Clinical Utility of an LC-MS/MS Seizure Panel for Common Drugs Involved in Drug-Induced Seizures

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BACKGROUND: Approximately 6% of new-onset seizures are drug-related, but there is currently no reliable way to determine if a seizure is drug-induced. Liquid chromatography–tandem mass spectrometry (LC-MS/MS) is a powerful tool that allows simultaneous detection of numerous analytes of diverse chemical nature in patient samples. This allows a single analysis to incorporate many compounds relevant to a particular clinical presentation, such as suspected drug-induced seizures. We investigated whether results from a seizure panel using LC-MS/MS could affect patient care.

METHODS: We developed a semiquantitative LC-MS/MS assay to detect 12 chemically diverse drugs implicated in drug-related seizures. We collected leftover serum and plasma samples from patients who had seized, performed solid-phase extraction, and analyzed the samples using a hybrid triple quadrupole/linear ion trap mass spectrometer. After assembling a team of medical and toxicology experts, we developed and used a scoring system to determine whether the results of the seizure panel would have affected patient treatment in each case where a drug was detected.

RESULTS: In an analysis of 157 samples from patients who seized, 17 (11%) were found to be positive for a drug on the seizure panel. The team of experts determined that the test results probably or definitely would have affected treatment in 7 (41%) of these cases.

CONCLUSIONS: A test that detects the presence of drugs implicated in drug-induced seizures can help physicians determine if an unexplained seizure is drug-related and thus potentially better direct patient care. Additionally, LC-MS/MS is an effective tool for answering clinically driven questions.

Traditional toxicological testing strategies group analytes by their chemical nature, which dictates the method of analysis. However, clinical questions are not constrained by the chemical nature of the analytes in question. Furthermore, each traditional chromatographic method has its limitations. Liquid chromatography with spectrophotometric detection has limited detection capability, and gas chromatography mass spectrometry often requires labor-intensive derivatization and is not adept at detecting polar compounds. Regardless of the methodology, comprehensive drug screens that are designed to detect hundreds of diverse compounds have compromised utility for detecting compounds (1, 2). Methods that are targeted to a narrower menu of drugs can be more analytically robust and can supply semiquantitative information that may aid in patient care. A single test that can detect numerous chemically diverse compounds that are relevant to a particular clinical presentation would represent a new paradigm in toxicological testing. Liquid chromatography–tandem mass spectrometry (LC-MS/MS), with its low detection limits and great flexibility in the types of analytes it can detect, is well suited to this new, clinically driven approach. To evaluate the potential of this paradigm, we addressed the clinical problem of determining whether a seizure was drug-related using a seizure panel.

Many seizures in patients presenting to emergency departments (EDs) have unexplained causes. Studies have estimated that 6% of new-onset seizures and up to 9% of status epilepticus cases are due to drug toxicity

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Nonstandard abbreviations: LC-MS/MS, liquid chromatography–tandem mass spectrometry; ED, emergency department; MDMA, 3,4-methylenedioxymethamphetamine; IS, internal standard; SRM, selected reaction monitoring.
(3, 4); however, drug screens typically do not include the majority of drugs that can cause seizures. The California Poison Control System has conducted several studies on drug-related seizures (5, 6). A retrospective study of 529 cases associated with seizures in California in 2003 revealed 386 seizures to be drug-related (7). The top 12 drugs that accounted for approximately 75% of all drug-related seizures were bupropion, citalogren, cocain, diphenhydramine, iloniazid, lamotrigine, methamphetamine, 3,4-methylenedioxyamphetamine (MDMA), quetiapine, tiagabine, tramadol, and venlafaxine. As proof of principle for laboratory testing driven by clinical problems, we designed a seizure panel using LC-MS/MS to detect these 12 drugs. Our objective was to use this method to test patient samples and perform a retrospective chart review to determine if a seizure panel would have improved patient care.

