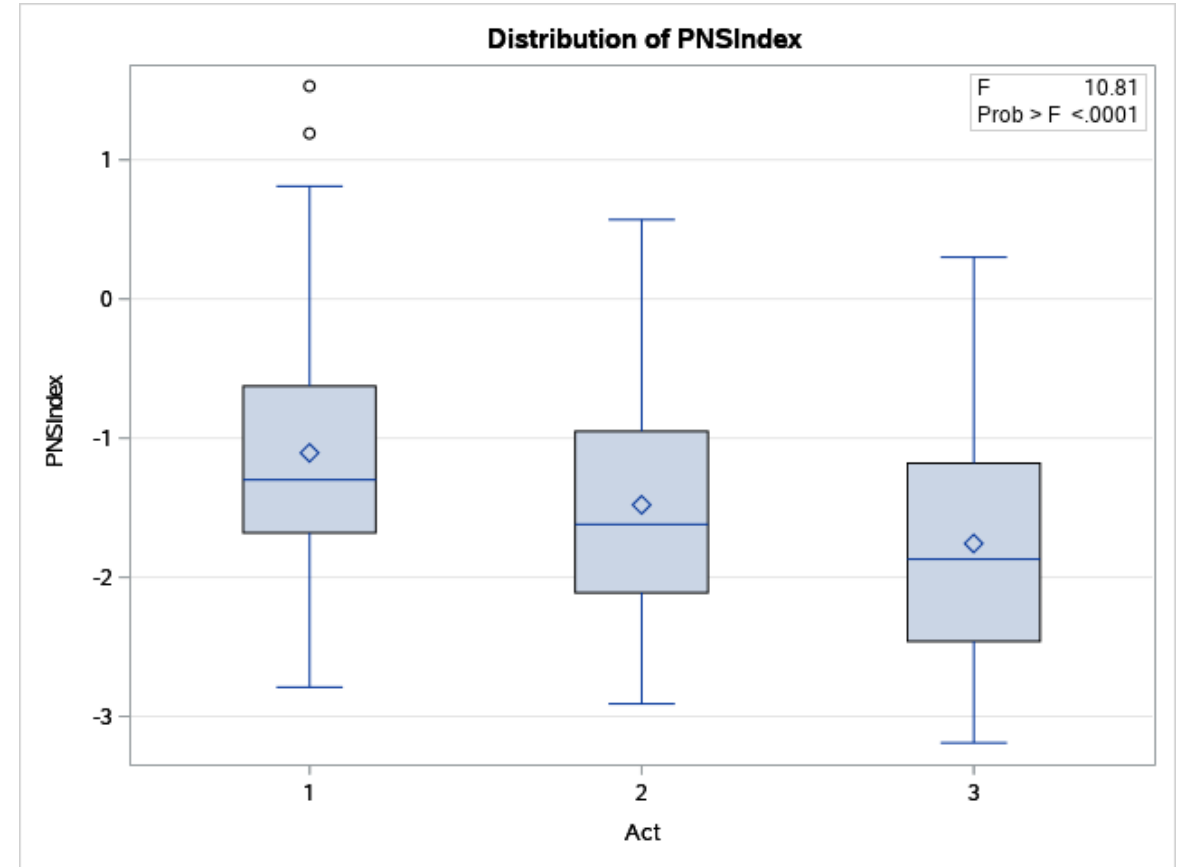
F=13.96 df =2,217 p<0.01

F=12.09 df =2,217 p<0.01

**FOR HUMANS** -- A comparison of non-linear measures showed that the average sympathetic nervous system index increased across the activities, while the average parasympathetic nervous system index decreased.

