



Determination of Cannabinoids, Cathinones, and Synthetic Fentanyl Using Wastewater-Based Epidemiology

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ABSTRACT

- Develop an analytical method capable of determining a wide range of new psychoactive substances (NPS) in wastewater.
- Optimize sample preparation and instrumental analysis.
- Determine the prevalence of NPS in four rural Southern Illinois communities.

BACKGROUND

- Unregulated new narcotics or psychoactive substances (NPS) are created to have similar effects of established prescribed and illicit drugs.¹
- NPS are often more potent than established abused drugs. For instance, carfentanyl is ~100 folds more potent than fentanyl and ~10,000 folds more powerful than morphine.¹
- The prevalence of substance use in a community is typically determined based on forensic evidence, criminal statistics, self-report surveys, and hospital reports, causing an underestimation of the drug consumption and a delay between a drug outbreak and intervention of law enforcement.²⁻⁴
- In the U.S., the total forensic identifications of NPS tripled from 2016 (684) to 2019 (2023).⁵
- Wastewater-based epidemiology (WBE) is a near real-time, cost-effective, comprehensive, and a non-invasive method of determining the prevalence of substance use in a population.²⁻⁴
- WBE utilizes the trace levels of drug residues in wastewater to back-calculate the mass loading and community consumption rate of drugs.²⁻⁴
- NPS are reintroduced in markets in quick succession to impede the efforts of law enforcement to prohibit their production and purchase; therefore, WBE can be established as an early warning system for NPS.²
- There are no reports of NPS prevalence in U.S. communities using wastewater-based epidemiology.

TARGET ANALYTES

Opioids / Analgesics furanyl fentanyl (FUF) valeryl fentanyl (VAF) butyryl fentanyl (BUF) 4-fluoro-isobutyryl fentanyl (FIBF) acetyl fentanyl (ACF) 4'-methyl acetyl fentanyl (MAF) 3'-methyl fentanyl (MEF) para-fluorobutyryl fentanyl (PFBF) benzyl fentanyl (BZF) cyclopropyl fentanyl (CPF) methoxyacetyl fentanyl (MOAF) carfentanyl (CAF) U-47700 (U477) U-48800 (U488) 4-ANPP (ANPP)	Cannabinoids 5-fluoro MDMB-PICA (MDMB) MAB-CHMINACA (MABC) MMB-FUBINACA (MMBF) 5-fluoro EDMB-PINACA (EDMB) (R)-5-fluoro ADB (RADB) AB-FUBINACA (ABF) MMB-CHMICA (MMBC) AB-CHMINACA (ABC) ADB-FUBINACA (ADBF) 5-fluoro AMB (FAMB) NM2201	Piperazines 1-(3-chlorophenyl) piperazine (CPP) MT-45	Indole 5-IT	Amphetamine 4-methylamphetamine (MAMP)	Anti-Convulsant gabapentin (GBP)	Benzodiazepine clonazepam (CZP)
Cathinones 4-methyl pentedrone (MPD) α-pyrrolidinopropiophenone (PPP) N-ethylpentylone (NEP) (±)-methcathinone (MC) 4-chloro-α-PVP (PVP) mephedrone (MMC) ethylone (ETO)	3,4-methylenedioxypropylvalerone (MDPV) α-ethylaminohexanophenone (EAHP) 2-methyl-4'-(methylthio)-2-morpholinopropiophenone (MMMP)					

ANALYTICAL METHOD

- An analytical method (UPLC-MS/MS based) capable of determining trace levels of New Psychoactive Substances in wastewater was developed and validated.

