in muscle, and 0.16-0.46 ppm in fat. Characterization was attempted only on liver and excreta. Acetochlor was detected at 5.6% of the TRR in liver, and analysis using the common moiety method indicated that EMA-type metabolites accounted for 12% of the TRR. In excreta, the principal ^{14}C-residue identified was *N*-(2-ethyl-6-methylphenyl) oxamic acid (Metabolite 27) at 10% of the TRR. Analysis of excreta using the common moiety method detected EMA-type metabolites at 19% of the TRR. This study was deemed inadequate because of insufficient characterization of ^{14}C-residues.

860.1340 Residue Analytical Methods - Plants

Tolerance enforcement methods: A tolerance enforcement method is available for determining residues of acetochlor and its EMA- and HEMA- producing metabolites in corn commodities. The method is a high performance liquid chromatography (HPLC) method using an oxidative coulometric electrochemical detector (OCED) and is listed as Method I in PAM Vol. II.

For this method, residues are solvent extracted into aqueous acetonitrile, concentrated, and base hydrolyzed to yield EMA and HEMA. The resulting residues are steam-distilled into dilute acid, adjusted to a basic pH, and partitioned into methylene chloride. HEMA is methylated using acidic methanol and residues of EMA and methylated HEMA (MEMA) are separated and determined using HPLC/OCED. Residues of EMA and HEMA are expressed in acetochlor equivalents and the validated method LOQ is 0.02 ppm for each analyte.

The Agency has previously noted that metolachlor metabolites can give false positive results for acetochlor EMA and HEMA residues. Instead of developing a separate confirmatory method for acetochlor residue, the registrant has provided adequate data indicating that the method available for metolachlor in PAM Vol. II (Method I) can be used to determine whether residues in corn commodities are due to metolachlor (positive) or acetochlor (negative).

Data collection methods: Adequate analytical methods are available for collecting data on residues of acetochlor and its EMA- and HEMA-type metabolites in/on plant commodities. Over the 20 years that residue data have been submitted for acetochlor, numerous analytical methods have been used as the definition of the regulated residue has been refined. Adequate method validation data were submitted for each of these methods.

In the earliest field trails, which included the analysis of only acetochlor and its EMA-type metabolites, residues were determined by a gas chromatography (GC) method using a nitrogen phosphorus detector (NPD). Residues of acetochlor and its EMA-type metabolites were extracted into acidic aqueous acetonitrile and sequentially acid and base hydrolyzed to yield EMA. The EMA residues were then steam distilled, partitioned into hexane, and cleaned up using a Florisil column. Residues of EMA were then determined by GC/NPD. The LOQ for this method is 0.02 pm for corn grain and 0.05 ppm for corn forage and fodder.

In more recent field trials, residues of acetochlor and its EMA- and HEMA-type metabolites were determined using variations of an HPLC/OCED method developed by Monsanto that is essentially the same as the tolerance enforcement method. Residues are extracted with aqueous acetonitrile or aqueous methanol, concentrated, and base hydrolyzed to yield EMA and HEMA. The residues are then steam distilled into dilute acid, adjusted to a basic pH, and partitioned into methylene

chloride. HEMA is methylated using acidic methanol and residues of EMA and methylated HEMA (MEMA) are separated and determined using HPLC/OCED. Residues of EMA and HEMA are expressed in acetochlor equivalents and the validated method LOQ is 0.02 ppm for each analyte.

Several other methods have also been used to collect data on residues of acetochlor *per se* in corn commodities. In one method, residues are extracted with acetonitrile, concentrated, diluted and partitioned into toluene. Residues are then concentrated, redissolved in hexane, and analyzed by GC/NPD or mass spectrometer detector (MSD). The validated method LOQ is 0.01 ppm. In another method (Method 244/01), residues are extracted with methanol, concentrated, diluted with a sodium chloride solution and partitioned into toluene. Residues are then cleaned up using solid phase amino and silica columns and determined by GC/NPD. The validated method LOQ is 0.01 ppm.

Residues of EMA- and HEMA-producing metabolites have also been determined using a GC/MSD method (Method 184). Residues are extracted with aqueous acetonitrile, concentrated and then base hydrolyzed by refluxing with NaOH and carbitol. Saturated sodium chloride solution is added to the hydrolysate and residues of EMA and HEMA are then partitioned into ethyl acetate. Residues are derivatized with heptafluorobutryic anhydride (HFBA) and then analyzed by GC/MSD. The LOQ is 0.01 mg/kg for both EMA and HEMA, or 0.02 mg/kg when expressed as acetochlor equivalents.

Another GC/MSD method (Report No. RJ1257B) has also been used for determination of Metabolite 57 residues in corn RACs and processed fractions. For this method, residues of Metabolite 57 are extracted with aqueous acetonitrile, concentrated and partitioned into ethyl acetate. Residues are then derivatized first to an isobutyl ester by heating in isobutanol/HCl and then using MTBSTFA to form tert-butyldimethylsilyl derivatives. The derivatized residues are then analyzed by GC/MSD. The validated method LOQ is 0.01 ppm.