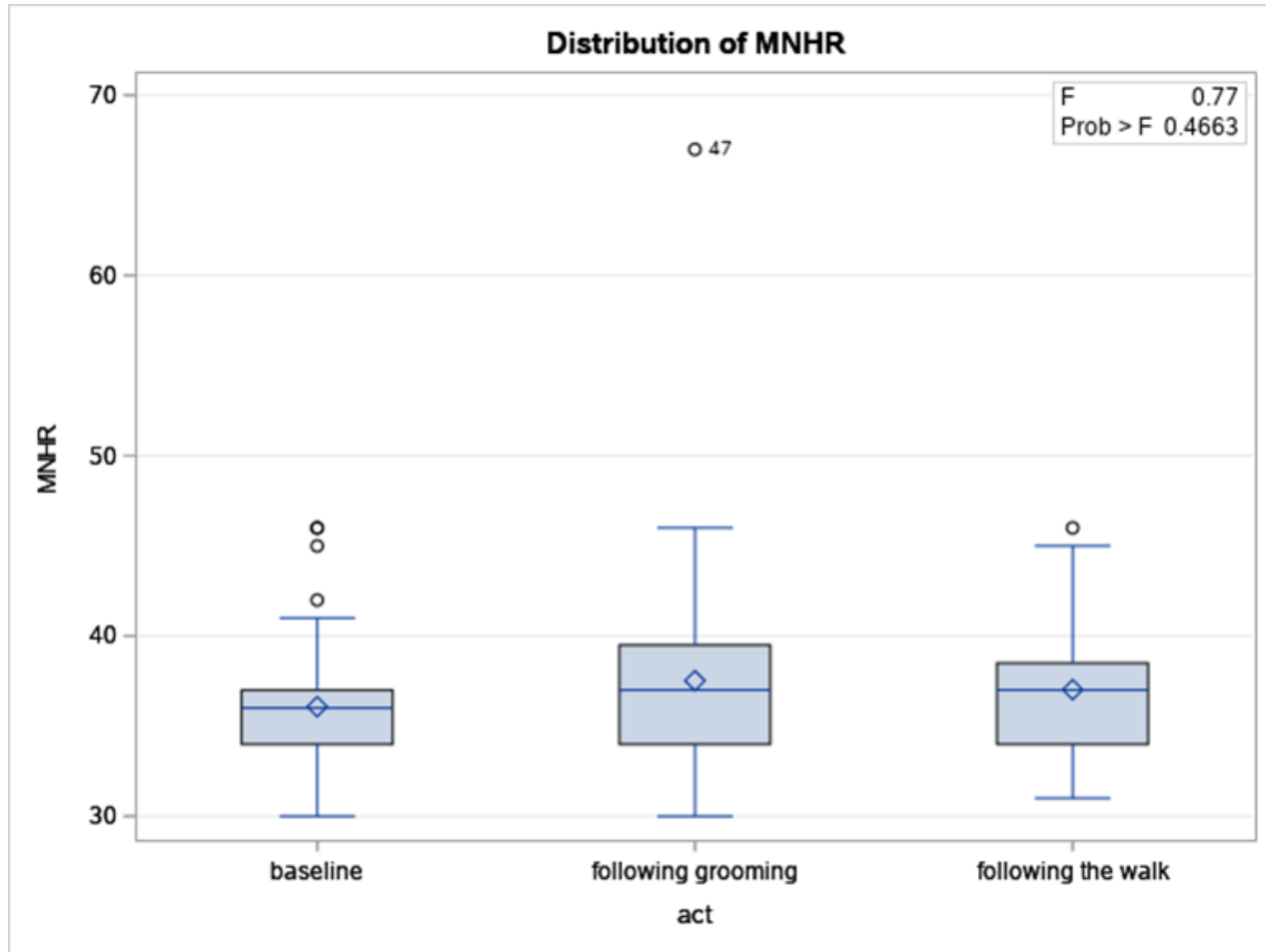


F=6.02 df =2,217 p<0.01

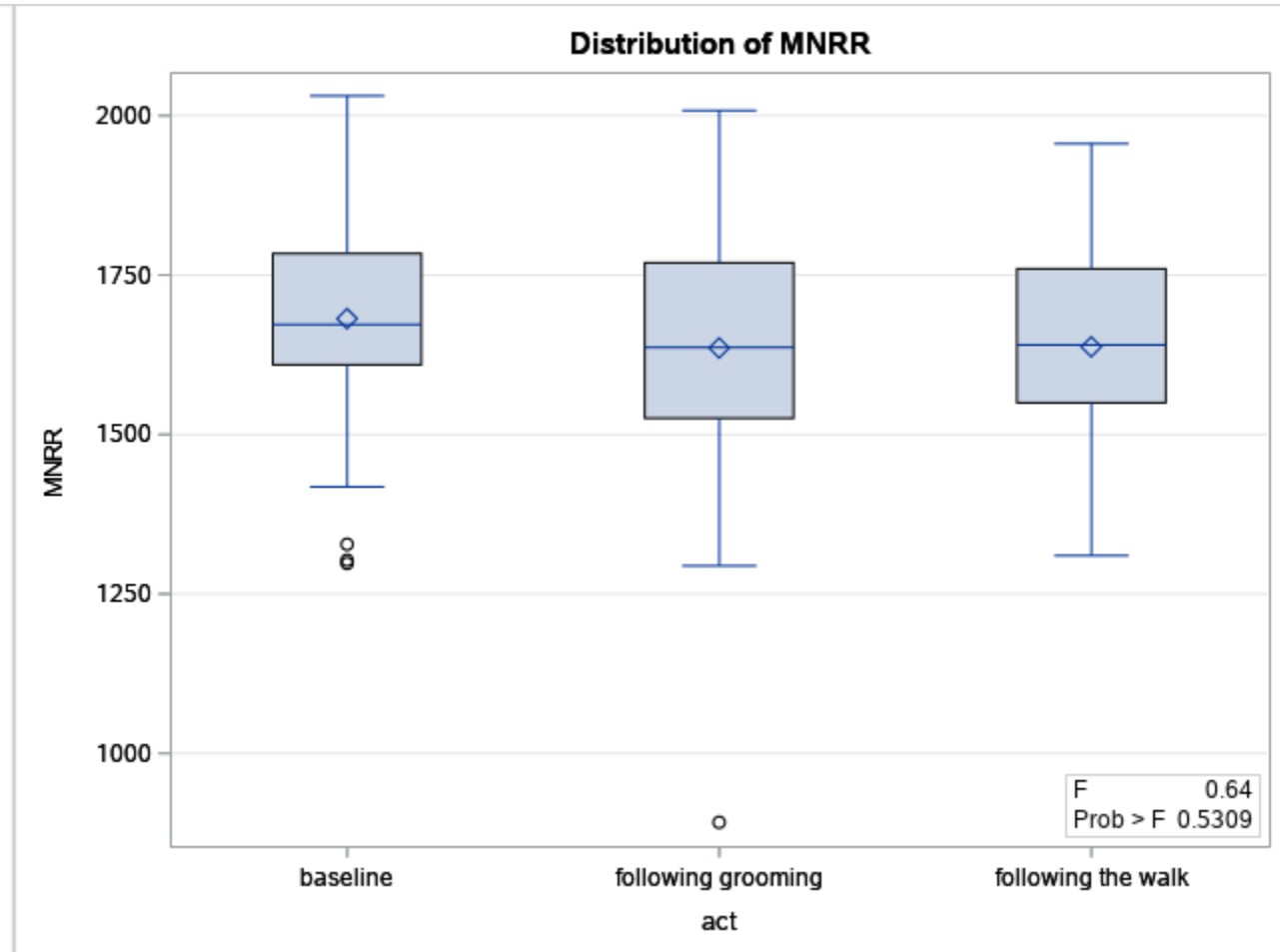


F=10.81 df =2,217 p<0.01

**FOR HORSES** -- No statistically significant differences were observed in the change in HRV measures between baseline, grooming and following the walk. Here we show mean HR and mean R-R interval.

