Electronic Supplementary Information

Experimental

All mass spectrometric data was generated on an API 4000 (Applied Biosystems) spectrometer as described in the Materials and Methods part. All MS data was acquired in ESI positive mode with a declustering potential of 60 V and a cell exit potential of 15 V. The mass transitions m/z 253→ 236, 253→ 210, 253→ 208, 253→ 180 and 253→ 167 were monitored in MRM mode at various collision energies in a single LC run. All product ion scans were collected using a collision energy of 25 eV.

Characterization of X-O-CBZ

The exact mass of $[M+H]^+$ was determined as m/z 253.0977 which verifies the suspected molecular formula $[C_{15}H_{13}N_2O_2]^+$ and reveals a deviation of only 0.5 mDa. The product ion mass spectrum of X-O-CBZ (see SI1) is similar to 2-OH-CBZ, 3-OH-CBZ, EP-CBZ, and OxCBZ. Depending upon the collision energy, all substances show fragment ions with m/z 236, 210, 180, and eventually 167. The fragmentation pattern allows a hypothesis about the position of the oxygen substituent (see SI2). Since the fragment ions 236 and 210 are formed, the substituent must be connected directly to the tricyclic ring. Presumably the substituent is not a hydroxy or oxo group because this would lead to an iminoquinone fragment ion (m/z 208) as in 2-OH-CBZ, 3-OH-CBZ and OxCBZ. Therefore a possible conclusion is the presence of an arene oxide, e.g. 2,3-epoxycarbamazepine that has been reported earlier as an intermediate in CBZ metabolism[1, 2]. In contrast to the hydroxylated metabolites, epoxides are not prone to form iminoquinone fragment ions. We also considered the numerous glucuronide and glutathione conjugates of the mono-oxygenated metabolites that have been reported[3, 4]. These conjugates are prone to cleavage during wastewater treatment and therefore not to be expected in the sample. Accordingly, a precursor ion scan of X-O-CBZ did not reveal any parent ion (data not shown).

