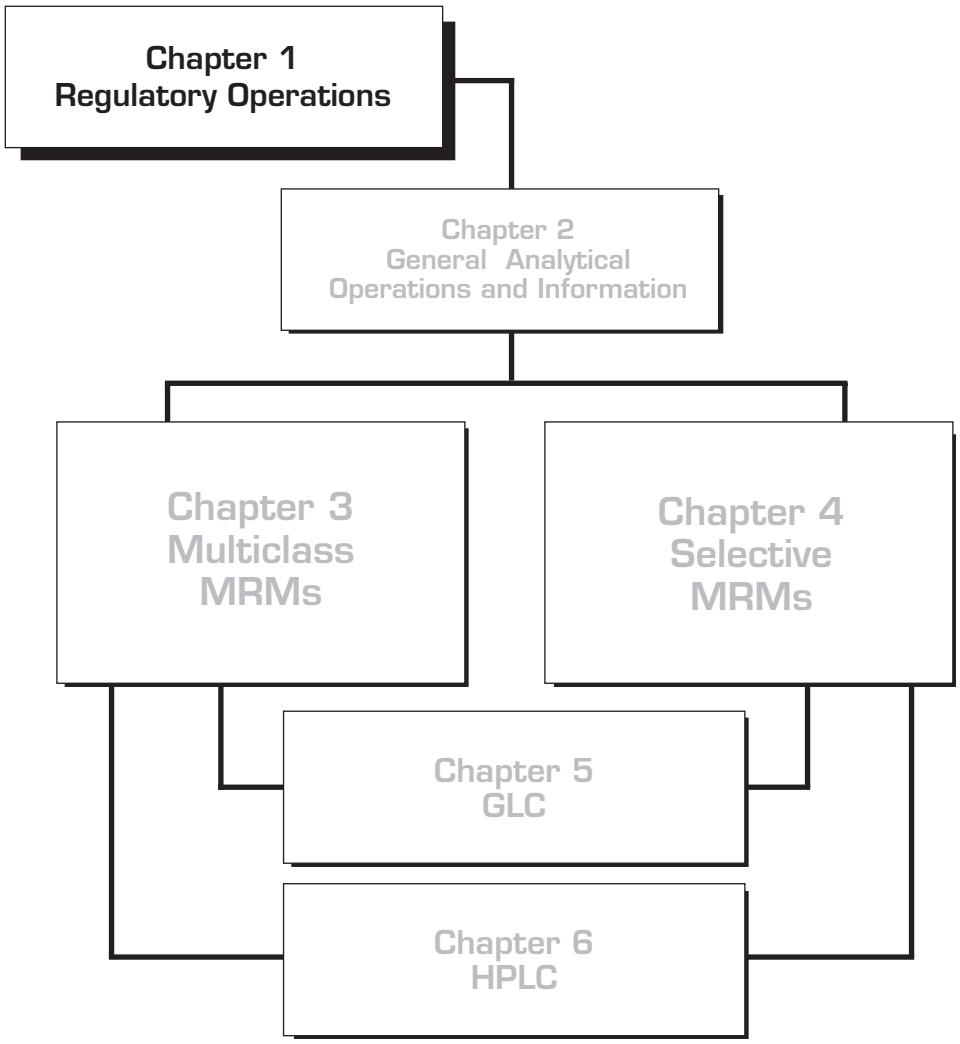

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101: REGULATORY POLICY

The expressed purpose of the Pesticide Analytical Manual is to publish analytical methodology used by the FDA in enforcing pesticide tolerances. To understand FDA's application of methodology published in the manual, it is important to understand pesticide tolerance regulations and related FDA regulatory operations. Material in Chapter 1 reflects FDA regulatory policies that affect its application of analytical methodology.

101 A: REGULATORY AUTHORITY

Information related to and the characteristics of pesticide tolerances include the following:

A tolerance is the maximum concentration of a pesticide residue that is legally permitted to remain in a food. The tolerance is not expected to be exceeded if the pesticide's registered use directions are followed.

The statutory authority for the Environmental Protection Agency's (EPA's) establishment of tolerances is provided by sections 408 and 409 of the Federal Food, Drug, and Cosmetic Act.

Tolerances established by EPA are set forth in Title 40 of the *Code of Federal Regulations* (CFR), Part 180 for raw agricultural commodities, Part 185 for processed food, and Part 186 for animal feed. The concentration of tolerances listed in 40 CFR 180, 185, and 186 is expressed in terms of ppm (*i.e.*, mg pesticide residue/kg food). In addition, certain pesticides are exempted from the need for tolerances; such exemptions are listed in 40 CFR 180.1001.

A tolerance for a pesticide residue on a raw agricultural commodity, *e.g.*, tomatoes, also applies to processed forms of that commodity, *e.g.*, canned tomatoes. In cases where processing may concentrate the residue, a food additive regulation may be issued in 40 CFR Part 185 to establish a higher tolerance on that processed commodity, *e.g.*, tomato paste.

A tolerance regulation specifies the composition of pesticide residue for which the limit applies; *i.e.*, a tolerance can apply to the parent form of the active ingredient only, parent compound plus one or more metabolites and/or degradation products, one or more metabolites and/or degradation products only, or some chemical moiety that can be measured analytically for calculating the pesticide residue. A chemical entity not specified by the tolerance regulation is not included in the residue for tolerance enforcement purposes (Section 104).

A tolerance regulation also specifies each individual food (*e.g.*, apples) or food group (*e.g.*, citrus fruit) to which the limit applies. No tolerance exists for a residue on a commodity unless the commodity itself or the group to which it belongs is specified.

In the examination of a food lot to determine whether it complies with tolerances, it is necessary to provide a sample for analysis that is representative of the lot in order to determine the average pesticide residue content

of the lot. Tolerances apply to that sample or a representative portion of that sample.