Materials and Methods

PATIENT SAMPLES

Leftover serum and plasma samples from patients presenting to the ED with a seizure were collected and refrigerated for <1 week before being tested. Assay results were not revealed to patients or health care providers. This work was approved by the University of California–San Francisco Institutional Review Board, which determined that patient consent was not necessary.

STANDARDS AND REAGENTS

We purchased LC-MS–grade water, acetonitrile, methanol, dichloromethane, formic acid, isopropanol, and ammonium formate from Fisher; Oasis HLB cartridges from Waters; all standards, including deuterated diphenhydramine and methamphetamine, from Cerilliant; and drug-free serum and plasma from Biological Specialty Corp.

SAMPLE PREPARATION

We added 25 μL of a 4 mg/L methanolic solution of the internal standards (ISs) deuterated diphenhydramine and methamphetamine (final concentration 98 μg/L) to 1 mL serum or plasma. These ISs were chosen for their strategic placement within the range of retention times. The solution was vortex-mixed, loaded onto an Oasis HLB cartridge that had been preconditioned with 3 mL methanol and 3 mL water, and then washed with 3 mL water and 3 mL 90% methanol by volume. Elution was performed using 3 mL dichloromethane: isopropanol (75:25 by volume) containing 2% formic acid. Extracts were evaporated to dryness and then reconstituted in 100 μL 2 mmol/L ammonium formate, pH 3.0.

LIQUID CHROMATOGRAPHY

For LC, we used an Agilent 1200 series and a Waters XTerra MS C18, 3.5-μm (100 × 2.1 mm) column, maintained at 25 °C, with a gradient of mobile phase A (0.5 mmol/L ammonium formate, pH 3.0) and mobile phase B (acetonitrile:10 mmol/L ammonium formate, pH 3.0; 90:10 by volume). The flow rate was 300 μL/min and the program was 0–0.5 min, 0% B; 0.5–1 min, 0–20% B; 1–6.5 min, 20%–60% B; 6.5–6.6 min, 60%–100% B; 6.6–8 min, 100% B; 8–8.1 min, 100%–0% B; 8.1–10 min, equilibrium with 0% B. The total analysis time was 10 min.

MASS SPECTROMETRY

We used a hybrid triple quadrupole/linear ion trap (LIT) mass spectrometer (Applied Biosystems 3200 QTRAP) equipped with a Turbolon-Spray ionization source. To optimize mass spectrometer settings, we used quantitative optimization in Analyst 1.4.2 software. Compounds were infused with a syringe pump at 20–100 μg/L in 50% mobile phase A and 50% mobile phase B. Positive ionization was performed with the following settings: ion spray voltage, 3500 V; curtain gas, 40; collision gas, medium; ion source gas 1 and 2 at 55 and 50 U, respectively; interface heater, on; and source temperature, 650 °C. We used selected reaction monitoring (SRM) as a survey scan, followed by a product ion scan that was triggered under certain conditions, in a feature called information-dependent acquisition (IDA). In SRM, 14 precursor-product ion pairs were scanned; the total scan time was 2.17 s. Q1 and Q3 were set at unit and low resolution, respectively; dynamic fill time was used. Under IDA conditions, the 3 most intense ions in the SRM spectrum >2000 cps at any point were selected for product ion analysis. Each ion could be selected in up to 3 occurrences before being excluded from analysis for 30 s. The product ion scan conditions were as follows: declustering potential 20 V, entrance potential 10 V, and the dynamic fill time used. The collision energy was set at 20 V with a spread of 15 V so that fragments were generated at 5, 20, and 35 V. These fragments were collected in the LIT and analyzed together by scanning from 50 to 400 m/z at a rate of 1000 atomic mass units (amu)/s with a scan time of 0.61 s. The source parameters were unchanged.