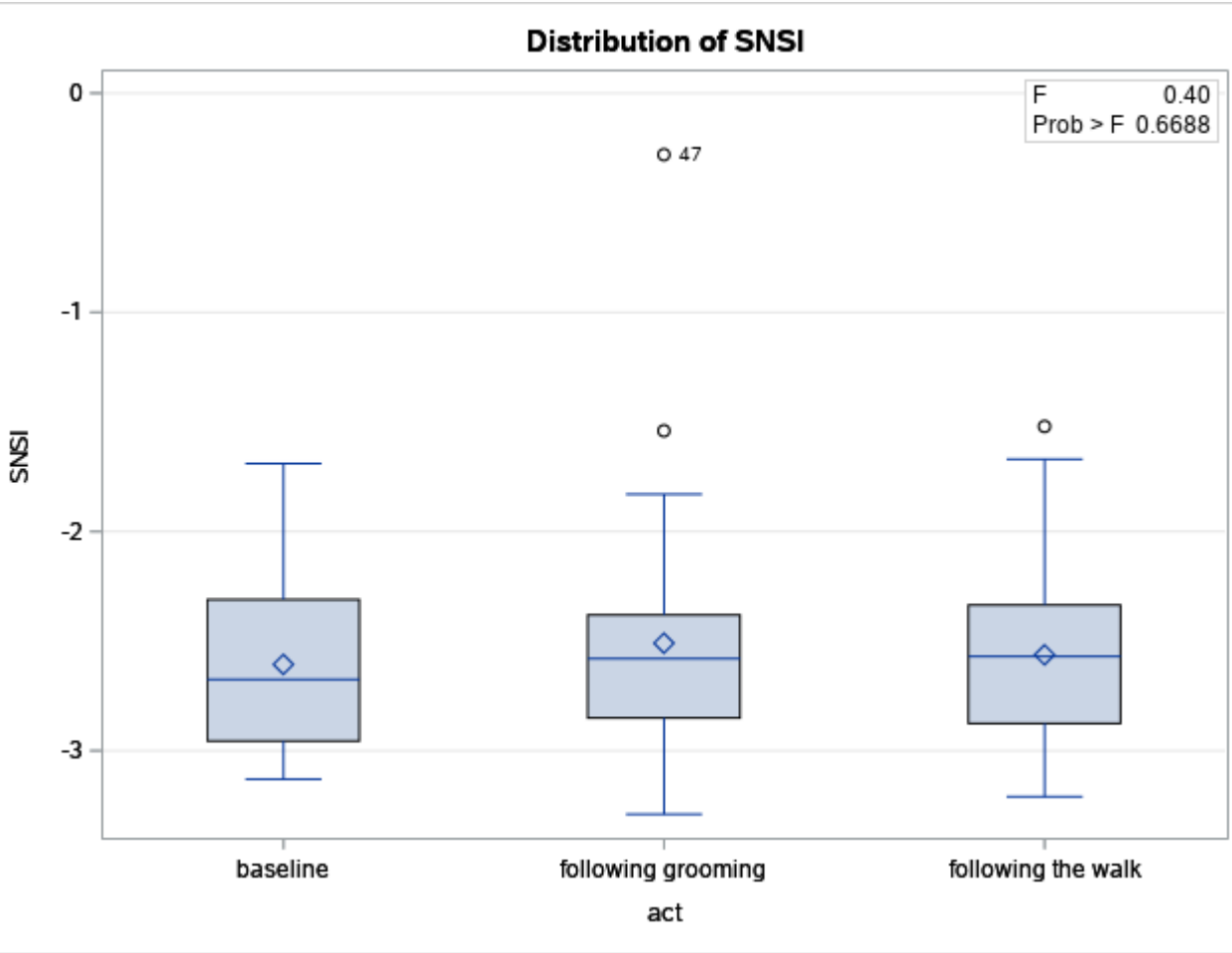


LAM F=0.77 df =2,105 p=0.47

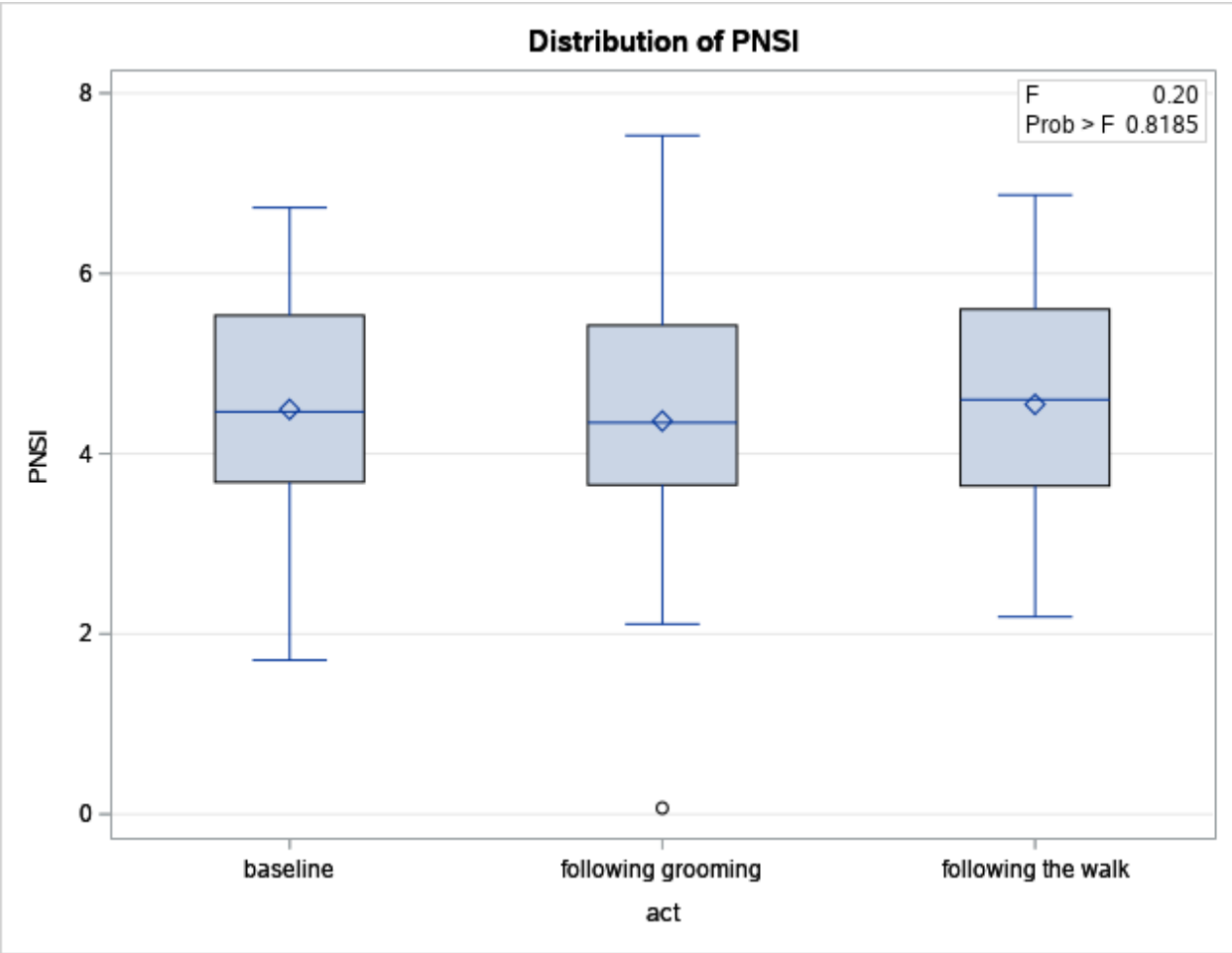


F=0.64 df =2,105 p=0.53

**FOR HORSES** -- No statistically significant differences were observed in the change in HRV measures between baseline, grooming and following the walk. Here we show SNS Index and PNS Index.



LAM F=0.40 df =2,105 p=0.67



F=0.20 df =2,105 p=0.82

# DOES HRV HELP TO EXPLAIN THE COMPLEXITY OF OUR WORK?

- Recall that we are interested in measuring STRESS, and especially the effects of EFP on mitigating stress.
- Up to this point we have reported in standard format the results of our measurements.
- However, there is more to this research!
- One of the ways that we have been exploring these data is to consider the agreement between the measurements from the various independent approaches that use different measurement techniques and scales.
- Can we determine if the different measures agree in their demonstration of stress responses?

How do these measures agree with each other?