Unless otherwise specified in a tolerance regulation, each tolerance applies to the whole portion of a food commodity that moves in commerce. In practice, however, some food commodities (mainly raw agricultural commodities) require further definition as to the portion of commodity to which a tolerance applies and which is to be analyzed.

In summary, a tolerance provides a means of ascertaining that a pesticide was properly used. If a pesticide residue is found to exceed a tolerance or be present in a food for which there is no tolerance, then the pesticide was not used in a manner consistent with the tolerance. Under Sections 402 (a) (2) (B) or (C) of the Federal Food, Drug, and Cosmetic Act, this constitutes a violation of the law; *i.e.*, the food commodity is adulterated because it contains an "unsafe" or illegal pesticide residue.

101 B: REGULATORY OPERATIONS

To fulfill its responsibility to enforce regulations on pesticide residues in foods, FDA maintains a comprehensive pesticide program, including the analysis of food for enforcement of pesticide tolerances. Although the majority of samples analyzed contain no violative residues, sample handling must be consistent for all analyzed samples, because it is impossible to know in advance which samples will contain violative residues.

Accordingly, the following procedures must be followed by FDA laboratories to establish that a product contains illegal pesticide residue(s):

1) A laboratory sample of food must be collected from a consignment in accordance with agency sample collection instructions [1]; this laboratory sample is then considered representative of the food consignment.

2) The portion of food taken from the laboratory sample (whole product or specified parts of product) must be in accordance with agency instructions, and that portion must be appropriately composited and comminuted (Section 102). From that resulting test sample, a test portion is taken for analysis; test portion size is dictated by requirements of the analytical method. Residues found in the test portion are considered representative of the average residue content of the original consignment.

   (Note that terminology related to food products in this chapter, *i.e.*, "consignment," "laboratory sample," "test sample," "test portion," reflects recommendations of IUPAC Analytical Chemistry Division, Commission on Analytical Nomenclature [2]. Common usage, however, usually refers to the test portion as "sample," and this convention is used throughout most of PAM I.)

3) The test portion must be analyzed by a published, official analytical method or one that has been validated for the specific pesticide/commodity portion, and findings of residues must be confirmed (Section 103). For FDA monitoring purposes, analytical methods must be capable of accurately

measuring pesticide residues as defined by the tolerance regulation at not only the tolerance limit but also the lower limit of quantitation (Section 105).

4) If the residue level found in the original analysis exceeds an established tolerance, or if no tolerance exists for the residue in that commodity, another analysis of a second test portion of the same composited test sample must be conducted by a second analyst (normally a senior analyst); the second analysis is referred to as a "check analysis."

5) If check analysis verifies that the residue violates a regulation, *i.e.*, the results of both original and check analyses exceed a tolerance and are in close agreement or are in close agreement for pesticide residues for which there is no tolerance, the analytical findings will support enforcement action against the food consignment. If the check analysis result is below a tolerance or if the results of the original and check analyses are widely divergent, enforcement action cannot be supported. Additional analyses may be required to resolve widely divergent analytical results.

101 C: MONITORING

The FDA pesticide program has two main objectives: (1) to enforce residue tolerances and (2) to determine incidence and level of pesticide residues in the food supply. The section above defines operations established to enforce regulations. Monitoring aspects of the programs can be accomplished simultaneously, because levels of all residues found are calculated and recorded, whether or not they support enforcement action. Section 104 provides information about reporting residues for monitoring purposes, as well as determining compliance with regulations.

References

[1] Investigations Operation Manual, Sample Schedule Chart 3, FDA, Rockville, MD

[2] Horwitz, W. (1990) *Pure Appl. Chem.* **62**, 1193-1208

102: PREPARATION OF ANALYTICAL SAMPLES

102 A: INTRODUCTION

This section contains directions for preparation of test samples of food from laboratory samples collected for pesticide residue analysis. The following topics are considered, but not all are pertinent to every sample situation: (1) instructions for portion of commodity to be analyzed for pesticide residues, (2) directions for compositing and comminuting food items, (3) procedures for samples that are to undergo special analyses, and (4) requirements for retention of reserve portions of test samples.

102 B: PORTION OF FOOD COMMODITY TO BE ANALYZED

As a general approach, the "portion of commodity" composited to create the test sample consists of the entire food commodity (*e.g.*, whole cantaloupe). For many raw and processed foods, however, only specific portions of the food are included in the composite for the test sample. To ensure uniformity and consistency in tolerance enforcement and related monitoring, it is necessary to adhere to the following instructions on the portion of commodity to be analyzed.

Raw Agricultural Commodities

EPA regulations [1] specify that a raw agricultural commodity examined for compliance with a pesticide tolerance consist of the "whole raw agricultural commodity." The regulations contain some specific instructions on what constitutes the whole raw agricultural commodity; *e.g.*, "caps (hulls) shall be removed and discarded from strawberries before examination for pesticide residues." Such instructions are provided for only nine individual food commodities (*e.g.*, bananas) and crop group commodities (*e.g.*, root vegetables).

Recognizing the limitation of these regulations, FDA developed directions for additional commodities, taking into account practical considerations of sample preparation (*e.g.*, removal of stones from peaches to facilitate preparation of a homogenate). Table 102-a is a compilation of EPA regulations and FDA directions. (An EPA rulemaking is expected to be initiated that would amend the above existing regulation to incorporate FDA's more complete instructions on the portion of commodity to which a tolerance applies and that is to be analyzed.)

In some instances, a pesticide tolerance regulation specifies an exception to directions in Table 102-a. For example, the tolerance for mevinphos residues on melons states that compliance with the tolerance is to be "determined on the edible portion with rind removed," [2] even though the tolerances for most other pesticides on melons apply to the whole commodity including the rind.