QUANTITATIVE AND QUALITATIVE IDENTIFICATION

The seizure panel was designed to be semiquantitative. With every batch of patient samples, we extracted and ran a blank (drug-free serum) and 3 calibrators at 25, 50, and 100 μg/L or 50, 100, and 500 μg/L in drug-free serum. For this preliminary method, the cutoff for positivity was set at or just below the concentration of the lowest calibrator used in the batch, either 20 or 50 μg/L.
These cutoffs applied to all of the drugs except isoniazid and lamotrigine, which had cutoffs of 500 μg/L because of their higher therapeutic concentrations in plasma (8). For all controls and calibrators, isoniazid and lamotrigine were present at 10-fold higher concentrations. In each batch of patient samples, we also included an aliquot from a frozen quality control pool consisting of all compounds at 50 μg/L in drug-free serum. For qualitative identification of the drugs, we performed a library search in Analyst 1.4.2 software; a purity of ≥70% between the unknown and library spectrum was required for a match.

ANALYTICAL VALIDATION
We tested extraction recovery by spiking the compounds into drug-free serum at 100 and 1000 μg/L in duplicate. After extraction, we compared the peak areas to peak areas of compounds spiked into extracts of drug-free serum. We tested linearity by spiking the compounds into drug-free serum at 0, 50, 200, 500, 2000, and 5000 μg/L and added IS to all samples as in a patient sample. We plotted the ratio of analyte peak area to IS peak area against the ratio of analyte concentration to IS concentration and used linear regression with 1/x weighting. We reviewed curve-fit parameters for the calibration curve of each run and tested between-run (n = 5) and within-run (n = 6) imprecision for all compounds at 50 μg/L. We checked ion suppression by infusing a 25 μg/L mixture of the compounds at 5 μL/min in 50% mobile phase A and 50% mobile phase B. At the same time, extracted drug-free serum was injected into the chromatographic system and the 2 flows were merged using a polyetheretherketone (PEEK) tee before the source entrance, in a postcolumn infusion system described elsewhere (9). Patient plasma samples were treated the same as serum samples, although plasma was not validated for this preliminary method.

CLINICAL UTILITY
We assembled a team of 3 medical and toxicology experts to review the medical records of patients with positive samples and determine whether the cause of the seizure was related to the drug detected and whether the test result would have been clinically useful to the health care provider managing the case. The team included an ED physician, a physician toxicologist, and a pharmacist (PharmD) toxicologist from the California Poison Control System. Charts were reviewed to determine the likelihood that the reported seizure was actually a seizure and not a fall or syncope—whether the seizure was witnessed was one of the criteria used. The seizure likelihood was designated A for “definitely a seizure,” B for “probably a seizure,” or C for “possibly a seizure.” Next, the team decided on 5 criteria to determine if the seizure was related to the drug found via LC-MS/MS screening. The criteria were adapted from the Naranjo Adverse Drug Reaction Probability Scale (10) and are listed in Table 1. Positive points were assigned in situations that suggested the seizure was likely to be drug-related; no points or negative points were assigned when the drug was not implicated in the seizure. All rankings were done by consensus of the team. The 5 criteria, taken together, create a range of points from −3 to +7. The team decided that in cases with a total score of <2 points, it was doubtful that the seizure was related to the drug detected. In cases with the maximum of 7 points, the seizure was determined to be definitely drug-related. Categories of “possibly drug-related” (2–4 points) and “probably drug-related” (5–6 points) were also designated.

The team’s final evaluation involved judging the usefulness of the test. Each of the 17 cases was reviewed to evaluate the test result’s potential impact on patient care. Two hypothetical situations were evaluated: first, the impact on acute care in the ED if the results of the seizure panel had been available with a short turnaround time (<4 h), and second, the impact on long-term care provided by a primary care physician if the test results had been available with a longer turnaround time (≥24 h). The test result for each case was judged as No (not useful), ED1 (possibly useful in the ED), ED2 (probably useful in the ED), ED3 (definitely useful in the ED), LT1 (possibly useful in the long term), LT2 (probably useful in the long term), and LT3 (definitely useful in the long term) (Table 2).