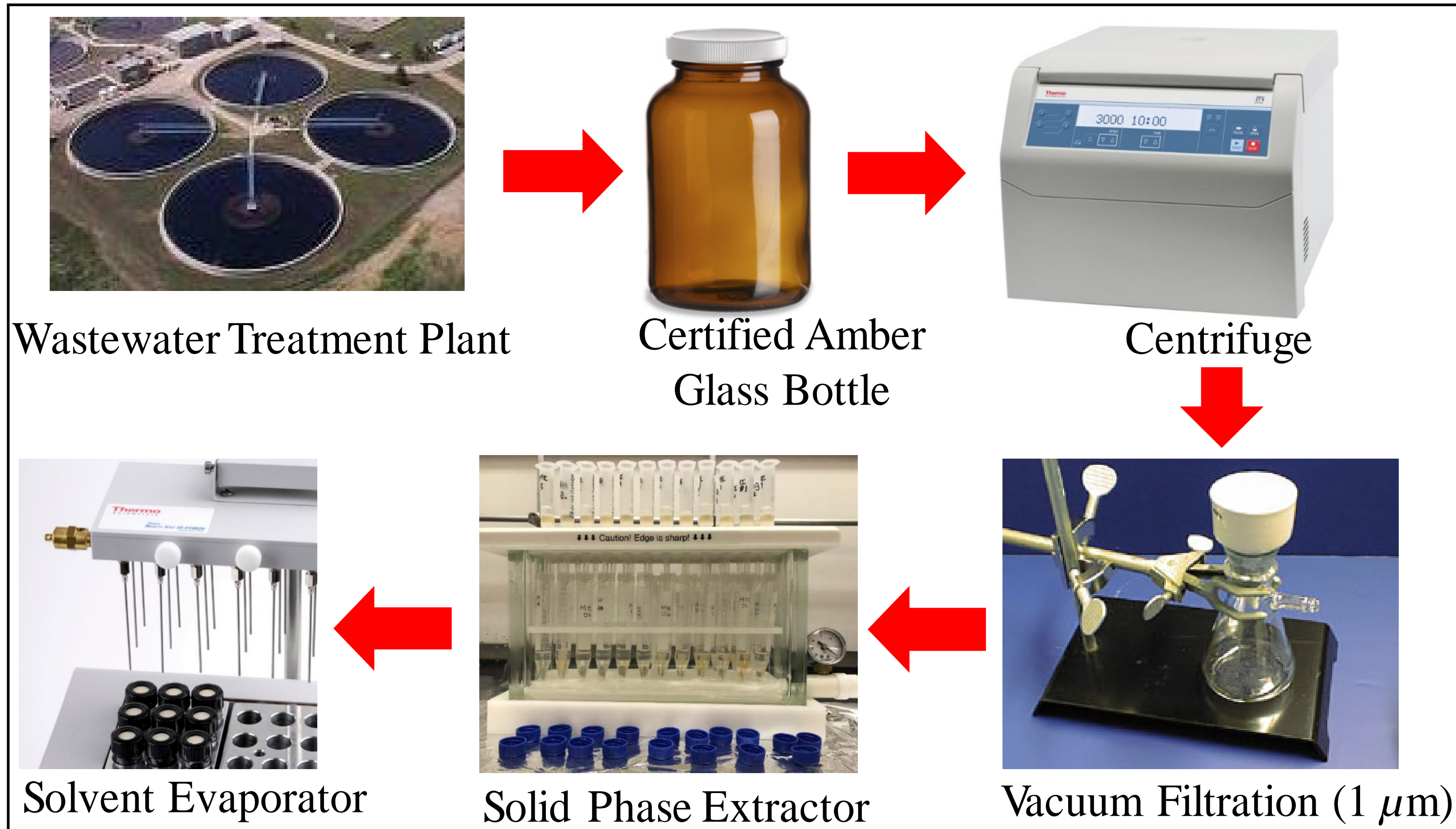


Figure 1. Schematic of sample preparation

RESULTS

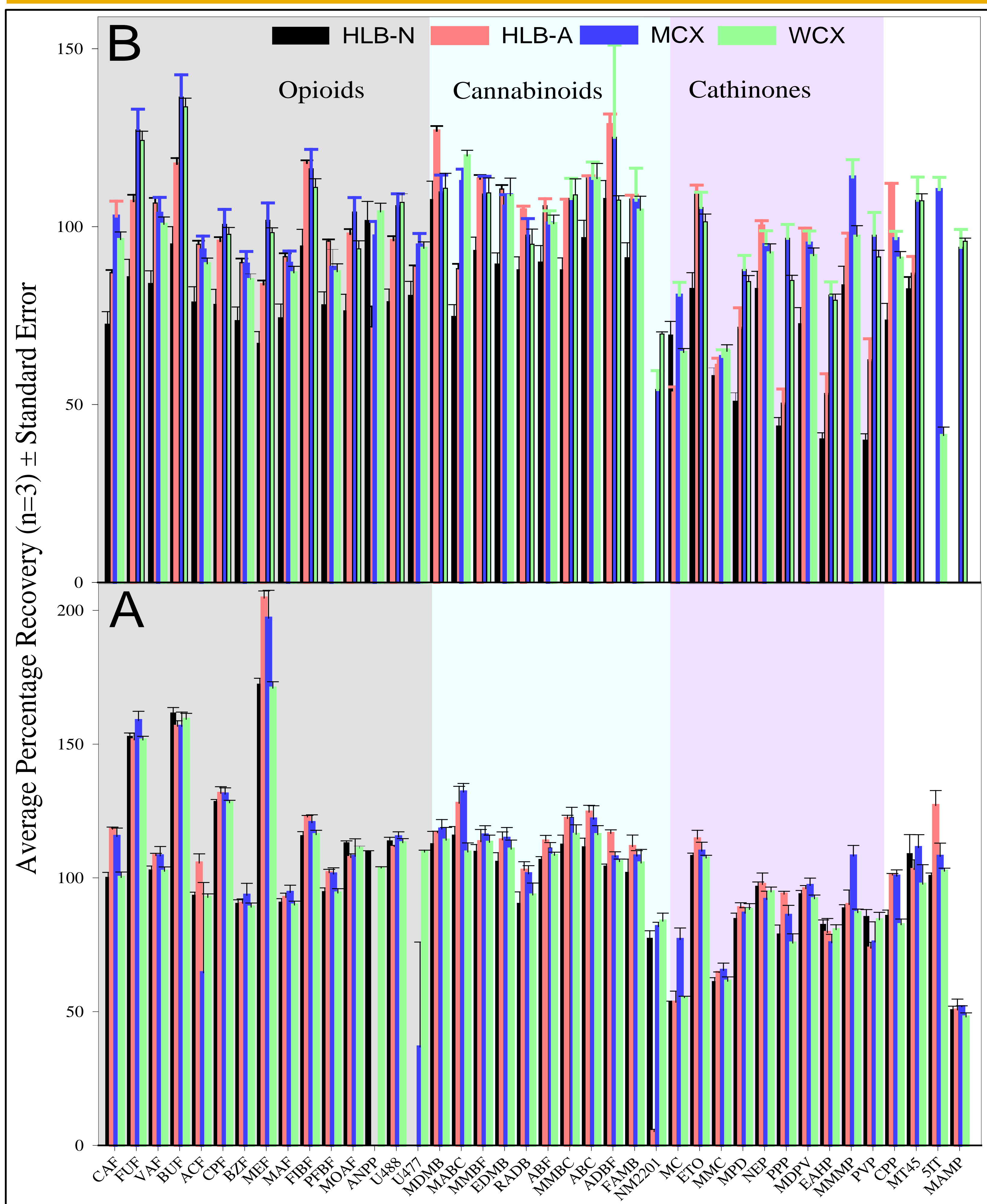


Figure 2. Absolute spiking recoveries of target NPS (n=3) at A) 50 or 100 ppb and B) 2 or 4 ppb using HLB-N, HLB-A (pH~2 sample using HLB), MCX, and WCX cartridges.

RESULTS (Contd.)