Follow these directions to prepare test samples of raw agricultural commodities:

- Use the entire raw agricultural commodity, as specified in Table 102-a.
- When a pesticide residue is found in a commodity for which the tolerance applies to a portion different from that specified in Table 102-a, prepare a new test sample in accordance with the pesticide's tolerance regulation.

Table 102-a: Portion of Raw Agricultural Commodity to be Analyzed for Pesticide Residues

Root and tuber vegetables group[1]	Where separate tolerances are established for root or tuber, analyze whole commodity after removing adhering soil by lightly rinsing in running water. Where a tolerance is established on a root vegetable including tops and/or with tops, and tops and roots are marketed together, analyze tops and roots separately. Neither the pesticide residue on the roots nor the pesticide residue on the tops shall exceed the tolerance level. For carrots, parsnips, and rutabagas, remove and discard tops.
Bulb vegetables (green or dry) group	Whole commodity after removing and discarding roots. Remove adhering soil by lightly rinsing in running water. In the case of dry bulb onions and garlic, remove and discard stems and outer sheaths (husk or parchment skin) that are easily removed.
Leafy vegetables (except Brassica vegetables) group	Whole commodity after removing and discarding obviously decomposed or withered leaves. In the case of rhubarb, analyze only the stem without leaves. Remove adhering soil from celery by lightly rinsing in running water.
Brassica (cole) leafy vegetables group	Whole commodity after removing and discarding obviously decomposed or withered leaves, except remove and discard all leaves from cauliflower and headed broccoli and use sprouts only from brussels sprouts.
Legume vegetables (succulent or dried) group	Whole commodity, including pods for succulent and without pods for dry.
Fruiting vegetables (except cucurbits) group	Whole commodity after removing and discarding stems and husks.
Cucurbit vegetables group	Whole commodity after removing and discarding stems.
Citrus fruits group	Whole commodity.
Pome fruits group	Whole commodity after removing and discarding stems.
Stone fruits group	Whole commodity after removing and discarding stems and stones.
Small fruits and berries group	Whole commodity after removing and discarding caps and stems, except for currants, where the stems are to be included.

[1] Members of food groups are listed in 40 CFR 180.34 (f) (9).

Peanuts	Whole peanut meat (kernel) after removing hulls.
Peanut hulls	Whole commodity after removing peanut meat.
Dates and olives	Whole commodity after removing and discarding stems and stones or pits.
Pineapples	Whole commodity after removing and discarding crowns (leaves at top of fruit).
Avocados and mangoes	Whole commodity after removing and discarding stones.
Bananas	Whole commodity including peel after removing and discarding crown tissue and stalk.
Miscellaneous raw fruits and vegetables not previously included	Whole commodity after removing and discarding obviously decomposed or withered leaves, stems, stones or pits, shells or husks; if commodity has adhering amounts of soil, remove by lightly rinsing in running water.
Almond hulls	Whole commodity after removing shell and nutmeat.
Cereal grains group	Whole commodity (grain) except for fresh corn (including sweet corn). Include kernels plus cob after removing and discarding husk.
Eggs	Whole commodity after removing and discarding shells.
Fish	Edible portion of the commodity after removing and discarding heads, tails, scales, fins, viscera, bones (if inedible), and skin (if inedible).
Crab (hard shell)	Edible portion of commodity after removing and discarding shells, gills, and viscera.
Crab (soft shell)	Edible portion of commodity after removing and discarding gills.
Shrimp and crayfish	Edible portion of commodity after removing and discarding heads, shells, and inedible tails of shrimp.
Lobster	Edible portion of commodity including tomalley (liver) after removing and discarding shells and stomachs (hard sac near head).
Oyster, clam, and other shellfish	Edible portion of commodity including the liquor, after removing and discarding shells.
Rabbits and other game	Edible portion of commodity after removing and discarding bones.

Processed Foods

In the absence of EPA regulations, FDA also developed the instructions listed in Table 102-b on the portion of processed food to be analyzed for tolerance enforcement purposes. These instructions, like the ones for raw agricultural commodities, ensure uniformity and consistency in FDA analysis of processed food for pesticide residues. The instructions take a practical approach for sample preparation of processed food; *e.g.*, fruit juice concentrates that are normally reconstituted before consumption are also reconstituted prior to analysis for pesticide residues. Therefore:

- Follow the directions in Table 102-b to prepare test samples of processed foods.

Table 102-b: Portion of Processed Food to be Analyzed for Pesticide Residues

Processed food consisting of one ingredient and sold in a ready-to-eat form (*e.g.*, canned fruits packed in syrup or their own juice, canned vegetables packed in water or brine, or frozen fruits or vegetables, dried fruits, single-strength juices, catsup)	Analyze the whole processed commodity including any liquid or other edible media in which the commodity is packed. Discard inedible media, *e.g.*, brine.
Processed food consisting primarily of one ingredient and sold in a form requiring further preparation before it is ready to eat (*e.g.*, fruit juice concentrates, dehydrated vegetables, and powdered potatoes)	Analyze the whole processed commodity after compensating for or reconstituting to the commodity's normal moisture content.
Processed food in a form not ready to eat, used as an ingredient or component of other food (*e.g.*, flour, tomato concentrates such as paste, and citrus oils)	Analyze the whole processed commodity on an "as is" basis.
Cheese	Analyze the whole commodity including natural cheese rind after removing and discarding waxed or oiled rinds.
Frozen seafood (*e.g.*, fish or shrimp)	Analyze the edible portion after thawing; discard water.
Canned seafood	Analyze the edible portion including edible liquor and media, such as oil, broth, or sauces in which commodity is packed. Discard media that is not edible.
Frog legs	Analyze the edible portion of commodity after removing and discarding bones.