Results
Extraction recovery was between 60% and 130% for all compounds except isoniazid, which had a recovery of 15% due to its polar nature. Isoniazid was still easily detectable at its cutoff concentration despite this low recovery. The signal-to-noise ratio was always detectable at its cutoff concentration despite this low recovery. The signal-to-noise ratio was always detectable at its cutoff concentration despite this low recovery. The signal-to-noise ratio was always detectable at its cutoff concentration despite this low recovery. The signal-to-noise ratio was always detectable at its cutoff concentration despite this low recovery.
# Table 2. Cases in which drugs were detected in samples from patients who had seized.

<table>
<thead>
<tr>
<th>Case no.</th>
<th>Age, years</th>
<th>Sex</th>
<th>Relevant patient history</th>
<th>Drug detected, μg/L</th>
<th>Therapeutic level of drug detected?</th>
<th>Previous reports of seizure with drug?</th>
<th>Did seizure occur after drug taken?</th>
<th>Absence of alternate causes?</th>
<th>Drug detected at toxic levels?</th>
<th>Seizure confirmed by other evidence?</th>
<th>Was seizure drug-related?</th>
<th>Score</th>
<th>Would test results have impacted patient care?</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>115</td>
<td>55</td>
<td>M</td>
<td>Witnessed fall, possible sz; 3 syncope episodes in 3 days; history CVA, sz disorder; low phenytoin and VPA levels</td>
<td>Cocaine, 20</td>
<td>NA</td>
<td>C</td>
<td>1</td>
<td>2</td>
<td>−2</td>
<td>1</td>
<td>1</td>
<td>Possibly 3</td>
<td>LT1, possibly switch to clonazepam for cocaine user</td>
<td></td>
</tr>
<tr>
<td>70</td>
<td>37</td>
<td>M</td>
<td>Witnessed grand mal tonic clonic sz, 3 min; history sz disorder since head injury 1999; low phenytoin (last dose 3 days ago)</td>
<td>Methamphetamine, 60</td>
<td>NA</td>
<td>B</td>
<td>1</td>
<td>2</td>
<td>−2</td>
<td>1</td>
<td>1</td>
<td>Possibly, 2</td>
<td>No, would still restart phenytoin</td>
<td></td>
</tr>
<tr>
<td>122</td>
<td>26</td>
<td>M</td>
<td>Friends presumed sz after fight; history anxiety, 1 sz; previously on phenytoin</td>
<td>Methamphetamine, 40</td>
<td>NA</td>
<td>C</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>Probably, 6</td>
<td>ED2, LT2, don’t restart on phenytoin</td>
<td></td>
</tr>
<tr>
<td>161</td>
<td>36</td>
<td>F</td>
<td>Unwitnessed sz, patient called paramedics confused; history sz, but none &gt;1.5 yrs; history drug abuse, noncompliant with sz meds</td>
<td>Methamphetamine, 220</td>
<td>NA</td>
<td>B</td>
<td>1</td>
<td>2</td>
<td>−1</td>
<td>1</td>
<td>0</td>
