

Examination of Pesticide Residues in Wine, Beer and their Constituent Products using High-Throughput Techniques to Maximize Extraction & Efficiency

ABSTRACT

Commercial red wine and beer samples were examined for their pesticide concentrations. In addition to the examination of the finished alcoholic beverage, the constituent agricultural products of wine a beer production: grains, malts hops and wine grapes; were also examined to determine the levels of pesticides found in those products. The sample preparation and extraction process efficiency and recovery were examined by processing samples using manual versus high-throughput techniques. The QuEChERS method was used to process a greater number of samples in a shorter period of time than other extraction methods.

METHODS & MATERIALS

Samples

The wine & component samples included:

- Malbec and Syrah grapes obtained from a commercial urban vintner
- Six red wines of the same varietals from South American, US, Europe and Australia.

The beer & component samples included:

- Two Briess grain samples (organic and non-organic)
- A dark malt sample
- Four hop samples (Cascade, Magnum, Centennial and CitraHops)
- Six US craft beer samples representing the same hop varieties examined.

Solid Sample Preparation

- 2.5 g of solid samples (grain and hop samples) were ground using SPEX SamplePrep 6970 EFM FreezerMill®
- Program:
 - Precool for twenty minutes
 - Five grinding cycles(16 impacts/seconds) 2 min/cycle
 - Each cycle had an additional 2 min of cooling before the subsequent grinding cycle.
- Grape samples were used whole and ground in the centrifuge tubes using the SPEX SamplePrep ShaQer® 1500 with ceramic mixing media.

Liquid Sample Preparation

- The alcohol content of each liquid material was noted in each sample for later calculations.
- No additional sample preparation was conducted on liquid samples.

Sample Extraction

The AOAC 2007.1 method for the extraction and clean-up of agricultural products using QuEChERS were followed.

Initial Extraction used the SPEX CertiPrep SPEXQuE® QuEChERS Kits Part # AOAC-ACE-50ML

- 10 to 15 g of sample were placed in a 50 mL centrifuge tube
- 10 to 15 mL of 1% acetic acid in acetonitrile (HPLC grade) were added to each tube.

Samples were shaken mechanically for one minute in the SPEX SamplePrep ShaQer® 1500 using ceramic mixing media. An identical set of samples was hand-shaken also using ceramic mixing media for one minute to compare the pesticide recovery when using mechanical versus hand-shaken methods.

The contents of the SPEXQuE® Acetate Tube (AOAC-ACE-50ML), 75 µL I.S. solution (QUECR-AOAC-IS) and a spiking standard solution (QUECR-AOAC-QC) were added to the samples and the tubes were then shaken for an additional one minute using both the mechanical and hand-shaken methods. The samples were then centrifuged at 1500 U/min for one minute.

In the second step of the extraction and clean up:

- 8 ml of the ACN layer was transferred to a SPEXQuE® PSA Tube (AOAC-PSA-15ML).
- The mechanically shaken samples were again shaken for 1 min in the SPEX SamplePrep ShaQer® 1500 while the hand-shaken samples were agitated for 1 min by hand before centrifugation.
- Samples were centrifuged at 1500 U/min for one minute
- Supernatant was transferred to a clean tube & evaporated to near dryness in a Reacti-therm and solvent exchanged to 1 mL DCM.
- Samples were spiked with GC/MS IS (CLPS-I90) prior to being analyzed by GC/MS.

Analytical Conditions

- Instrument: Agilent GC-MS in Scan mode w/EIC (35-450 m/z)
- Column: DB-5 or eq. capillary column (30 m x 0.25 mm x 0.25 µm)
- Run program:
 - 55 °C x 1 min; 20 °C/min to 200 °C, hold 1 min; 30 °C/min to 310 °C, hold 3 min
- Detector: 280 °C
- Injector: 150 °C