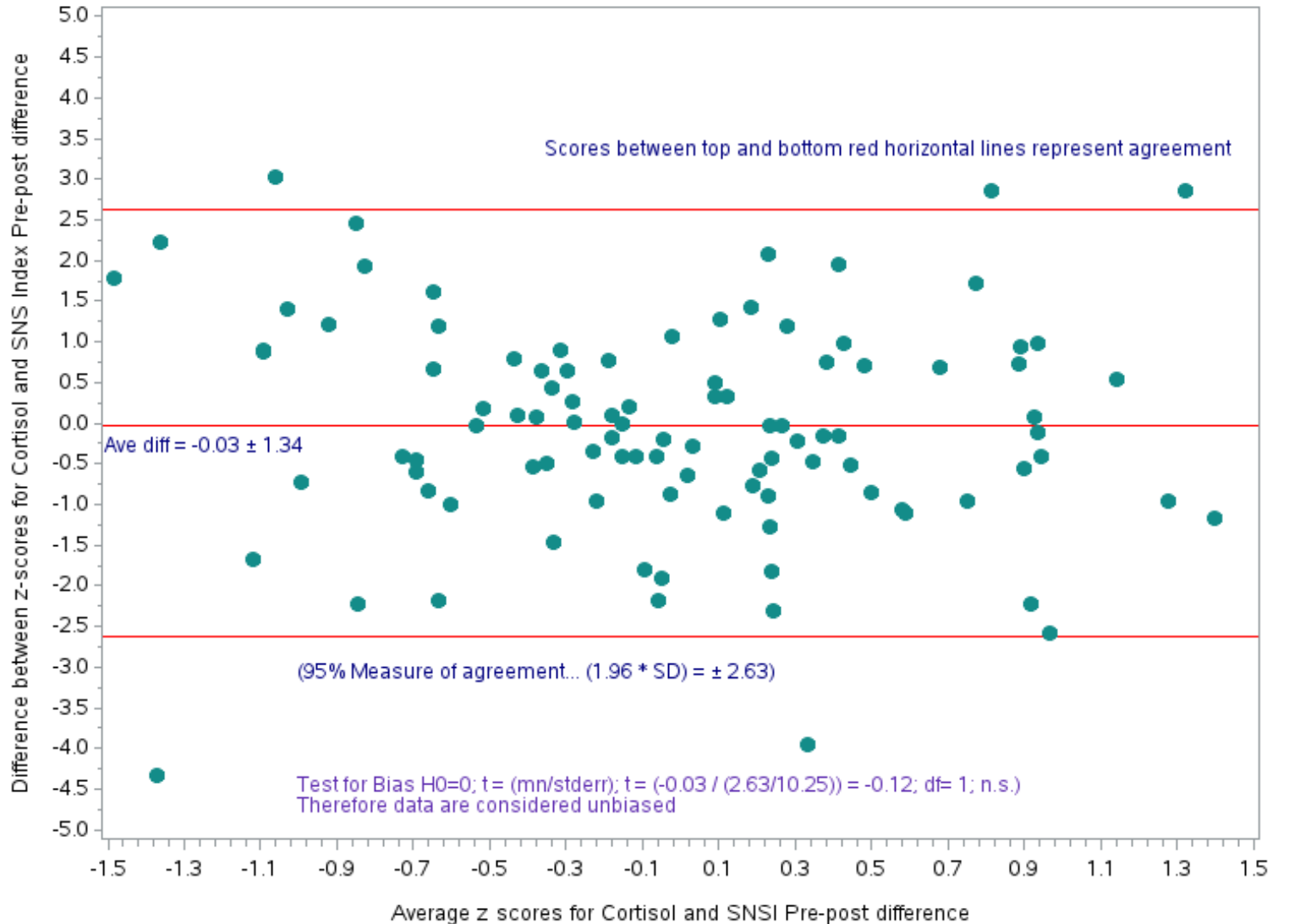


Measures of agreement helped us to show the consistency between stress response measures that use different scales

Here we transformed the data using z scores of the pre to post differences of cortisol and the SNS Index