<td>Possibly, 3</td>
<td>LT1, consider restart phenytoin but maybe not because patient noncompliant with prescriptions</td>
<td></td>
</tr>
<tr>
<td>171</td>
<td>49</td>
<td>M</td>
<td>Jailed that day; multiple witnessed tonic clonic sz; long history nonepileptic sz; previously controlled by valium</td>
<td>Methamphetamine, 110</td>
<td>NA</td>
<td>B</td>
<td>1</td>
<td>2</td>
<td>−2</td>
<td>1</td>
<td>1</td>
<td>Possibly, 3</td>
<td>LT1, would restart on an antiepileptic</td>
<td></td>
</tr>
<tr>
<td>21</td>
<td>74</td>
<td>M</td>
<td>1.5-min tonic clonic witnessed by paramedics; history stroke, diabetes, Parkinson disease; vomited after lunch, hypoglycemic; head CT negative</td>
<td>Citalopram, 40</td>
<td>Yes</td>
<td>A</td>
<td>1</td>
<td>2</td>
<td>−2</td>
<td>0</td>
<td>0</td>
<td>Doubtful, 1</td>
<td>No, would treat hypoglycemia</td>
<td></td>
</tr>
<tr>
<td>141</td>
<td>29</td>
<td>M</td>
<td>Witnessed 45-s tonic clonic sz; history alcohol abuse; last drink 24 h ago; no history detox or alcohol withdrawal sz</td>
<td>Venlafaxine, 170</td>
<td>Yes</td>
<td>A</td>
<td>1</td>
<td>2</td>
<td>−2</td>
<td>0</td>
<td>0</td>
<td>Doubtful, 1</td>
<td>LT2, venlafaxine is confounding cause of seizure, change to drug that does not lower seizure threshold since patient is alcoholic</td>
<td></td>
</tr>
<tr>
<td>68</td>
<td>24</td>
<td>M</td>
<td>Witnessed sz while playing soccer; no postictal state, could be syncopic; 1 sz in childhood history depression</td>
<td>Bupropion, 30</td>
<td>No, less</td>
<td>C</td>
<td>1</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>Possibly, 4</td>
<td>LT2, consider stopping bupropion; important to know patient was taking it during sz</td>
<td></td>
</tr>
<tr>
<td>31</td>
<td>21</td>
<td>M</td>
<td>Unwitnessed sz in shower; hit head; history autism, epilepsy; 1st sz in 3 y</td>
<td>Lamotrigine, 4200</td>
<td>Yes</td>
<td>C</td>
<td>1</td>
<td>2</td>
<td>−2</td>
<td>0</td>
<td>0</td>
<td>Doubtful, 1</td>
<td>LT1, helpful to know that lamotrigine is in the low therapeutic range; change drug or increase dose?</td>
<td></td>
</tr>
<tr>
<td>79</td>
<td>22</td>
<td>F</td>
<td>Sz witnessed by mother; heavy alcohol use night before, missed a.m. dose of lamictal; 2 sz in morning 10y history epilepsy, sz when off lamotrigine</td>
<td>Lamotrigine, 4100</td>
<td>Yes</td>
<td>A</td>
<td>1</td>
<td>2</td>
<td>−2</td>
<td>0</td>
<td>0</td>
<td>Doubtful, 1</td>
<td>ED1, tell parent to take lamotrigine; evidence that level was on the low end of therapeutic; LT2, consider raising dose, discuss compliance</td>
<td></td>
</tr>
</tbody>
</table>