102 C: COMPOSITING AND COMMINUTING THE LABORATORY SAMPLE

Laboratory samples are comminuted or homogenized so that the relatively small (25-100 g) test portion taken for analysis is representative of the entire sample. Meaningful residue data can only be obtained when sample representation is preserved. Specialized equipment is employed to provide as much homogeneity as possible for the particular commodity.

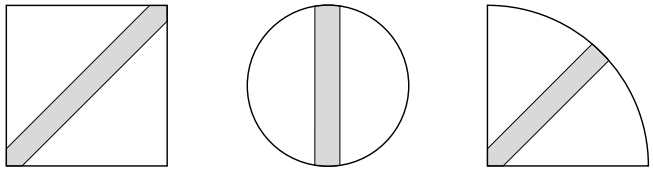
The general procedure for comminuting the commodity is:

- Comminute the test sample, prepared according to Section 102 B directions.
- Use comminuting operations (grinding, chopping, *etc.*) that produce the best possible homogenate for that commodity. Chopping procedures adequate for fruits and vegetables are often inadequate for homogenizing commodities such as dried hays and fish. See Section 203 for useful information about equipment and procedures.
- After comminuting, handle the homogenate carefully to minimize loss of residue by volatilization and concentration of residue through physical separation of product.

Exceptions to the general rule occur in several situations related to commodity type and special directions for the particular analysis. For example, removal of pits or caps from large quantities of small units can be too time consuming; similarly, melting, dicing, shredding, or blending a large unit of a food commodity such as butter or cheese is impractical. The following procedures are to be used in these situations:

- When the commodity consists of small units (*e.g.*, grains, cherries, nuts, dried peas and beans), mix and reduce by quartering to approximately 4 lb or 4 qt. Prepare the portion of commodity from this smaller amount, according to the appropriate directions in Table 102-a or 102-b. Chop or grind that material to obtain >1 lb or >1 qt comminuted sample.
- When the laboratory sample consists of large units of commodities of homogeneous nature (*e.g.*, butter, cheese), prepare the test sample by taking equal portions from each packaged unit. Select the appropriate portion of commodity (for cheese, see Table 102-b) and comminute by dicing, shredding, or blending.
- When the laboratory sample consists of large blocks, wedges, or wheels of cheese, take fraction shown as shaded area in Figure 102-a. Select the appropriate portion of commodity for cheese (Table 102-b) and comminute by dicing, shredding, or blending.

Figure 102-a
Fraction of Cheese Units to Take for Compositing



102 D: HANDLING SAMPLES FOR SPECIAL ANALYSES

Analyses for residues of ethylenebisdithiocarbamates (EBDCs) require special handling of the laboratory sample. EBDCs decompose rapidly as soon as the crop surface is broken and residues contact water, enzymes, and sugars [3]. Recoveries continue to decrease with time of contact in aqueous crop solution. Samples for EBDC analysis must either be analyzed immediately or frozen for storage.

Follow these directions for samples to be analyzed for EBDCs:

- Select representative units for EBDC analysis prior to chopping, grinding, or blending the laboratory sample.
- If the individual units are small and free flowing (*e.g.*, grains, beans, berries), mix well and take whole units for analysis.
- If the individual units are large, take wedges from each unit. Analyze immediately or freeze immediately after cutting.
- If the commodity contains free juices (*e.g.*, tomatoes, apples, oranges) and also requires cutting in pieces to fit into the digestion apparatus, freeze representative whole units before cutting. Dice frozen units without allowing them to thaw; mix and take sample for analysis.

102 E: RETENTION OF RESERVE PORTIONS

The following directions apply to all test samples (*i.e.*, comminuted material prepared from appropriate portion of commodity):

- Select three 1 qt portions from the total sample homogenate; identify them, respectively, as: "original analysis," "check analysis," and "reserve."
- Take analytical test portions from the "original analysis" and "check analysis," as appropriate, and analyze. Store the "reserve" portion in a freezer to provide to the claimant if requested.
- In addition to their sample homogenate, remaining fractions of large samples (*e.g.*, blocks of cheese) must be retained in a manner that prevents decomposition of product and/or residue. This requires that all products be frozen until findings of original analysis have been verified. The amount of commodity retained is governed by the extent of analysis required on the sample. However, in no case should portions be <1 qt each (or for products of high density, <1 lb) for original, check, and reserve.

References

[1] 40 CFR 180.1(j)

[2] 40 CFR 180.157

[3] Cullen, T.E. (1964) *Anal. Chem.* **36**, 221-224

103: METHOD APPLICATION IN REGULATORY ANALYSIS

103 A: INTRODUCTION

The Pesticide Analytical Manual (PAM) is published to provide analytical methodology for determination of pesticide residues in foods. Method application in a tolerance enforcement program imposes certain requirements and restrictions, and this section provides information about FDA operations concerning choice of method, identification, quantitation, and confirmation of residues(s), and documentation of analysis. Procedures established to support enforcement also apply to analyses that result only in monitoring data (*i.e.*, nonviolative samples), to the degree required in a program that fulfills both needs. (When PAM methods are used by other organizations for different purposes, *e.g.*, environmental monitoring without regulatory consequences, application requirements may vary from those expressed here.)

103 B: CHOICE OF METHOD

To support enforcement action against a commodity, original, check, and any additional analyses must be performed using official methods or methods shown by the analyst to support the validity of the result. The following minimum evidence is required to demonstrate that a method is valid for a particular analyte in a particular commodity:

1) Reagent blank analysis performed using reagents only (no commodity) shows no detector responses that could be mistaken for the analyte.

2) Historical or concurrent analyses of a residue-free lot of the same or similar commodity show no interfering detector responses.