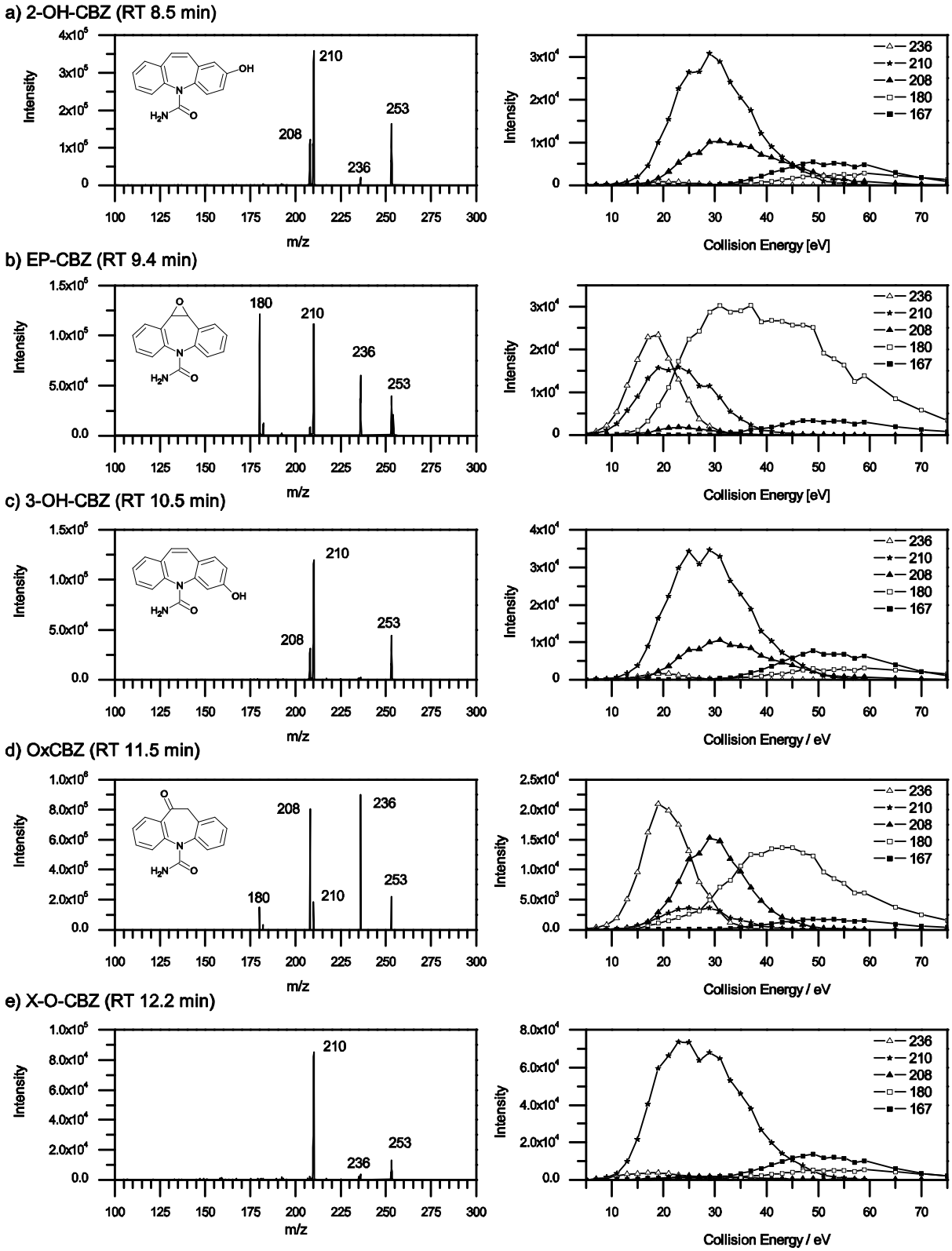

Fig. SI1: *MS analysis of 2-OH-CBZ (a), EP-CBZ (b), 3-OH-CBZ (c), OxCBZ (d) and X-O-CBZ (e) in 4000-fold concentrated wastewater; left: product ion spectra; right: distribution of fragment ions with varying collision energies after determination in MRM mode. These informations enable to distinguish compounds that show very similar product ion spectra.*