Continued on page 130
Table 2. Cases in which drugs were detected in samples from patients who had seized. (Continued from page 129)

<table>
<thead>
<tr>
<th>Case no.</th>
<th>Age, years</th>
<th>Sex</th>
<th>Relevant patient history</th>
<th>Drug detected, µg/L</th>
<th>Therapeutic level of drug detected?</th>
<th>Was it a true seizure?</th>
<th>Previous reports of seizure with drug?</th>
<th>Did seizure occur after drug taken?</th>
<th>Absence of alternate causes?</th>
<th>Drug detected at toxic levels?</th>
<th>Seizure confirmed by other evidence?</th>
<th>Was seizure drug-related?, score</th>
<th>Would test results have impacted patient care?, notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>113</td>
<td>31</td>
<td>M</td>
<td>Witnessed sz, 2 min; CT negative; history sz in context of drug intoxication or withdrawal; history schizoaffective, polydipsic, admitted to psychiatry</td>
<td>Quetiapine, 2280</td>
<td>No, 8 times more</td>
<td>B</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>Definitely, 7</td>
<td>ED2, admit to monitored bed instead of psychiatry; LT3, monitor better in institution</td>
</tr>
<tr>
<td>62</td>
<td>58</td>
<td>F</td>
<td>Twitching in sleep, altered mental status in ED, improved with naloxone, worsened by benzodiazepines in nursing facility; no history sz</td>
<td>Diphenhydramine, 50; isoniazid, 530</td>
<td>No, less (both)</td>
<td>C</td>
<td>1</td>
<td>2</td>
<td>-2</td>
<td>0</td>
<td>0</td>
<td>Doubtful, 1</td>
<td>ED2, rule out isoniazid OD; LT2, consider raising isoniazid dose because of low level</td>
</tr>
<tr>
<td>162</td>
<td>37</td>
<td>F</td>
<td>Witnessed sz, 1–2 min; history alcohol withdrawal sz &lt;4; last alcohol night before</td>
<td>Diphenhydramine, 20</td>
<td>No, less</td>
<td>A</td>
<td>1</td>
<td>2</td>
<td>-2</td>
<td>0</td>
<td>0</td>
<td>Doubtful, 1</td>
<td>No, would treat alcohol withdrawal</td>
</tr>
<tr>
<td>190</td>
<td>50</td>
<td>F</td>
<td>Witnessed 30 min generalized tonic-clonic sz in ED; renal failure; hypokalemic; head CT negative</td>
<td>Diphenhydramine, 70</td>
<td>No, less</td>
<td>A</td>
<td>1</td>
<td>2</td>
<td>-2</td>
<td>0</td>
<td>0</td>
<td>Doubtful, 1</td>
<td>No, would treat hypokalemia</td>
</tr>
<tr>
<td>178</td>
<td>38</td>
<td>F</td>
<td>3 sz, responsive throughout; history sz off phenytoin for 3 weeks</td>
<td>Diphenhydramine, 30</td>
<td>No, less</td>
<td>C</td>
<td>1</td>
<td>2</td>
<td>-2</td>
<td>0</td>
<td>0</td>
<td>Doubtful, 1</td>
<td>No, would restart phenytoin</td>
</tr>
<tr>
<td>185</td>
<td>54</td>
<td>M</td>
<td>Passed out in shelter; synapse of szs; history advanced dementia; CT and MRI of brain negative</td>
<td>Diphenhydramine, 30</td>
<td>No, less</td>
<td>C</td>
<td>1</td>
<td>2</td>
<td>-1</td>
<td>0</td>
<td>0</td>
<td>Possibly, 2</td>
<td>ED1, negative result so consider phenytoin for new-onset sz disorder; LT1, negative result so consider full sz workup or monitor patient</td>
</tr>
<tr>
<td>105</td>
<td>20</td>
<td>F</td>
<td>Sz while driving, accident; no trauma; blue emesis; history drug OD, depression; no history sz; CT negative; later determined OD; “6–8 sleeping pills” and fight with friends/parents</td>
<td>Diphenhydramine, 700</td>
<td>No, 9 times more</td>
<td>A</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>Definitely, 7</td>
<td>ED1, treat OD, order APAP, GI decontamination, charcoal, NG tube, cardiac monitor; LT3, obviates full seizure workup</td>
</tr>
</tbody>
</table>

Notes:
- sz, seizure; CVA, cerebrovascular accident; VPA, valproic acid; NA, not applicable; CT, computed tomography; OD, overdose; MRI, magnetic resonance imaging; APAP, acetaminophen; GI, gastrointestinal; NG, nasogastric.
- 1, definitely; 2, probably; 3, possibly.
- *No: ED1, possibly useful in acute care; ED2, probably useful in acute care; ED3, definitely useful in acute care; LT1, probably useful in long-term care; LT1, probably useful in long-term care; LT1, definitely useful in long-term care.
ear between 0 and 5000 μg/L for all drugs, with an r value of at least 0.999. The CVs for within-run and between-run precision were <6% and 19%, respectively. Ion suppression was calculated to be <3-fold at the retention times that the compounds eluted, and this suppression never prevented detection of any compound at its cutoff concentration in the controls.

Of 157 patient samples tested, 17 samples were positive for 1 or more of the drugs included in the panel. The most common drugs detected were diphenhydramine (6 samples), methamphetamine (4), and lamotrigine (2). One patient sample was positive for both diphenhydramine and isoniazid. Three drugs (MDMA, tiagabine, and tramadol) were not detected in any patient samples. The drugs detected and the corresponding concentrations are listed for each case in Table 2. Whether the drug was detected at therapeutic, less than therapeutic, or greater than therapeutic concentrations (multiples of the upper limit of normal) is also indicated.

