3.3.4 Analytical Method and Method Modifications

The analytical method for zeta-cypermethrin in/on tomatoes was based on the methodology described in FMC report P-3421 (Section 7.0, Reference 3). The methodology involved a solvent extraction with 2:1 acetone/water, followed by evaporation to remove acetone. The resulting aqueous extract was partitioned with hexane. The hexane was passed through a silica gel solid phase extraction (SPE) cartridge, and analyzed by a gas chromatograph (GC) equipped with an electron capture detector (ECD).

The complete analytical method is included in Appendix III with the analytical flow scheme presented in Figure 2.

3.3.5 Instrumentation and Quantitation

A Hewlett Packard (HP) gas chromatograph equipped with a DB-5 (5% diphenyl methylpolysiloxane, J&W Scientific) megabore column, an HP 6890 autosampler, a $^{63}$Ni Electron Capture Detector (ECD), and Perkin Elmer Nelson Turbochrom® computer software was used for the determination of zeta-cypermethrin. An HP 5890 Series II GC equipped with an HP 7673 autosampler, an HP 5972 MSD, a J&W Scientific DB-5 narrowbore capillary column and a MS ChemStation (Window Based) was used for qualitative confirmation of zeta-cypermethrin residues. Detailed instrument parameters for all detector systems are listed in Appendix IV.

zeta-Cypermethrin was quantitated by a multiple point external standard calibration method. A computer spreadsheet program (Microsoft® Excel 97) was used for calculation and reporting. A full description of the calculations for quantitation can be found in Appendix V.
Method for zeta-Cypermethrin in/on Tomatoes

Matrix

Acetone/DI Water Mix (2:1)

Evaporate Acetone

Aqueous Extract

Hexane Partition

Concentration

Silica Cartridge SPE

Concentration

GC/ECD Quantitation
Appendix III

Residue Analytical Method for zeta-Cypermethrin on Tomatoes

Step 1 Accurately weigh 5.0 grams of tomatoes into a 50mL plastic centrifuge tube.

Step 2 Fortify all recovery samples. Allow solvent in fortification samples to evaporate.

Step 3 Add 30mL of acetone/water (2:1, v/v)

Step 4 Vortex for 10 minutes

Step 5 Concentrate to ca. 10-15mL. Partition 2x10mL hexane. Centrifuge if needed to eliminate emulsions.

Step 6 Combine hexane fractions and concentrate to ca. 3mL.

Step 7 Condition a silica gel SPE cartridge (1g, 6mL CV) with one CV of 5% ethyl acetate/hexane followed by one CV of hexane.

Step 8 Load the 3mL sample onto the cartridge. Rinse the centrifuge tube with 3mL of hexane and add the rinse to the cartridge. Drain the cartridge.

Step 9 Wash the cartridge with 4mL of 5% ethyl acetate/hexane.

Step 10 Elute and collect zeta-cypermethrin with 9mL of 5% ethyl acetate/hexane into a graduated centrifuge tube.

Step 11 Concentrate to < 1mL using N-Evap.

Step 12 Dilute to 1mL with hexane.

Step 13 Analyze for zeta-cypermethrin by GC/ECD.
Appendix IV

Instrument Parameters for zeta-Cypermethrin Analysis

INSTRUMENT : HP 6890 GC

COLUMN : J&W DB-5, (5% phenyl) methylpolysiloxane,
         15 m x 0.543 mm, 1.5 µm film thickness

INLET : Splitless Injection Mode

DETECTOR : Electron Capture

TEMPERATURE PROGRAM:
  Injection Port : 250 °C
  Oven : 200 °C / 1 minute (initial)
        : 20 °C / minute (ramp)
        : 280 °C / 10 minutes (final)

COLUMN GAS FLOW : He, Carrier, (Ar/Me, Make Up) ~30 mL / minute He,

INJECTION VOLUME : 2 µL

RUN TIME : 15 minutes

RETENTION TIMES : ~4.9 minutes
Appendix IV (continued)

Instrument Parameters for Confirmation of zeta-Cypermethrin by Mass Spec

INSTRUMENT: HP 6890 GC/5972A MSD

COLUMN: J&W DB-5MS, 5% phenyl methyl silicone, 15 m x 0.25 mm, 0.25 µm film thickness

INLET: Splitless Injection Mode
(Cyclo-double gooseneck liner)

DETECTOR: Mass Selective Detector

TEMPERATURE PROGRAM:
Injection Port: 250°C
Oven: 120°C/1 minute (initial)
10°C/minute (ramp 1)
200°C/0 minute (hold)
20°C/minute (ramp 2)
280°C/3 minutes (hold)
Transfer line: 280°C

COLUMN GAS FLOW: He, Carrier, ~1 mL/minute

INJECTION VOLUME: 2 µL

RUN TIME: 16 minutes

RETENTION TIME: ~6.8 minutes (zeta-cypermethrin)

ION MONITORED: 141, 169, and 198
From Zeta-cypermethrin on Peas – Report P-3441

3.3.4 Analytical Method and Method Modifications

The analytical methodology for zeta-cypermethrin in/on peas and processed peas (washed whole peas, canned peas, cooked peas, microwaved peas, steamed peas, and pea puree (baby food)) is similar to the method used in a previous zeta-cypermethrin crop analysis program detailed in FMC report P-3324 (Section 7.0, Reference 2). The method involved the extraction of zeta-cypermethrin from a homogenized matrix using an acetone/water (70/30) mixture. The resulting extract solution was then centrifuged and the extract solution decanted into a graduated cylinder.