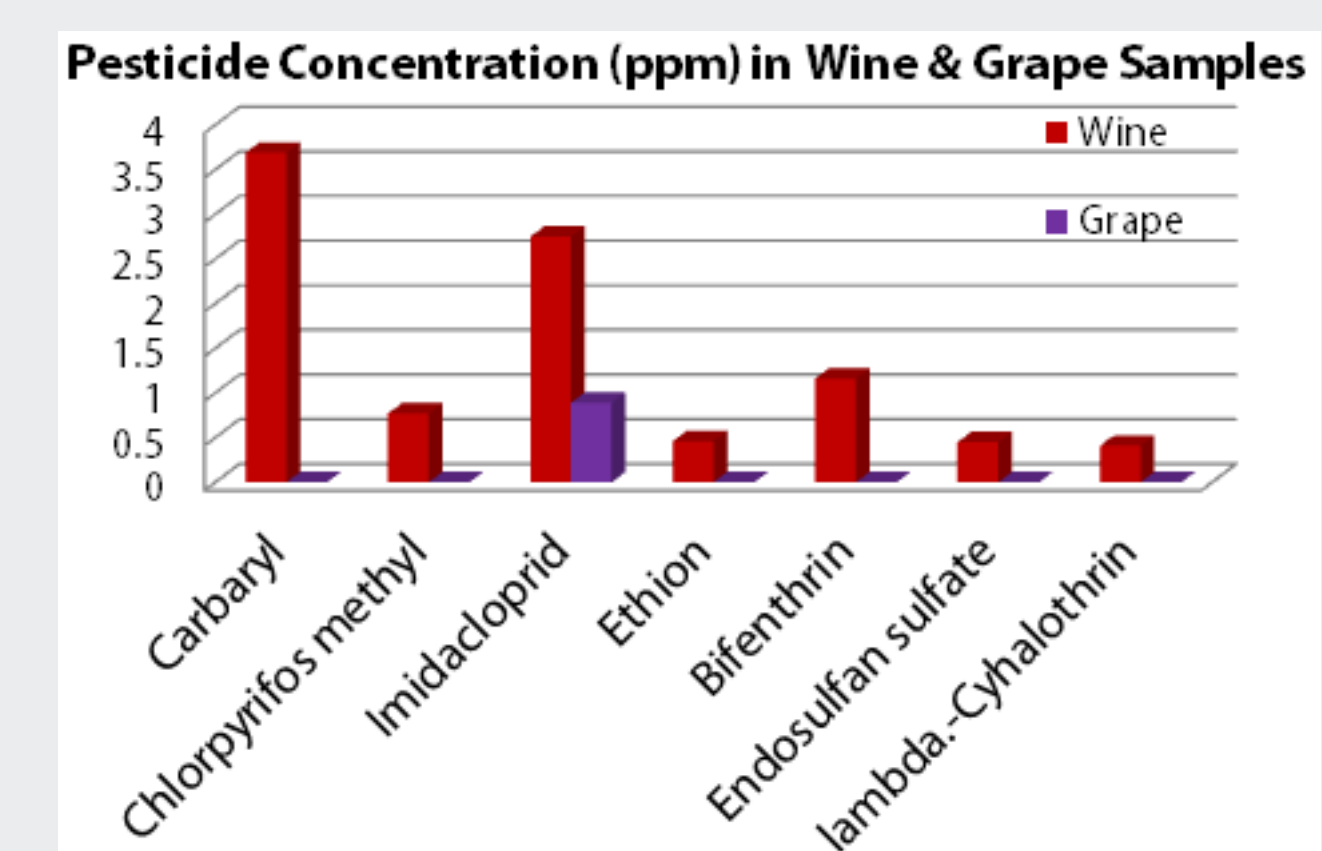


RESULTS & CONCLUSIONS

Wine and Wine Grapes

In the grape samples, only one pesticide, Imidacloprid, was found at levels above the spiked levels with a concentration of 0.9 ppm. The pesticide residue was only detectable in the mechanically shaken samples and not found in the hand-shaken samples.

The wine samples contained pesticide residues for several compounds: Carbaryl (3.7 ppm), Imidacloprid (2.8 ppm) and pesticide residues under 1.5 ppm for Ethion, Bifenthrin, Chlorpyrifos methyl, Endosulfan sulfate and Lamba-Cyhalothrin. The pesticide residues were increased by 5-15% using mechanical shaking techniques over the manual shaking. Comparison of the wine grapes and the finished wine products showed that Imidacloprid was found in both materials.