Table 2 also summarizes the scoring for each of the 17 cases based on the adverse drug reaction criteria determined by the team of experts (Table 1). All cases received 1 point for the first criteria, since there are reports of all of the panel drugs causing seizures under certain circumstances. Likewise, all cases received 2 points for the second criteria, since drugs were detected in all of the samples when the seizure occurred, indicating that the seizure occurred after exposure to the drug. The third criteria allowed a range of points (from −2 to +2) to be assigned based on the possibility of an alternate cause for the seizure. Positive points were assigned when no alternate cause could be determined, whereas negative points were assigned when a clear (−2) or possible (−1) alternate cause was noted in the medical charts. For example, case 141 notes a history of alcohol withdrawal seizures, and the medical chart notes that the patient’s last drink was 24 h before the seizure; this case received −2 points for the third criteria. Alternatively, case 105 was assigned +2 since the patient had no history of seizures, a negative computed tomography scan of the head, and a history of drug overdose and depression. All cases involving cocaine, methamphetamine, and MDMA were assigned 1 point for the fourth criteria, since any concentration of these drugs could be potentially toxic. No points were assigned when a therapeutic drug was detected at therapeutic concentrations, although it is possible that some of these drugs can lower the seizure threshold in some patients even at therapeutic concentrations (e.g., bupropion (3)). In 2 cases, 113 and 105, a drug (quetiapine and diphenhydramine, respectively) was present far above therapeutic concentrations; those cases were assigned 1 point. The fifth criteria allowed 1 point in cases with additional evidence supporting the occurrence of an adverse drug reaction. For example, 1 point was assigned to case 113 (quetiapine overdose) because the medical chart indicated that the patient had prolonged somnolence beyond a postictal state, and somnolence is consistent with that particular drug overdose.

The likelihood of each of the 17 seizures being related to the drug detected was determined by summing the points from the 5 criteria. Table 2 lists the final score for each case, and of the 17 patient cases, 3 were probably or definitely drug-related. Two cases (113 and 105) where the seizures were deemed “definitely drug-related” were for overdoses of quetiapine and diphenhydramine, respectively, at concentrations that were 8 and 9 times higher than therapeutic concentrations (8).

The team’s findings on the usefulness of the test results in each case are summarized in Table 2. Patient care would have at least possibly been impacted in all but 5 cases. The results in 4 cases probably or definitely would have affected acute care (ED), and long-term care probably or definitely would have been affected in 7 cases. Overall, of the 17 cases, patient treatment probably or definitely would have been affected in 7 (41%) if the results of the seizure panel had been available prospectively.

For the 2 overdose cases that were deemed to be “definitely drug-related,” the test was determined to be probably or definitely useful to both the acute and long-term care of the patients. In the seizure resulting from overdose of quetiapine (case 113), the team decided the ED would have used the test information to justify admitting the patient to a monitored bed rather than the psychiatric ward (the outcome in this case). Furthermore, long-term care would be affected by instituting better monitoring of the patient at his home institution. In the diphenhydramine overdose (case 105), the patient presented to the ED after a minor motor vehicle accident caused by her seizure, and the history of overdose was neither clear nor immediately available. Therefore, the team of experts decided the ED would have used the test results to consider treating an acute overdose such as gastrointestinal decontamination (placing a nasogastric tube, giving activated charcoal), placing the patient on a cardiac monitor, and ordering an acetaminophen level, since many over-the-counter drugs containing diphenhydramine also contain acetaminophen. The patient’s long-term follow-up care also would have been affected since the high diphenhydramine concentration would obviate a full seizure workup, which is otherwise standard in patients with new-onset seizures. Additionally, since the semiquantitative results for both samples indicated concentrations far above therapeutic, psychiatric evaluations for suicidal ideation may have been triggered.