3) Recovery of the analyte, added to a residue-free sample at or near the level of residue in the violative sample, is in the range 80-110%. If a residue-free lot is not readily available, recovery determination may be performed on another portion of the sample of interest by fortifying it with at least twice the level of residue found (*e.g.*, add 2 ppm if original analysis is 1 ppm).

Validation tests may be performed by either the original or check analyst and must be carried out concurrently with analysis of the sample of interest. The method used to determine any illegal residue found in a commodity for the first time by a laboratory must be validated in this way.

Analytical methods may be taken from the following sources, in decreasing order of preference:

PAM

Analytical methods from the PAM are suitable for regulatory analysis. As specified by regulation in the CFR, PAM contains the methods FDA uses for determining compliance with pesticide tolerances [1]. Volume I methods are capable of determining more than one residue (multiresidue methods, MRMs) and are most often used for routine analysis.

PAM I MRMs are presented as choices of several extraction, cleanup, and determinative modules. At the beginning of each method section in Chapters 3 and 4, method combinations that have undergone interlaboratory validation are listed. Absence of a particular combination of modules from this list does not prevent its use in regulatory analysis, but the analyst must provide suitable supporting evidence, as described above, that the combination is indeed valid for the particular commodity and residue combination.

PAM II methods are designed to determine residues of a single pesticide (single residue methods, SRMs). SRMs are published in PAM II for residues of all pesticides subject to EPA tolerances. These methods, usually from the pesticide manufacturer that petitioned for the tolerances, may be used by FDA to target residues not determined by MRMs. SRMs are also useful for check analyses when a residue has been determined first by an MRM, especially when a residue determined by an MRM is known to represent only part of the expected residue of a particular pesticide. For example, it may be known that the MRM does not completely recover the residue, or that the tolerance definition of the pesticide residue includes metabolites not detectable by the MRM. In these cases, MRMs may detect the presence of a residue but further analysis must be performed using a PAM II SRM to determine the total residue.

Other "Official" Methods

AOAC International maintains a system of interlaboratory testing through which methods passing the requirements are designated "AOAC Official"; many PAM I methods have undergone this process and are AOAC Official for certain residue/commodity combinations. Other AOAC official methods are not included in PAM but are acceptable for use in FDA regulatory analyses; AOAC methods have also been designated in the CFR as methods suitable for use [2]. In some cases, the AOAC method is preferable; *e.g.*, AOAC method 977.19 [3], official for hexachlorobenzene and mirex in adipose tissue, is preferred to PAM I method 304, through which the two chemicals involved are incompletely recovered.

Other Published Methods

When investigational evidence suggests that a commodity may contain a residue for which no PAM I or other official method is acceptable, published methods from the scientific literature must be sought. Use of such a method must be supported by documented evidence of its applicability to the residue and commodity in question, in the hands of the analyst who performed the regulatory analysis. The requirements described above are the minimum acceptable as supporting evidence.

Liquid-liquid partitioning and column chromatography cleanup steps that vary from established versions only in the use of proportionately smaller amounts of reagents are usually considered equivalent to the original; such methods are often referred to as "scaled-down" or "miniaturized." Miniaturized procedures that are published, validated, and used routinely in FDA are included as method modules in PAM I, *e.g.*, the 4 g Florisil column of Section 302 C1, developed as an alternative to the 20 g column. Certain other miniaturized versions of PAM I method modules have not undergone validation, but analyses performed in this way are considered adequate to support enforcement action and to assume equivalent coverage of residues for monitoring purposes. Extraction steps that involve smaller sample weight and extractant volume have not yet been studied sufficiently to

endorse, however; it is recommended that miniaturization be applied only to steps subsequent to filtration of the original extract.

103 C: TENTATIVE IDENTIFICATION

Application of methods in this manual results in tentative identification of residues based on the analyte's behavior matching that of a reference standard. "Behavior" of the analyte refers to its recovery through methods, including the eluate(s) in which it elutes from cleanup columns, its GLC or HPLC retention time, and the response it elicits from selective detectors or detection systems.

PAM I tables list test results of chemical behavior through various analytical procedures. The tables are provided to supply analytical chemists with information useful in residue identification. Typically, a sample is analyzed by a particular method and the extract examined by one or more determinative steps. When a GLC or HPLC response is recorded, the analyst measures the retention time of the response, calculates a relative retention time, and refers to the appropriate PAM I table (*e.g.*, Appendix I, PESTDATA; Table 403-a) to find chemicals that elute at that approximate time. Detector response data and molecular formulas included in PESTDATA offer further clues about which residue is likely to have caused response by the GLC element-selective detector used. Other details of chemical behavior through methods provide the analyst with means to eliminate certain candidate chemicals from consideration and strengthen the case for others; *e.g.*, if the method included a column chromatographic cleanup step and more than one eluate was used, only those chemicals known to elute in the pertinent eluate should be considered further.

In most cases, this preliminary evaluation provides the analyst with a limited number of potential candidates for residue identification. A reference standard solution of each likely chemical is then chromatographed on the appropriate GLC or HPLC system for direct comparison with the residue. In no circumstance is a table of data alone adequate for residue identification.

The expertise of the residue chemist is most critical during the determinative step of the analysis. The chemist's knowledge of pesticide usage and the chemistry and metabolism of pesticides is invaluable to the correct interpretation of evidence. Familiarity with commonly encountered artifacts from commodities, reagents, and environmental contaminants helps avoid incorrect conclusions.

103 D: RESIDUE QUANTITATION

Quantitation of residues is considered appropriate when:

1) The level of residue in a commodity is quantitated according to standard practices of GLC (Section 504) and HPLC (Section 606). In addition:

   a) Peak sizes of sample and standard match within ±25%.

   b) Time between sample and standard injection is ≤1 hr; chromatographic sequence ends with a standard.

   c) Replicate injections of standard and sample have been made when determining difficult-to-chromatograph residues.

