

**THE JOCKEY CLUB
THOROUGHBRED SAFETY COMMITTEE
RECOMMENDATIONS
AUGUST 17, 2008**

Recommendation: TCO₂ Testing

The Thoroughbred Safety Committee is very concerned by the practice of supra-physiologic alkalization, or “milkshaking,” in Thoroughbred racing, and has met with several recognized authorities on the effects of bicarbonate loading in racehorses.

After a review of existing research, a survey of “milkshaking” policies in 29 states, and consultation with the Racing Medication and Testing Consortium (RMTC) on effective control of these alkalinizing substances, the Thoroughbred Safety Committee calls for:

“The adoption of RMTC recommendations on the prohibition of the use of alkalinizing substances by the RCI Model Rules Committee, RCI Board of Directors, and all state racing commissions.”

The attached RMTC recommendations establish uniform threshold levels, pre-race sampling protocols, and effective testing procedures to detect prohibited levels of carbon dioxide in Thoroughbred race horses. The RMTC staff will meet with representatives of the RCI Model Rules Committee and state racing commissions at their earliest convenience to provide further explanations and any necessary clarifications.

Further, as an interim measure, all North American Thoroughbred racetracks should consider implementation of these recommendations by “house rule.”

In addition, the Thoroughbred Safety Committee strongly encourages all state racing commissions to publish the TCO₂ levels of each horse tested to ensure the public that testing is being conducted and participants are below the regulatory thresholds.



RECOMMENDED “BEST PRACTICES” FOR TCO₂ TESTING AND SAMPLE COLLECTION

DEFINITIONS:

Bicarbonate ion – Bicarbonate ion (HCO₃⁻) is a normal constituent of the blood. Bicarbonate is naturally produced by the reaction of carbon dioxide (CO₂) with water (H₂O) to produce carbonic acid (H₂CO₃), which dissociates to a bicarbonate ion and a proton (H⁺). Acid/base metabolism in the body is regulated by this chemical equation:



TCO₂ – The total concentration of all forms of carbon dioxide (Total Carbon Dioxide) in the sample including bicarbonate and carbonate as well as dissolved CO₂. The TCO₂ of the plasma or serum is used for regulatory purposes. The dissolved carbon dioxide is only a small fraction (about 3%) of the total carbon dioxide.

Total CO₂ analyzer – A TCO₂ analyzer adds acid to the plasma or serum sample thereby converting all the carbonate, bicarbonate and carbonic acid to water and CO₂. The concentration of CO₂ is measured represents the total concentration of available CO₂ in the sample or TCO₂.

Measurement Uncertainty (MU) – The measurement uncertainty is a value (with units of concentration) that is determined experimentally and characterizes the variability of the analytical process. The measurement uncertainty is used to eliminate all reasonable variability originating from the measurement process. The measurement uncertainty is a property of the method and is therefore unique to each laboratory unless measures have been taken to standardize methods between laboratories.

Regulatory Threshold – The threshold is the maximum permitted concentration of TCO₂ that has been established by the regulatory body.

Decision Level – The regulatory threshold including the measurement uncertainty is the decision level. For TCO₂ the decision level is the regulatory threshold level plus the measurement uncertainty for the laboratory providing the testing.

REGULATORY THRESHOLD:

The 37 millimoles/L regulatory threshold for TCO₂ is a statistically based threshold designed to identify a state of metabolic alkalosis in horses. Most racing regulators in the U. S. apply a threshold of 37 millimoles of total carbon dioxide per liter of plasma/serum. The decision level for the regulation of TCO₂ is 37 millimoles/L plus the measurement uncertainty.

COLLECTION OF SAMPLES:

Blood samples should be collected 45 minutes (+ or – 15min) pre-race and approximately 3 hours after furosemide administration. The samples must be handled in a consistent manner and cannot be frozen. If samples are obtained pre-furosemide a lower regulatory threshold is necessary and the horses must be kept in a secure detention barn until race time.

Furosemide (Lasix/Salix) produces an elevation in Total Carbon Dioxide averaging approximately 1.7mml/l for a standard 250mg IV dose administered four (4) hours pre-race. The furosemide effect has been considered in the 37mml/l regulatory threshold.

In harness racing, it is necessary to collect samples from horses about 30-45 minutes pre-race. The horse needs to be reasonably well cooled off before being sampled. A horse that is hyperventilating from exercise may produce an artificially low TCO₂.

Horses selected for post race TCO₂ testing should be required to remain in the test barn for a minimum of one and one-half hours before being sampled for TCO₂ testing. This is to permit the TCO₂ value to approach accurate values. Post-race sampling of horses for TCO₂ testing should be discouraged.

INSTRUMENTATION:

Most laboratories doing TCO₂ testing laboratories are utilizing Beckman instruments. Beckman EL-ISE analyzers were used to establish the international threshold for TCO₂ testing. Other TCO₂ instruments may be used for TCO₂ analysis but must be demonstrated to be equivalent and consistent with Beckman equipment.

Blood samples must be processed and tested using standardized, reproducible, validated procedures. Furthermore, test samples submitted for TCO₂ testing should be tested within 120 hours in order to limit sample degradation that results in decreased TCO₂ values.

COMMENTARY:

Regulators should be advised that an elevated TCO₂ concentration is indicative only of metabolic alkalosis and is not proof of the administration of sodium bicarbonate or other alkalinizing substances. For example, the feeding of certain feeds with highly positive dietary cation-anion balances (the DCAB is a measure of the difference between the concentrations of the major cations (primarily sodium and potassium) and the major

anions (primarily chloride and sulfate) in the feed) may cause metabolic alkalosis that is characterized by increased TCO₂ values. Furthermore, the administration of furosemide produces a mild metabolic alkalosis that results in an average increase in the TCO₂ of 1.7 millimoles/L.

Regulators should conduct an investigation to determine the cause of an elevated TCO₂ value, which should include but not necessarily be limited to, the quarantining of the horse and the taking of periodic blood samples.

Trainers should be advised that elevated TCO₂ values, regardless of causation, are violations of the rules and that the penalties for excessive TCO₂ values are severe. Therefore, trainers and their veterinarians should work closely to identify any procedures or practices that may elevate the TCO₂ value.

Split sample analyses can be done with TCO₂ testing but the split sample must be run at the split sample laboratory in parallel with the official sample at the official laboratory in order to avoid delays in testing that result in lower TCO₂ values as a result of sample degradation.