

TRH Stimulation Test Instructions

In the fall of 2013, the Equine Endocrinology Group first recommended the thyrotropin-releasing hormone (TRH) stimulation test, which is particularly useful for diagnosis when horses have signs of early PPID or normal resting ACTH concentrations.¹ TRH causes the pituitary gland to release more hormones, and ACTH concentrations increase to a higher level in horses with PPID. This test is easily performed by taking a baseline blood sample, injecting TRH intravenously, and collecting a second blood sample exactly 10 minutes later.² TRH stimulation tests should only be conducted January through June. Previous studies have reported an intermittent frequency of chewing, licking, yawning, flehmen and coughing following the IV administration of TRH in horses.

Procedure:

1. The TRH stimulation test can be conducted after hay is fed, but not within 12 hours after a grain meal.
2. Collect baseline blood sample in a purple-top (EDTA) tube:
 - a. Time 0 (T0 — pre-TRH administration).
3. Administer 0.5 mg (horses <250 kg) or 1.0 mg (horses >250 kg) of TRH intravenously:
 - a. Set cellphone timer for 10 minutes.
4. **Exactly 10 minutes (T10) relative to TRH administration:**
 - a. Collect a second blood sample in a separate purple-top (EDTA) tube.
5. Label purple-top (EDTA) tubes accordingly (T0 or T10).
6. Submit plasma from each time point (T0 and T10) for measurement of ACTH.

Interpretation of ACTH Results

Seasonal Interpretation of Results		PPID unlikely	Equivocal	PPID likely
Baseline ACTH or TRH time 0 (T0) (pg/mL)	Dec – Jun	< 15	15 – 40	> 40
	Jul and Nov	< 15	15 – 50	> 50
	Aug	< 20	20 – 75	> 75
	Sept – Oct	< 30	30 – 90	> 90
10 min (T10) after TRH (pg/mL)	Jan – Jun	< 100	100 – 200	> 200
	Jul – Dec	< 100	TRH stimulation testing should not be used in these months due to potential false-positive results.	

¹Equine Endocrinology Group Recommendations, 2023.

²Goodale L, Hermida P, D'Oench S, Frank N. Assessment of compounded thyrotropin-releasing hormone for the diagnosis of pituitary pars intermedia dysfunction in horses. ACVIM Annual Forum, Seattle, Washington. *J Vet Intern Med* 2013;27:656.