Beer, Grain, Malt and Hops

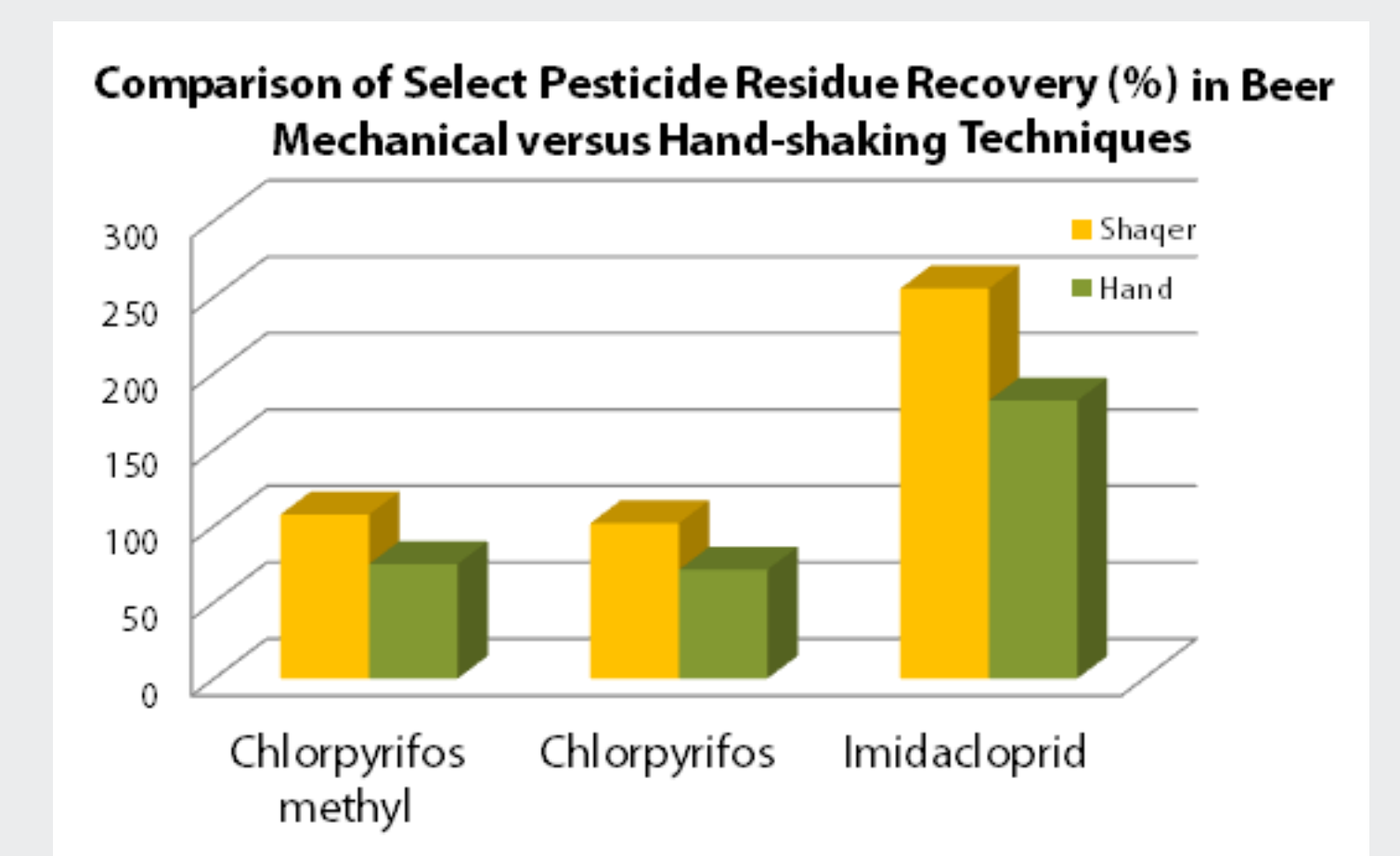
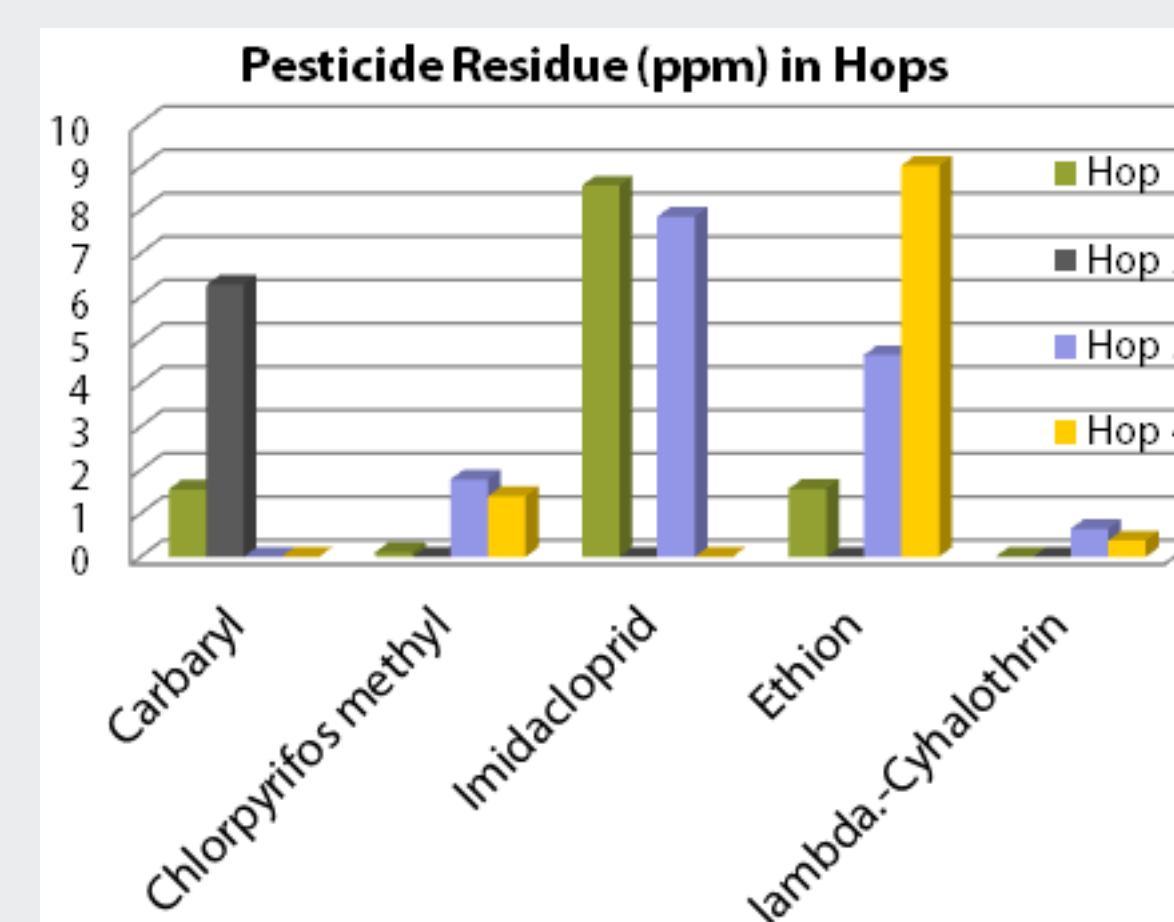
Two samples of the Briess grain were examined: traditionally grown Briess and organic Briess. The overall concentration of pesticide residues was lower in the Organic Briess samples than in the traditionally grown Briess sample. Bifenthrin was detected at 0.5 ppm in each sample type and Ethion, which had a concentration of 3.6 ppm in the traditional grain samples and 1.4 ppm in the organic grain samples.

The use of mechanical shaking increased the pesticide residue recovery in the grains by 10-25% over manual shaking methods.