2) Directions from Section 104 have been followed for summing levels of related residues for purposes of determining compliance with regulations.

3) Determination occurred at conditions that provided a limit of quantitation as directed in Section 105.

103 E: CONFIRMATION OF IDENTITY

Because analytical processes are subject to possible error in interpretation or measurement, confirmatory evidence must be developed to increase confidence in the tentative residue identification (Section 103 C). Attempting to define minimum confirmation requirements that are adequate for every situation is impractical. Instead, a philosophy of confirmatory analyses and discussion of certain minimum expectations are presented.

The extent of confirmatory effort will be influenced by the significance of the sample, nature and level of the residue, sample history, purpose of the analysis, and practical considerations such as time, cost, number of other samples, *etc.* The choice of confirmatory procedures depends on the tentative identity of the pesticide, amount of residue available for testing, sample type, and availability of instrumentation required for confirmatory tests.

The logic of most confirmatory schemes relies upon presumptive evidence; *i.e.*, if the behavior of an unknown in a particular analytical technique is the same as that of a reference standard, it is presumed that they are the same chemical. Any analytical technique that measures a single property of an analyte may err in distinguishing between two chemicals that behave the same; *e.g.*, two different chemicals may have the same GLC retention time. To avoid this potential error, either the analytical technique must inherently measure more than one property of the analyte or more than one analytical technique must be employed, each of which measures a different property. Following this logic, FDA laboratories confirm most residues by one of two approaches:

1) FDA requires that mass spectrometry (MS) be used to confirm the identity of any residue found for the first time. Modern laboratories usually have access to compact, highly automated mass spectrometers, configured as either mass-selective or ion trap detectors for GLC. Such instruments are capable, in some instances, of simultaneously detecting, quantitating, and confirming the residue, especially when reference standards are available. Errors may still occur, however, if GLC-MS is operated in the single ion monitor mode, which is not adequate to distinguish between two chemicals that elute from the column at the same time and are detected at the same m/z. For identification to be confirmed, GLC-MS must be operated by monitoring at least three ions [4].

   Full spectrum MS on a high resolution instrument may be required for full structural elucidation of previously unidentified chemicals. Such analyses usually require research MS instruments, operated by specialists, that offer several different modes of ionization that may be needed for unambiguous identification.

2) Residues previously reported may be confirmed by less rigorous techniques, such as additional chromatographic analysis (GLC or HPLC), with different

columns, mobile phases, and/or detectors. Confirmation of identity requires an accumulation of corroborating evidence sufficient to prove that the residue and reference standard must in fact be identical because they behave the same way in different tests. Such evidence is provided by measurement of a different chemical or physical property in each test used.

Selective MRMs (Chapter 4) are designed to be applicable to residues of a single chemical type; the steps of the method provide some built-in confirmation because only residues with that chemistry are recovered and determined. Other available confirmatory analyses are referenced as part of these method descriptions.

The GLC determinative steps included with multiclass MRMs (Section 302 DG1-DG23) offer a series of alternatives that can be used as confirmatory analyses. To the degree that a detector is selective to a single element or group, its use inherently provides some confirmatory evidence during the original tentative identification. Additional chromatography using other element-selective detectors is ideal for confirming a residue that contains appropriate elements. For example, many chemicals contain both nitrogen and phosphorus, others both phosphorus and sulfur. Complementary evidence from phosphorus and sulfur mode flame photometric (FPD-P and FPD-S), N/P, and electrolytic conductivity nitrogen mode (ElCD-N) detectors, as shown in Table 103-a, can provide excellent confirmatory information.

Table 103-a: Information Provided by Use of Element-Selective Detectors

Detector	Response	Conclusion
Example 1		
N/P[1]	Measurable	Either N or P in molecule
and		
FPD-P[2]	None	N in molecule
Example 2		
N/P	None	No N or P in molecule
and		
FPD-P[2]	Measurable	Large amount of S in molecule (verify with FPD-S[3])
Example 3		
N/P	Measurable	Either N or P in molecule
and		
FPD-P	Measurable	P in molecule; N also possible (verify N with E1CD-N[4])

[1] responds to both N and P
[2] responds to P; large amount of S can also cause response
[3] responds to S; presence of P can also cause response
[4] responds to N only

No detector is completely element-specific, however, and the analyst must be aware of the potential for false-positive responses. Replacement of the relatively nonselective electron capture detector with element-selective detectors encouraged development of methods with minimal cleanup. These methods are popular because they reduce analytical time and solvent volumes, and because they permit determination of residues that are removed by traditional column chromatographic cleanup steps (Section 301). However, extracts from such methods contain relatively large amounts of co-extractives, to which even selective detectors may respond. Columns of dissimilar polarity should be used with the various element-selective detectors to strengthen confirmation.

Special precautions are necessary when determining residues in which nitrogen is the only element to which element-selective detectors respond. Because many nonpesticidal organonitrogen chemicals occur naturally in foods, chromatograms from nitrogen-selective detectors often display a pattern of responses related to the commodity being analyzed. Additional columns and/or cleanup steps should be employed to confirm identification of residues found with nitrogen detectors.

Other techniques are available for confirmation if chromatography with element-selective detectors is not applicable or available. Chemical or photolytic derivatization and thin layer chromatography are among those most frequently employed.

103 F: DOCUMENTATION

FDA regulatory operations require that analytical results be adequate to support enforcement actions, if necessary, in a court of law. The analytical package accompanying a recommendation for enforcement action must demonstrate that the requirements described above for sampling, sample handling, and analysis have been met. Descriptions of analytical operations must be documented in a format readily understandable to another residue chemist and explainable to nonscientists. To that end, FDA established the following minimum requirements for documenting analyses that support recommendations for enforcement action against food or feeds because of the presence of violative pesticide residues.