In conjunction with the field rotational crop studies on sorghum, soybeans, and wheat, residues EMA-, HEMA-, and HMEA-type metabolites were determined using a HPLC/OCED method (RES-004-90) that is derived from the enforcement method. For this method, residues are extracted with aqueous acetonitrile, concentrated, and base hydrolyzed, distilling the resulting residues into an acidic solution. Residues are then partitioned into methylene chloride and analyzed using HPLC/OCED. The LOQ is 0.01 ppm for each metabolite class and 0.03 ppm for the combined acetochlor residues.

860.1340 Residue Analytical Methods - Livestock Commodities

Tolerance enforcement method: Although tolerances are not currently required for acetochlor residues in livestock commodities, an enforcement method is available for determining residues of acetochlor and its EMA- and HEMA-producing metabolites in milk and cattle commodities. This HPLC/OCED method is listed as Method A in PAM Vol. II (180.470) and is similar to the enforcement method for corn commodities.

For this method, residues are solvent extracted into aqueous acetonitrile, concentrated, and base hydrolyzed to yield EMA and HEMA. The resulting residues are steam distilled into dilute acid,

adjusted to a basic pH, and partitioned into methylene chloride. HEMA is methylated using acidic methanol and residues of EMA and methylated HEMA are separated and determined using HPLC/OCED. Residues of EMA and HEMA are expressed in acetochlor equivalents and the validated method LOQ is 0.01 ppm for each analyte.

Data collection methods: Adequate methods are available for determining residues in livestock commodities. Samples from the initial cattle, poultry and swine feeding studies were analyzed for residues of acetochlor and its EMA-producing metabolites using a GC/NPD method similar to the method used in the earliest analyses of plant samples. Residues are extracted into aqueous acetonitrile and sequentially acid and base hydrolyzed to yield EMA. The EMA residues are then distilled, partitioned into hexane, and cleaned up using a Florisil column. Residues of EMA are then determined by GC/NPD. The method LOQ is 0.02 pm for regulated livestock commodities.

In the other ruminant feeding study using dosing of an HEMA-type metabolite, residues in milk and tissues were determined using an HPLC/OCED method (MSL-10380) that is essentially identical to the above enforcement method.

860.1360 Multiresidue Methods

The FDA PESTDATA database dated 11/01 (PAM Volume I, Appendix I) indicates that acetochlor *per se* is completely recovered using Multiresidue Methods Section 302 (Luke Method; Protocol D) and 303 (Mills Method; Protocol E), but is only partially recovered by Method 304 (Protocol F). The registrants have also submitted data testing six acetochlor metabolites (3 - EMA metabolites; 2 - HEMA metabolites; and an EHMA metabolite) under the Multiresidue Method Testing protocols. None of these metabolites were recovered under the protocols.

860.1380 Storage Stability

The requirements for storage stability data are fulfilled for plant and livestock commodities. The available storage stability data adequately support the sample storage intervals and conditions from the crop field trials, rotational crop trials, processing studies, and livestock feeding studies.

Storage stability data are available indicating that acetochlor *per se* is stable in frozen corn, soybean, and peanut forage for up to 36 months, and residues of EMA and HEMA metabolites are stable in frozen corn grain, forage, and fodder for up to 49 months.

Storage stability data are also available indicating that Metabolite 57 is stable in frozen corn forage, fodder and grain for up to 52 weeks.

Storage stability analyses conducted in conjunction with the extensive field rotational crop studies indicated that residues of representative EMA-, HEMA-, and HMEA-type metabolites are stable at -18°C for up to 13 months in soybean seeds, forage, and hay and for up to 25 months in wheat forage, grain, and straw, and sorghum grain and silage.

In conjunction with the livestock feeding studies, storage stability data are available indicating that EMA-type metabolites are stable in frozen milk, eggs, liver, muscle, kidney, and fat for at least 16

weeks, and that the HEMA metabolites are stable for at least the 1 month that milk and tissues samples were stored prior to analysis.

860.1400 Water, Fish, and Irrigated Crops

Acetochlor is not registered for direct use on water and aquatic food and feed crops; therefore, no residue chemistry data are required under these guideline topics.

860.1460 Food Handling

Acetochlor is not registered for use in food-handling establishments; therefore, no residue chemistry data are required under this guideline topic.

860.1480 Meat, Milk, Poultry, and Eggs

Based on the use of acetochlor on corn, there is the potential for exposure of livestock to acetochlor residues in their diet. The MTDBs of acetochlor residues for livestock are calculated below in Table 7. Based on reassessed tolerances, the MTDB is 3.03 and 3.77 ppm for beef and dairy cattle, respectively, and the MTDB for both poultry and swine is 0.04 ppm.

Monsanto has submitted three studies reflecting the feeding of EMA-type metabolites to cattle, poultry and swine, and one study reflecting the feeding of an HEMA-type metabolite to dairy goats. Summaries of these studies are presented below. In considering the available livestock metabolism data and the feeding studies in conjunction with a requested increase in the tolerance on corn forage to 3.0 ppm, the Agency (G. Herdon, W. Dykstra, and C. Lewis; DP Barcodes D214735 and D214738; 6/25/96) reaffirmed an earlier conclusion by the HED MARC that there is no reasonable expectation of finite residues occurring in livestock commodities [40 CFR §180.6(a)(3)]. Therefore, tolerances for livestock commodities are not currently required.