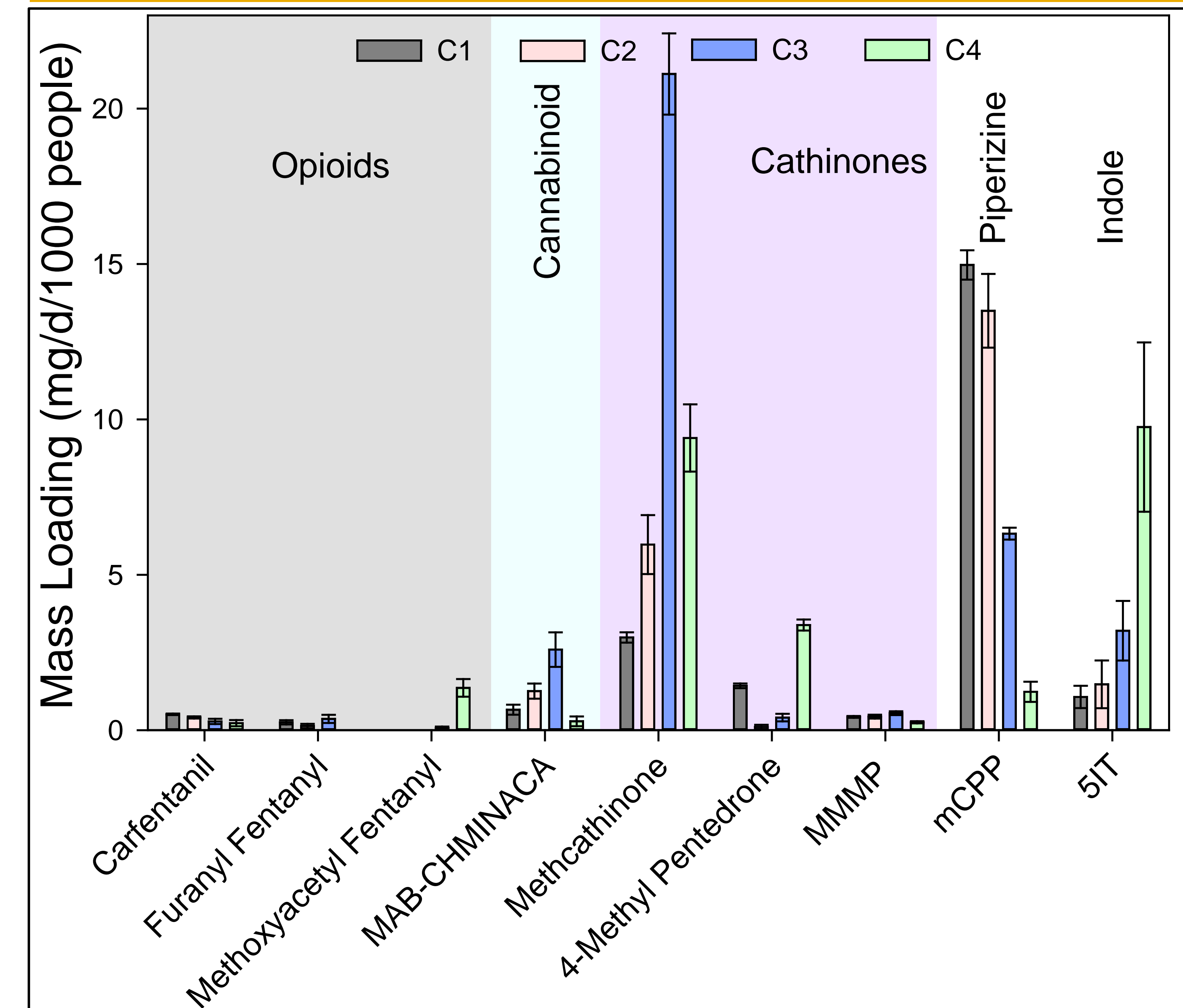


Figure 3. Prevalence of NPS in four rural Southern Illinois Counties

- Nine NPS were quantified in four rural communities in Southern Illinois.
- Methcathinone (MC), 1-(3-chlorophenyl) piperazine (mCPP), and 5-IT were the three most prevalent NPS, each more frequent in different rural areas.

CONCLUSIONS

- The analytical method for detecting 40 NPS, and gabapentin was optimized and used to monitor the occurrence of the 42 target analytes in four Southern Illinois communities.
- The average absolute recoveries of the target drugs during extraction optimization primarily ranged from 48% to 132% at 2 or 4 ppb.
- Methcathinone and MAB-CHMINACA were more prevalent in a University student-dominant community.
- 5-IT, 4-methyl pentedrone, and methoxyacetyl fentanyl were predominant in a town with the lowest per-capita income, while mCPP was more prevalent in communities with the higher per-capita income.

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