Analytical Reports

Analytical reports for both original and check analyses must provide complete information on the sample and on sample handling steps used, as well as the following analytical information:

1) Method reference or memorandum of analysis, including description of any modifications made to the referenced method

2) Measured weights and volumes used for sample weight calculations

3) Volume of final sample solution or eluate

4) Details of residue determination record

   a) Sample solution/eluate identity (*e.g.*, 6% ethyl ether/petroleum ether)

   b) Sample weight per unit volume

c) Volume and sample weight equivalent injected

d) Retention time or distance and retention time relative to the marker compound appropriate to the system

e) Peak size; area or height for sample(s) and standard(s)

5) Calculation of results; may also appear on chromatograms

6) Column and detector used for each injection

Chromatograms

Each chromatogram must be labeled with identity of solution, volume and weight injected, date, analyst's initials, and time of injection. Time is not required if a continuous chromatogram is submitted. The following chromatograms must be submitted:

1) Marker compound. For each column-detector combination used, a chromatogram of the reference standard used as a marker compound for that combination; it is preferable for additional reference standards also to be included in a mixed standard solution appropriate to the determinative step.

2) Sample and standard chromatograms used for quantitation

3) Any other chromatograms (for both sample and standard) associated with additional tests to confirm identity of the residue

Chromatographic Data

The following information must be included on the chromatograms. (If a data collection system is used, much of the required information can be automatically entered into the chromatographic report.)

1) Brand name and model of chromatograph

2) Column: type, size, liquid phase, plus solid support and percentage loading for GLC packed column; coating identification, film thickness, length and internal diameter for GLC wide bore or capillary column; packing identification, length, and internal diameter for HPLC column

3) Temperatures: column, detector, injector, transfer lines, furnaces, *etc.*

4) Description of injector and/or inlet system

5) Gas flow rates and identities for carrier, fuel, purge and make-up gases (GLC); mobile phase, gradient if applicable, flow rate (HPLC)

6) Detector type, including design, mode of operation, and specifics related to operation of the particular detector, such as ion exchange resin, electrolyte, and electrolyte flow rate of the electrolytic conductivity detector

7) Range settings of electrometer, integrator, linearizer, *etc.*, along with pertinent detector voltages

8) Parameters for signal measurement: recorder span and speed, integrator settings, *etc.* (recommended chart speed is 1 cm/min or 0.5"/min)

References

[1] 40 CFR 180.101(c)

[2] 21 CFR 2.19

[3] *Official Methods of Analysis* (1990)15th ed., Association of Official Analytical Chemists, Inc., Arlington, VA

[4] Sphon, J.A. (1978) *J. Assoc. Off. Chem.* **61**, 1247-1252

104: ANALYTICAL RESULTS

104 A: INTRODUCTION

Three separate but related responsibilities must be managed within any regulatory program of analyses for pesticide residues: (1) determination of residue identity and calculation of level, (2) reporting residue identity and level, and (3) judging whether the presence of that level of residue is in compliance with regulations or whether it warrants enforcement action.

The first responsibility, residue identification and calculation, is a scientific endeavor treated in the remaining chapters of this manual; regulatory requirements for these operations were discussed in Section 103. Regardless of the purpose of the analysis, the basic instructions remain the same. Appropriate application of Chapters 3 or 4 methods, combined with the precepts of accurate quantitation described in Sections 504 and 606, will permit the chemist to ascertain the presence or absence of particular residues and to calculate the quantity of each residue present in the sample.

Beyond the scientific endeavor, the use to which analytical results are put is within the province of the organization sponsoring the work. In this section, current FDA procedures for reporting residues and determining compliance with prevailing regulations are noted. Operations are often dictated by the needs of the dual-purpose FDA pesticide program: to monitor incidence and levels of pesticide residues in foods and to enforce regulations concerning permissibility of these residues.

104 B: REPORTING

In this context, reporting the presence and levels of pesticides refers to laboratory entry of information into a computerized data base. FDA has monitored residue trends for more than 30 years by reporting results of all analyses, including those in which no residues were found, into a data system. Pertinent information deduced from accumulated data includes the decline in residue levels of persistent chlorinated hydrocarbons in the years following their ban and the identity of terminal residues in different commodity types. One goal of the residue reporting system is to retain all pertinent information obtained during the analysis; of special interest is the precise identity of the residue. Accumulated reports of residue analyses are used to answer many questions about the prevalence of residues in the food supply; most prominently, the data are used to prepare an annual report of residue findings [1].

The following reporting practices have evolved over the years to produce the most meaningful possible data for agency interpretation:

Portion of Commodity

The exact portion of food that is analyzed for pesticide residues is dictated by the purpose of the analysis. Section 102 reflects agency procedures for the portions of particular commodities used in FDA regulatory monitoring. Once analyzed, residue levels are calculated and reported based on the exact portion of food taken for analysis as indicated in Table 104-a.

Table 104-a: Portion of Commodity for Calculation and Reporting of Residue Levels

Commodity	Report Results On:
Raw agricultural commodities	Whole commodity as prepared for analysis
Milk products[1]	Whole product
Juice concentrates or powders	Reconstituted basis
Concentrated/dehydrated products consumed "as is" or used as ingredients	Whole product, "as is"
Dehydrated vegetables intended for use *after* reconstitution	Calculated equivalent weight of original product before dehydration
Other processed foods	Whole product

[1] Includes whole, low fat, skim, and other milk products. Note that reporting residues in whole milk on the whole basis (effective 10/1/91) does not change the way of determining compliance with regulations for residues whose tolerances in milk are on the fat basis.

