Immunochemical monitoring of psilocybin and psilocin to identify hallucinogenic mushrooms.

**Abstract**

Development of rapid and reliable immunochemical methods for monitoring psilocybin (4-phosphoryloxy-N,N-dimethyltryptamine; Pyb) and psilocin (dephosphorylated metabolite; Psi), the psychoactive compounds contained within hallucinogenic mushrooms (magic mushrooms), is desirable in order to identify these mushrooms and regulate their illicit use. Because no antibody was publicly available for this purpose, we generated two independent monoclonal antibodies (mAbs) against Pyb or Psi, and then developed enzyme-linked immunosorbent assays (ELISAs) by using them. To generate the specific antibodies, novel immunogenic conjugates were prepared by linking Pyb or Psi molecules to carrier proteins by modifying their 2-(N,N-dimethylamino)ethyl side chains. Spleen cells from mice immunized with these conjugates were fused with P3/NS1/1-Ag4-1 myeloma cells, and hybridoma clones secreting anti-Pyb and anti-Psi mAbs were established. These mAbs were characterized for their biochemical features and then applied to competitive ELISAs, which used microplates coated with Pyb or Psi linked with albumin. These ELISAs enabled the determination of Pyb or Psi with measurable ranges of ca. 0.20–20 or 0.040–2.0 μg/assay (limit of detection was 0.14 or 0.029 μg/assay), respectively. The related tryptamines were satisfactorily discriminated as exemplified by the cross-reactivity of the ELISA to determine Pyb (or Psi) with Psi (or Pyb) that were found to be 2.8 % (or...