The dark malt sample contained three detectable pesticide residues above the spiked range: Imidacloprid (0.75 ppm), Bifenthrin & Endosulfan sulfate (<0.5 ppm) in the mechanically shaken samples.

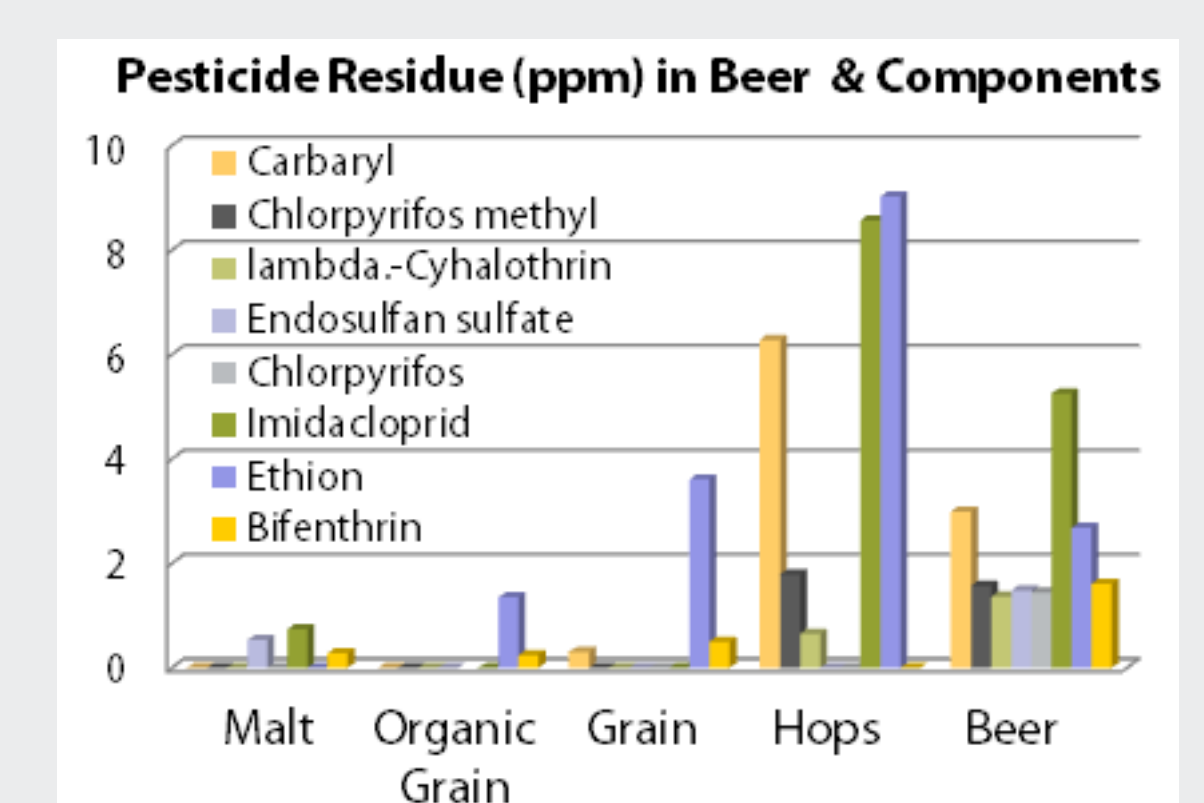
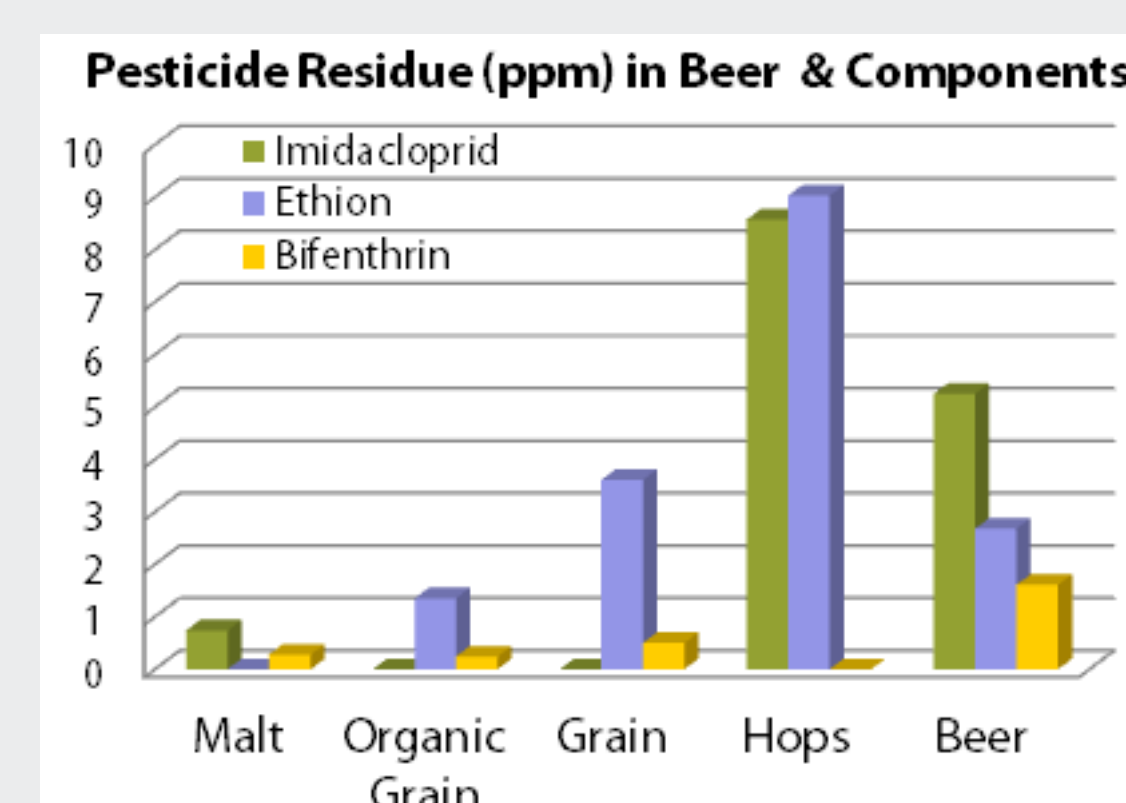
The hop samples showed the highest concentrations of pesticides of all the samples. Hops also contained numerous organic compounds such as: beta-Pinene, Caryophyllene, .alpha-Humulene, Lupulon, Vitamin E, Phytosterol. These organic compounds interfered with the detection and quantitation of some pesticides which were seen in other beer components. Additional sources of sample clean-up would need to be employed to remove more of these interfering organic compounds.