## Bland Altman Plot of Agreement (Human Participants)

Graph of Pre to Post Activity Change in Cortisol and SNSI



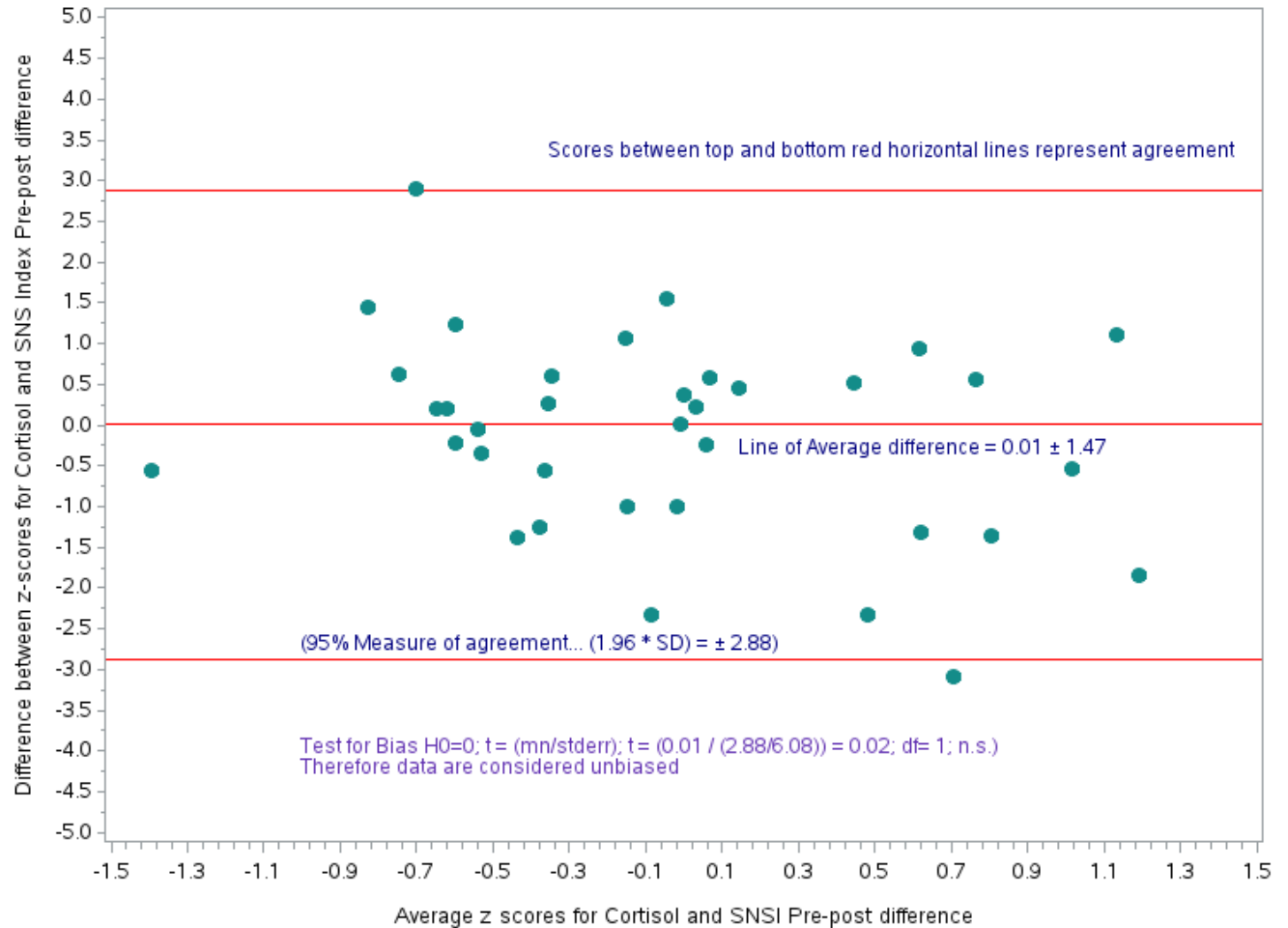


Measures of agreement helped us to show the consistency between stress response measures that use different scales

