

Topiramate Treatment May Interfere With Urinary Cortisol Measurement By Radioimmunoassay (RIA) - A Case Report

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Topiramate (Topamax™, Janssen-Ortho), a commonly prescribed anti-convulsant agent, is a sulfamate substituted monosaccharide that exerts its action by (i) reducing the frequency of action potentials generated when neurons are subjected to a sustained depolarization, (ii) increasing the activity of type A gamma-aminobutyric acid (GABA-A) receptors, and (iii) antagonizing kainate/alpha-amino-3-hydroxy-5-methylisoxazole-4-propionic acid (AMPA) subtypes of the excitatory glutamate receptor (1,2). Topiramate is readily absorbed in the gut, reaching peak plasma concentrations in 2-3 hours. Absorbed topiramate distributes mainly to body water with a volume of distribution of 0.80-0.55 L/kg. Approximately 18% of the absorbed drug is metabolized into 6 derivatives through hydroxylation, hydrolysis and glucuronidation, while the remaining 82% is eliminated unchanged through the kidneys (2,3). We report here the first case that topiramate treatment may have interfered the measurement of urinary cortisol by radioimmunoassay (RIA).

A 39-yr old female presented with fatigue, mild weight loss, poor appetite and some suspicious striae on the abdomen, and was investigated for possible hypercortisolism using 24-hr urinary free cortisol measured by RIA. The result of 3950 nmol/L or 12,442 nmol/d (Reference Interval or RI: <275 nmol/d) was considered inconsistent with the mild clinical presentation and laboratory findings. The latter included normal plasma cortisol, potassium and bicarbonate, and normal urinary potassium (Table 1). When topiramate was stopped and the urinary cortisol measured again 2

months later, the normal result of 209 nmol/d suggested that topiramate treatment was the likely culprit causing the spurious cortisol result.

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Urinary cortisol was measured by (i) competitive RIA with and without extraction with dichloromethane using the Coat-A-Count kit from Diagnostic Products Corporation, Los Angeles (4), (ii) electrochemiluminescence immunoassay (ECL) by the Roche Elecsys 2010, and (iii) tandem mass spectrometry (MS/MS). Plasma cortisol was measured using the Roche Elecsys 2010. Topiramate (Topamax™) and Tylenol™ tablets were obtained from the Pharmacy Department at the authors' institution, ground, dissolved in phosphate buffer, and centrifuged before analyzed for cortisol by RIA. Tylenol™ tablets served as a matrix control for the cortisol analysis of the topiramate solution. Topiramate.

concentration was determined by liquid chromatography/mass spectrometry (LC/MS).

The measurement of urinary cortisol on the patient's sample using RIA, with and without prior organic extraction, showed similarly elevated results of approximately 4,000 nmol/L. When measured by ECL, the urinary cortisol result was comparable to that obtained by RIA, at 4,841 nmol/L. However, when measured by MS/MS, the cortisol result was 16.6 nmol/L or 52.3 nmol/d, well within the reference interval of 13.8 - 152 nmol/d. This finding strongly suggested that the cortisol results by both RIA and ECL were factitious. This is not surprising, as interferences of the urinary cortisol immunoassays have been described previously (5-7). However, topiramate has never been reported to interfere with the cortisol measurements by immunoassays. To examine if topiramate interfered with the assays directly, topiramate tablets were dissolved in phosphate buffer to generate topiramate solutions of three different concentrations, which were then subjected to the RIA cortisol measurement. At 3,000 µmol/L, topiramate alone gave a cortisol value of 22 nmol/L. As the topiramate concentration increased, so did the cortisol value, reaching 417 nmol/L for the 93,000 µmol/L topiramate solution (Table 1).

Table 1 Laboratory Findings		
Initial Biochemistry Results		
Analyte	Concentration	Reference Interval (RI)
Fasting Glucose	4.2 mmol/L	4.0 – 6.0
Plasma Potassium	3.7 mmol/L	3.5 – 5.0
Plasma Cortisol	255 nmol/L	178 – 625
Plasma Creatinine	74 µmol/L	44 – 106
Plasma Bicarbonate	22 mmol/L	22 – 30
Urine Potassium	17 mmol/L	< 20
Detailed Analysis of the Urine Samples and Topiramate Solutions		
Original Urine Sample (3.15L)	Remarks	
Cortisol (RIA)	3950 nmol/L	Repeated with different dilutions; RI < 275 nmol/d
Cortisol (RIA) – no extraction	(12,443 nmol/d) 4120 nmol/L	
Cortisol (Elecys) – no extraction	4841 nmol/L	
Cortisol (MS/MS)	16.6 nmol/L (523 nmol/d)	RI: 13.8 – 152 nmol/d
Topiramate (LC-MS)	50 µmol/L	
Urine Sample 2 months later		
Cortisol (RIA)	209 nmol/d	Topiramate stopped for 2 months
Topiramate Phosphate Solution		

Cortisol (RIA)	22 nmol/L	3,000 µmol/L topiramate added
Cortisol (RIA)	57 nmol/L	15,500 µmol/L topiramate added
Cortisol (RIA) – no extraction	64 nmol/L	
Cortisol (RIA)	417 nmol/L	93,000 µmol/L topiramate added

In a similarly prepared 4 Tylenol™ solution, cortisol was undetectable, excluding any non-specific effects on the cortisol measurement due to tablet filler or phosphate buffer solution. The apparent dose-dependent effect of topiramate on the cortisol measurement points to possible cross-reactivity of topiramate with the cortisol antibodies used in these assays. To assess the specificity of this topiramate effect, we ran the 15,500 µmol/L topiramate solution through the Elecsys 2010 for total testosterone, chorioembryonic antigen (CEA), and CA125. All three immunoassays showed results at or below the detection limit (results not shown).

Having shown that topiramate interfered with the cortisol measurement directly and specifically, we then sought to confirm the presence of topiramate in the patient's urine sample, and whether the amount of topiramate present would be sufficient to explain the discrepancy observed. Using LC/MS, the original urine sample that gave a cortisol value of 3,950 nmol/L was shown to contain only 50 µmol/L of topiramate. On a molar basis, it is difficult to imagine how such a low concentration of topiramate would generate the dramatic increase in the cortisol result observed. It is possible that topiramate metabolites, rather than the parent compound, are responsible for the assay interference. Unfortunately, measuring topiramate in a selected-ion-monitoring mode, the LC/MS method would not have been able to detect, if any, topiramate metabolites present. Nevertheless, the fact that cessation of topiramate treatment was associated with a normal urinary cortisol by RIA strongly implicated a role of the initial topiramate treatment in causing the factitious urinary cortisol

result by either RIA or ECL. Further studies determining the cross-reactivity of topiramate metabolites in cortisol assays based on RIA and ECL will be helpful in elucidating the source and the mechanism of this interference.⁵

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