The hops showed high levels of Ethion (1.6-9.0 ppm), Imidacloprid (8.5 ppm), and Carbaryl (up to 6.5 ppm)



The beer samples contained detectable pesticide residue levels for Carbaryl (3 ppm), Ethion (2.5 ppm) and Imidacloprid (5.25 ppm). The use of the mechanical shaking techniques increased the pesticide recoveries in beer up to 35% over manual shaking.

The most commonly detected pesticides in the beer and beer components were Carbaryl, Ethion, Bifenthrin, Chlorpyrifos methyl, Chlorpyrifos, Imidacloprid, lambda Cyhalothrin, and Endosulfan sulfate. Imidacloprid was found at the highest levels in the hops and the finished beer products with a smaller concentration found in the malt. Ethion was found in all the beer and beer components with the highest levels in the hops, grain and finished beer. Bifenthrin was found highest in the finished beer with residue also found in the malt and both grains.



The highest pesticide levels detected in all the products tested were found in the beer hops with Imidacloprid detected at 8.5 ppm. The pesticides found in the beer components were also found in the finished beer products at up to 5 ppm (Imidacloprid) with most pesticides in the finished beer products being detected below 1-2 ppm. The use of mechanized shaking increased the recovery of the pesticides in all the sample types from 5-35%.

The wine and wine grapes showed one consistent pesticide residue (Imidacloprid) up to 3 ppm in the finished wine. The wine grapes only showed a detectable pesticide residue level in samples that were mechanically shaken versus hand-shaken.