Here we transformed the data using z scores of the pre to post differences of cortisol and the SNS Index

## Bland Altman Plot of Agreement (Horse Participants)

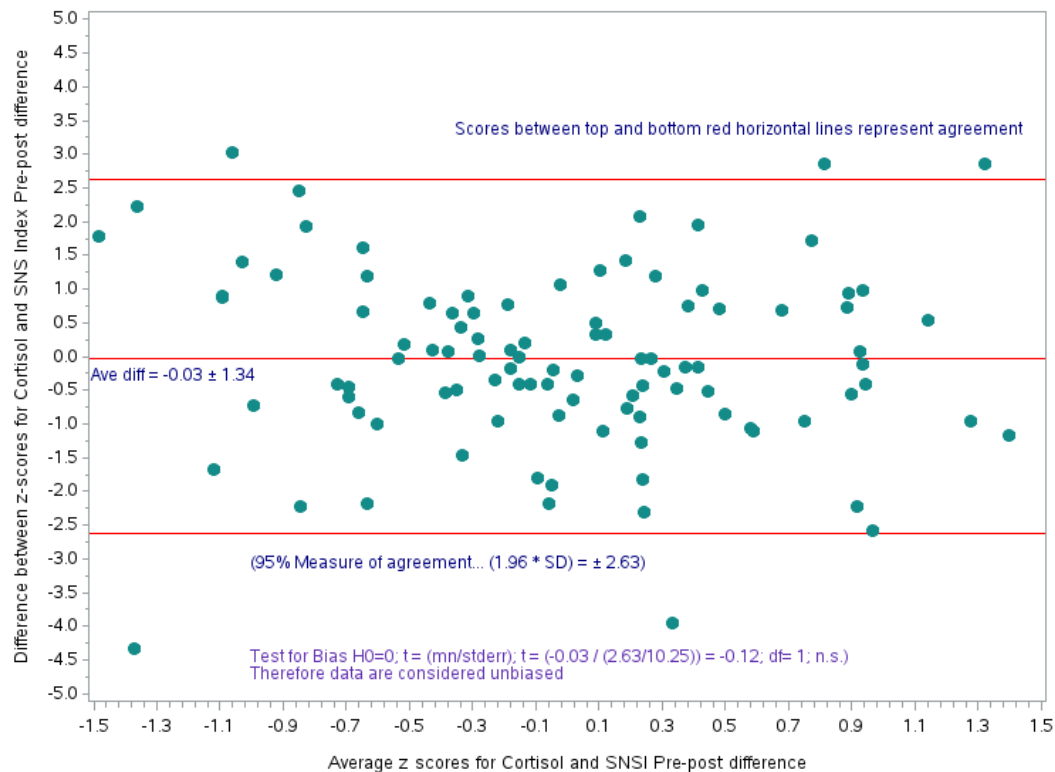
Graph of Pre to Post Activity Change in Cortisol and SNSI