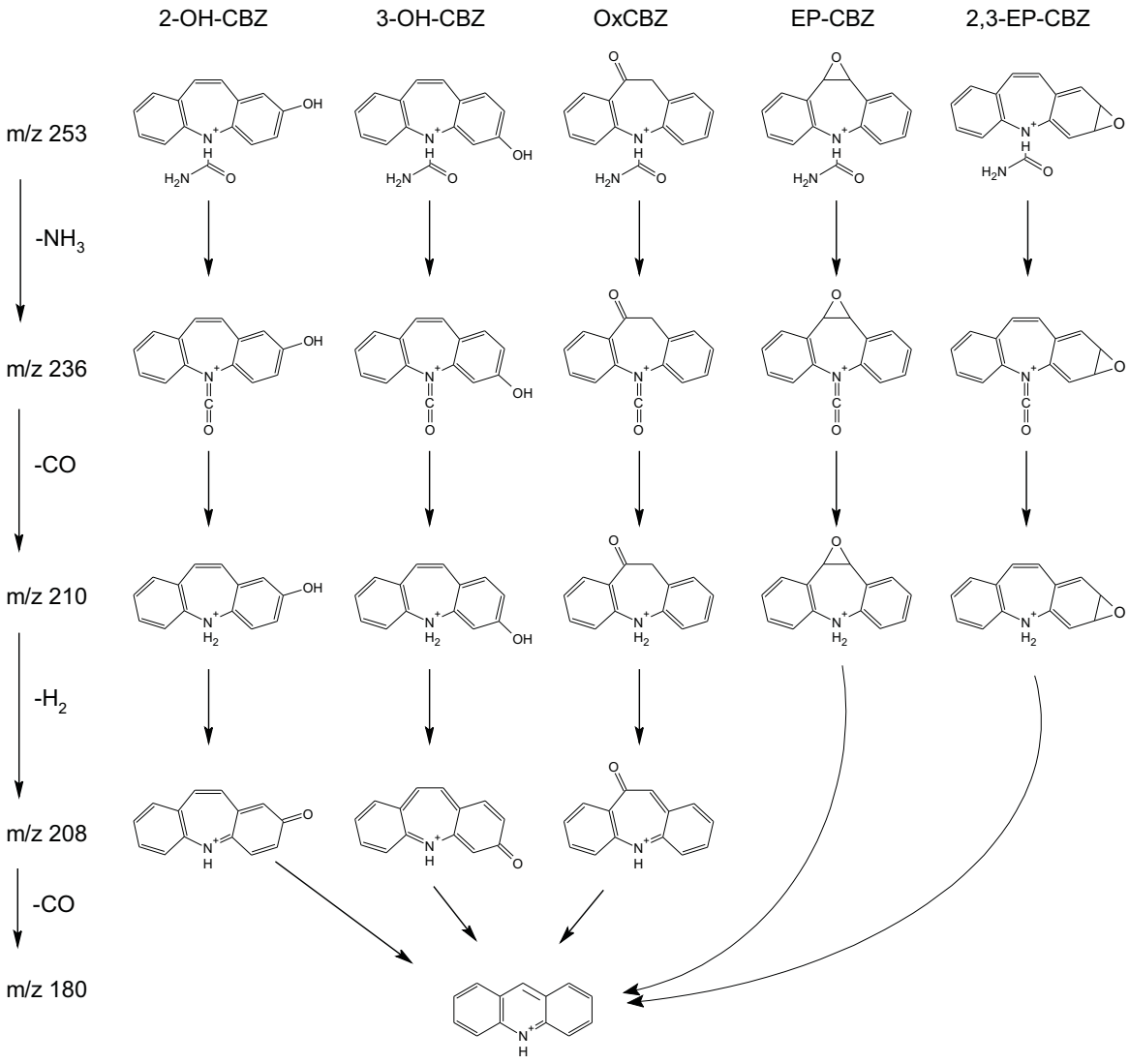

Fig. SI2: *Proposed ESI-MS fragmentation pattern for 2-OH-CBZ, 3-OH-CBZ, OxCBZ, EP-CBZ and 2,3-EP-CBZ.*

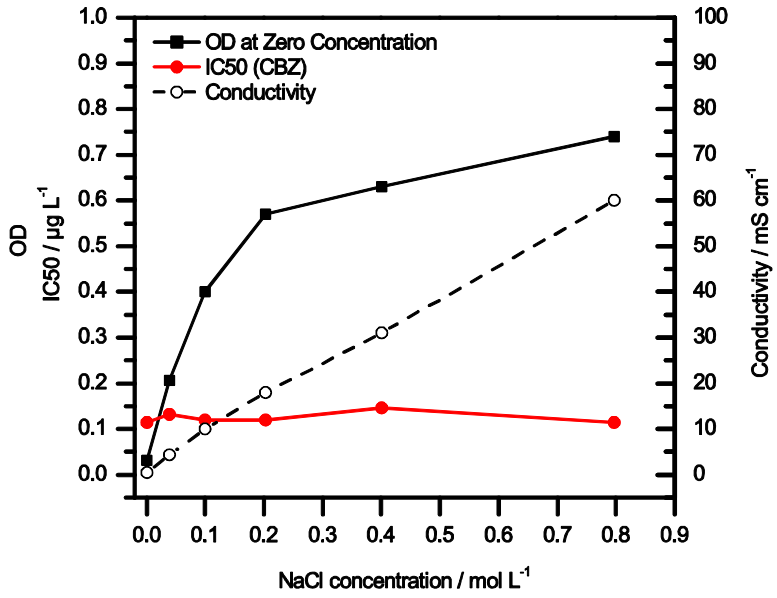

Fig. SI3: *Dependence of OD at zero concentration and test midpoint on sodium chloride concentration in the ELISA reaction mixture. Solutions were buffered with 8 mM phosphate at pH 7.6.*

1. K. Lertratanangkoon and M. G. Horning, *Drug Metab. Dispos.*, 1982, **10**, 1-10.
2. R. E. Pearce, G. R. Vakkalagadda and J. S. Leeder, *Drug Metabolism and Disposition*, 2002, **30**, 1170-1179.
3. J. L. Maggs, M. Pirmohamed, N. R. Kitteringham and B. K. Park, *Drug Metab. Dispos.*, 1997, **25**, 275-280.
4. M. K. Mahajan and C. A. Evans, *Rapid Commun. Mass Spectrom.*, 2008, **22**, 1032-1040.