Five samples were positive for cocaine or methamphetamine, which are detected more rapidly and less
expensively by urine immunoassay. Nonetheless, these drugs were included in the panel to give more complete coverage of seizure-inducing drugs. Furthermore, the semiquantitative results of the test may yield information on whether the drug was present at toxic concentrations. A positive urine screen for the cocaine metabolite benzoylecgonine indicates that the patient ingested cocaine in the last 2–4 days, whereas a positive serum cocaine result in the seizure panel indicates same-day exposure, which is more likely to induce a seizure.

In 4 cases, mainly when the seizure was doubtfully drug-related, the team decided that the results of the seizure panel probably would not have been useful in patient management. For example, in case 21, citalopram was detected at therapeutic concentrations, and the seizure was most likely related to hypoglycemia. Therefore, the test results would not have affected patient care, since the patient was already appropriately treated for hypoglycemia. With seizures of known etiology, such as this one, a seizure panel test probably would not be ordered by the physician in the first place.

However, seizure panel results were considered useful in 9 other cases where the seizure was only possibly or doubtfully drug-related. In case 62, the concentration of isoniazid was found to be subtherapeutic and was therefore unrelated to the seizure, though the result probably would have affected care in the ED by helping rule out an isoniazid-induced seizure. The team also decided that long-term patient care probably would be affected by the primary care physician verifying patient compliance and reevaluating the patient's isoniazid dose. In this case, part of the effect is unrelated to seizure management. In another example, case 185, diphenhydramine was detected below the therapeutic range, which the team considered to be a negative result but possibly useful to both acute and long-term patient care. In the absence of an overdose situation, the ED might more strongly consider administering phenytoin for a potential new-onset seizure disorder, and the primary care physician might consider a full seizure workup. The seizure panel might have been similarly useful in many of the 140 cases in this study with negative results by ruling out common sources of toxic exposure.

Discussion

This preliminary LC-MS/MS method successfully detected numerous chemically unrelated drugs that are commonly implicated in drug-induced seizures. Results from such a seizure panel have the potential to affect both acute and long-term patient care in a variety of ways. Besides helping to rule in or rule out drug toxicity as a potential cause for a seizure, semiquantitative concentrations of prescription drugs can alert a primary care physician about the need to query or counsel the patient regarding compliance, request quantitative serum levels, or adjust dosing. Further studies are needed to determine if LC-MS/MS testing can improve patient outcomes or reduce ED costs by either improving throughput or reducing other testing needs such as imaging.

The seizure panel, which targets a narrow menu of drugs related to a particular clinical problem, is an example of what could become a new paradigm in toxicology testing. Targeted assays provide improved detection and specificity compared with comprehensive drug screens and have the advantage of supplying semiquantitative information that can inform the physician that a drug is not present, is present at therapeutic concentrations, or is present at toxic concentrations.

LC-MS/MS is a powerful tool in the clinical laboratory that is well suited to answering clinically driven questions. More panels of drugs or analytes could be developed to target other specific clinical presentations. For example, a cardiac drug panel could be developed to include β blockers, calcium channel blockers, and drugs that cause QRS widening. One way to implement strategies with numerous targeted panels using specialized instrumentation such as LC-MS/MS would be to create a regional toxicology laboratory, coordinated with state poison control centers that refer cases (11). The turnaround time in this model would be too long to affect acute patient care, but this study indicates that semiquantitative toxicology results can be useful to long-term care.

Because this work was a pilot study to determine the clinical utility of a seizure panel, additional analytical validation is necessary before clinical use. This includes analysis of possible interferences from licit and illicit drugs, more fully validating plasma and hemolyzed sample types, adding more internal standards to compensate for matrix effects, using multiple blank matrices to better analyze ion suppression, and optimizing the extraction method for recovery of isoniazid. These improvements to the assay would allow a negative result to more confidently rule out the seizure panel drugs as a cause for a seizure. However, the panel cannot entirely rule out a drug etiology since it does not include every drug implicated in the California Poison Control System study.

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