# CONCLUSION IS THAT THERE IS CONSISTENCY ACROSS MEASUREMENT SYSTEMS - *Notice the differences are very close to zero*

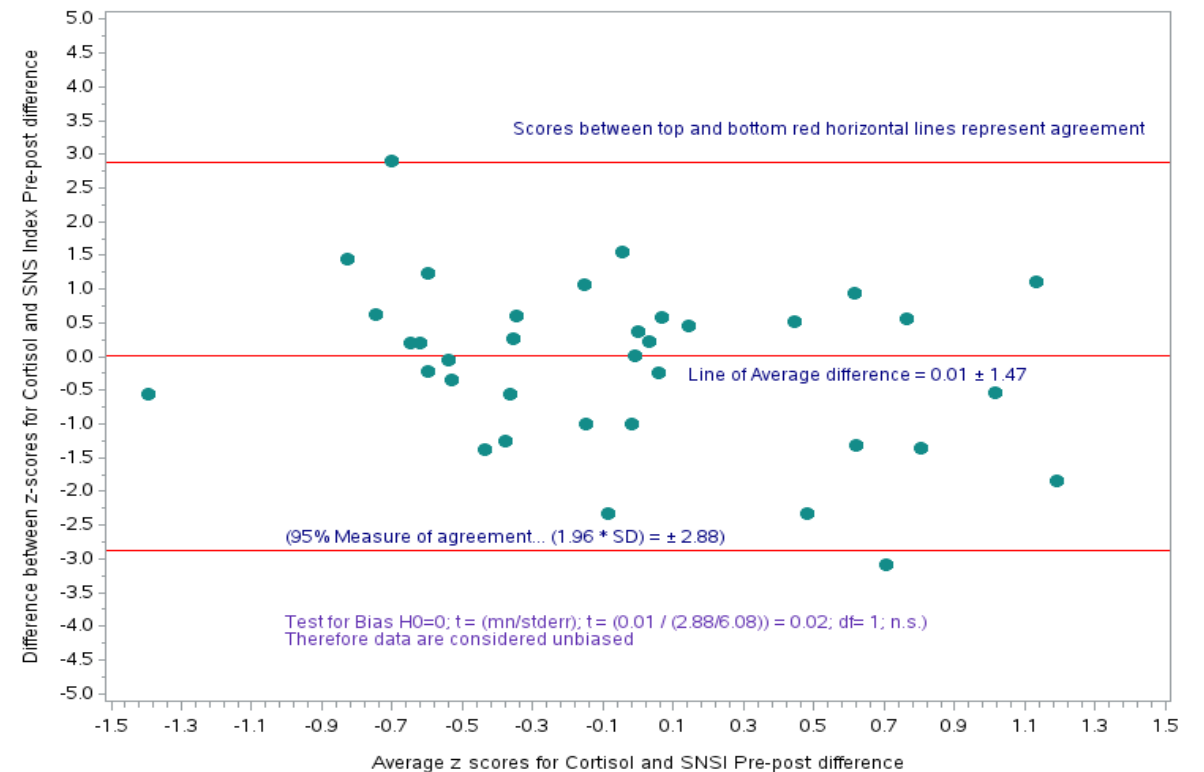
## HUMAN

**Bland Altman Plot of Agreement (Human Participants)**  
Graph of Pre to Post Activity Change in Cortisol and SNSI



## HORSE

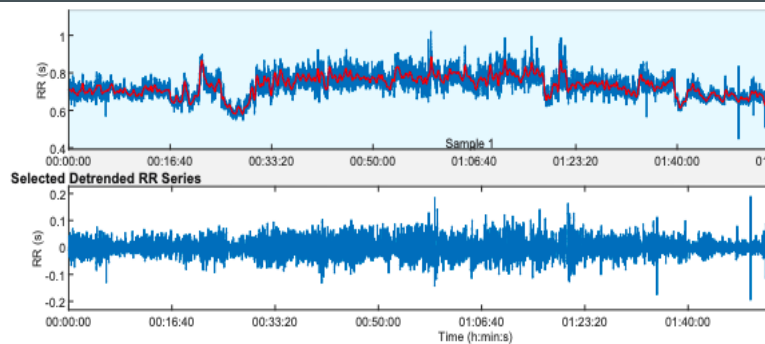
**Bland Altman Plot of Agreement (Horse Participants)**  
Graph of Pre to Post Activity Change in Cortisol and SNSI





# DOES HEARTRATE VARIABILITY CONTRIBUTE TO OUR UNDERSTANDING OF STRESS?

- Heart rate variability is complex because:
  - It provides information about two complementary systems that control heart rate (parasympathetic system and sympathetic system)
  - It can be influenced by both intrinsic (somatic stimuli) and extrinsic (environmental stimuli) factors
  - It provides measures across three different measurement domains: time domain, frequency domain, and the non-linear relationship that expresses entropy – aka the randomness or variance within the measures.
  - **MOST IMPORTANT:** as an objective measure HRV agrees with other objective measures, and supports subjective measures, as demonstrated by our research.



# ACKNOWLEDGEMENTS

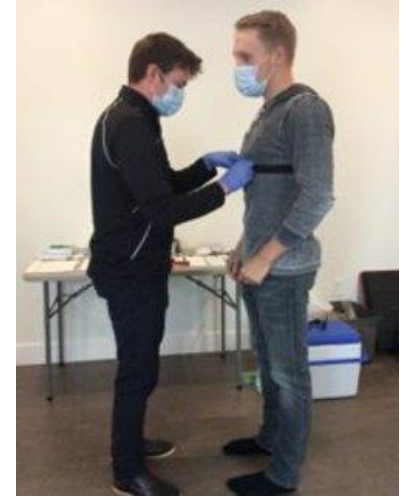
Horses and Humans  
Research  
Foundation: Funding



Serene View  
Ranch Team



UPEI Students



Technical support



Veterans and Horses

**Research Team**

---

QUESTIONS