A 1-gram aliquot with the addition of sodium chloride was then partitioned with hexane. The hexane fraction was concentrated to ~1 mL. The hexane fraction containing zeta-cypermethrin was cleaned up with a silica gel bonded SPE cartridge and the final solution containing zeta-cypermethrin was quantitated by a GC equipped with an ECD. The LOQ was validated at 0.05 ppm for zeta-cypermethrin by acceptable and reproducible recoveries of the analyte from laboratory fortified control samples. Based on a linear regression in each analytical data set, the LOD was calculated to be 0.03 ppm for zeta-cypermethrin in all pea samples. The method flow scheme is listed in Figure 3 and a detailed method description is listed in Appendix III.

3.3.5 Instrumentation and Quantitation

3.3.5.1 Instrumentation

An HP 6890 GC equipped with an HP 6890 autosampler, an ECD detector, a J&W Scientific DB-5 [(5% Phenyl) methylpolysiloxane] megabore capillary column, and a Perkin Elmer Nelson Turbochrom® computer software package was used for the quantitation of zeta-cypermethrin. An HP 6890 GC equipped with an HP 6890 autosampler, an HP 5972A MSD, a J&W Scientific DB-5 narrowbore capillary column and a MS ChemStation (Windows Based) was used for qualitative confirmation of zeta-cypermethrin residues. Detailed instrument parameters for both detector systems are listed in Appendix IV.

3.3.5.2 Quantitation

Zeta-Cypermethrin residues were quantitated by a multiple point external standard calibration method. A computer spreadsheet program (Microsoft® Excel 97) was used for calculation and reporting. A full description of the calculations for quantitation can be found in Appendix V.
Analytical Method for zeta-cypermethrin in/on Peas and Processed Peas

1. Weigh 5 grams of sample into a 50 mL Corning clear polypropylene centrifuge tube.

2. Add ~40 mL of acetone/water (70/30). Screw cap in place.

3. Horizontally shake the centrifuge tube for ~30 minutes. Centrifuge at 2000 rpm for ~3 minutes.

4. Decant the solution into a 100 mL graduated cylinder.

5. Repeat steps 2 and 3.

6. Decant the solution into the above 100 mL graduated cylinder. Bring final volume to 100 mL. Stopper and invert the cylinder to mix.

7. Place a 20 mL aliquot (1 gram) into another 50 mL Corning clear polypropylene centrifuge tube. Add ~2 grams of NaCl.

8. Partition acetone/aqueous twice with ~10 mL of hexane. Centrifuge at 2000 RPM for 3 minutes if emulsions occur.

9. Pipet each hexane layer into a 50 mL Corning clear polypropylene centrifuge tube.

10. Concentrate the combined hexane layers on the N-Evap (use a temperature setting at ~40°C and nitrogen setting at ~10 psi) to ~1 mL.

11. Condition a 1-gram silica gel SPE cartridge with one CV of 5% ethyl acetate/hexane followed by one CV of hexane. Allow hexane to drain to the top of the frit. Load the ~1 mL sample onto the cartridge. Rinse the centrifuge tube with ~1 mL of hexane and add the rinse to the cartridge. Allow hexane to drain to top of the frit.

12. Wash the cartridge with 5 mL of 5% ethyl acetate/hexane. Discard wash.

13. Elute the zeta-cypermethrin off the cartridge with one CV of 5% ethyl acetate/hexane and collect in a 13 mL centrifuge tube.

14. Concentrate on the N-Evap to less than 1 mL and adjust final volume with hexane to 1 mL or required final volume.

15. Inject on GC/ECD.
Appendix IV

**Instrument Parameters**
(Sample Analysis)

**INSTRUMENT:** HP 6890 GC

**COLUMN:** J&W DB-5, [(5% Phenyl) methylpolysiloxane], 15 m x 0.53 mm, 1.0 µm film thickness

**INLET:** Splitless Injection Mode
(Cyclo-double gooseneck liner)

**DETECTOR:** Electron Capture Detector

**TEMPERATURE PROGRAM:**
- **Injection Port:** 250°C
- **Oven:** 200°C/1 minute (initial)
  14°C/minute (ramp)
  280°C/8.2 minutes (hold)
- **Detector:** 300°C

**COLUMN GAS FLOW:** He, Carrier, ~11.5 mL/minute

**DETECTOR GAS FLOW:** Argon/10% methane, ~60 mL/minute

**INJECTION VOLUME:** 2 µL

**RUN TIME:** ~15 minutes

**RETENTION TIME:** ~6.1 minutes (zeta-cypermethrin)
**Instrument Parameters**
(Confirmation of zeta-cypermethrin)

**INSTRUMENT:** HP 6890 GC/5972A MSD

**COLUMN:** J&W DB-5MS, [(5% Phenyl) methylpolysiloxane], 15 m x 0.247 mm, 0.25 µm film thickness

**INLET:** Splitless Injection Mode
(Cyclo-double gooseneck liner)

**DETECTOR:** Mass Selective Detector

**TEMPERATURE PROGRAM:**
- Injection Port: 250°C
- Oven:
  - 120°C/1 minute (initial)
  - 10°C/minute (ramp 1)
  - 200°C/0 minute (hold)
  - 20°C/minute (ramp 2)
  - 280°C/3 minutes (hold)
- Transfer line: 280°C

**COLUMN GAS FLOW:** He, Carrier, ~1 mL/minute

**INJECTION VOLUME:** 2 µL

**RUN TIME:** 16 minutes

**RETENTION TIME:** ~6.7 minutes (zeta-cypermethrin)

**ION MONITORED:** 141, 169, and 198