Nature of the Residue

Whenever possible, pesticide residues in foods are identified and calculated as individual chemicals (*i.e.*, parent compound, metabolites, and degradation products). Although many pesticides are formulated and marketed as technical mixtures of related chemicals, their residues are calculated separately whenever chromatographic conditions and availability of separate reference standards permit.

Residues are reported as the individual isomer or congener that is identified and calculated during the determination. If the residue can be identified and its level calculated only by comparison to a mixed or technical reference standard, the residue may be reported as such in the data system.

Residues of polychlorinated biphenyls (PCBs) are calculated by comparison to commercial mixtures known as Aroclors (Section 504 D). Levels are reported in terms of the particular Aroclor(s) used as reference standard.

Residues Measured from Derivative or Breakdown Product

Some analytical methods convert the residue(s) to a derivative or breakdown product so that common determinative steps can be used. In these cases, some convention must be established for reporting the residues. Specific instances of this situation are:

Acids and Phenols. Residues of acids and phenols are converted to respective methyl esters/ethers by Section 402 method, to produce chemicals that can be measured by GLC. Reference standard solutions are prepared from standards of the ester/ether, if available, or from standards of the acid/phenol carried through the procedure. Levels of residue are calculated and reported as the acid or phenol.

Benomyl, Thiophanate-Methyl, and Carbendazim (MBC). Residues of these benzimidazole pesticides and related residues are determined by Section 404. MBC, the most common residue, may result from use of benomyl, which converts rapidly to MBC; thiophanate-methyl, which converts slowly; or carbendazim, a fungicide that is the same chemical as MBC. Residues are reported according to assumptions made about the source of MBC found.

In absence of evidence to the contrary, a residue of MBC is assumed to result from use of benomyl. MBC residues are quantitated by comparison to a reference standard of MBC and the level converted to the equivalent benomyl level, which is reported.

However, if MBC and thiophanate-methyl are both found in the sample, or if investigatory evidence indicates the commodity was treated with thiophanate-methyl, the quantitated level of MBC is converted to equivalent thiophanate-methyl and reported as such.

EBDCs. Tolerances for EBDCs are established in terms of parts per million (ppm) zineb, one of the EBDCs. Residues of EBDCs are determined by methods that convert these chemicals to carbon disulfide, which is measured and calculated as zineb. This analytical approach precludes identification of which EBDC was present. By convention, FDA laboratories report levels found as "EBDC (identity unknown)," unless investigatory evidence suggests which of the EBDCs was used on the product.

Because registrations for zineb have been cancelled, supplies of the chemical may not be available for use as a reference standard. If necessary, another EBDC analytical standard may be used and appropriate molecular weight conversion made to permit reporting residues in terms of ppm zineb.

Significant Figures

The level of each residue that appears at or above its limit of quantitation for the method is calculated (Section 105 discusses limit of quantitation). Residues are calculated and reported in ppm. Unless the quality of the chromatography or other factors necessitates fewer significant figures, levels are reported as follows:

≥100 ppm	to nearest ppm
10 to 99.9 ppm	to nearest 0.1 ppm
1 to 9.99 ppm	to nearest 0.01 ppm
0.010 to 0.999 ppm	to nearest 0.001 ppm

Trace

Residues that are detectable by the method but present at less than the limit of quantitation are reported as "Trace."

Confirmation

Identities of residues are confirmed before reporting, according to the principles discussed in Section 103. Confirmation of nonviolative levels of frequently found residues do not require confirmation in every sample; frequency of confirmation is at the discretion of the laboratory.

104 C: DETERMINING COMPLIANCE WITH REGULATIONS

For monitoring purposes, all residues of the same pesticide are calculated and reported as individual chemicals. For tolerance enforcement purposes, however, the residue definition as stated in the particular regulation applies; in some situations, not all residues are included in the total residue for determining compliance with the tolerance. FDA's Compliance Policy Guide [2] provides criteria that must be met to initiate an enforcement action for violative pesticide residues found in a food commodity. Directions included in this section assume that quantitation has been accurately performed, according to directions in Sections 504 and 606, and that individual residues have been reported into the data system.

General Rule for Multicomponent Residues

Residues that consist of more than one isomer of a technical pesticide, or that consist of parent and degradation products, are added together to determine compliance with existing tolerances, insofar as the degradation products are included in the tolerance expressions of 40 CFR Parts 180, 185, or 186 [3].

Special Situations

BHC. Determining compliance of residues of BHC is complicated by the fact that γ-BHC, also known as lindane, is marketed as a separate pesticide and is also an isomeric component of technical BHC, which may have up to six different isomers. At one time separate tolerances for BHC and for lindane were established, and the possibility existed that both might be used on the same commodity. Currently, U.S. tolerances for BHC have been revoked, but residues are still found in domestic and imported commodities; U.S. tolerances for lindane remain for several commodities.

When residues of BHC isomers are found in a commodity, the quantity of each isomer that is present is calculated against an individual reference standard, according to the general principle; quantities of α, β, and δ isomers are then added together. If the amount of γ-BHC is $<1/3$ the total of α+β+δ, the total of the four isomers is considered to be a residue of BHC. If the γ-isomer is $>1/3$ the total of α+β+δ, the amount in excess of $1/3$ (α+β+δ) is considered to be lindane and the remainder of the γ plus α, β, and δ is considered BHC. Appropriate regulatory action is decided based on these calculations.

Chlordane, Heptachlor, Heptachlor Epoxide. Two factors complicate the residue situation for chlordane: (1) chlordane is a multicomponent mixture whose terminal residue pattern varies considerably, and (2) one component of technical chlordane is heptachlor, which was marketed as a separate pesticide. At the time when both chlordane and heptachlor were registered for food use, the possibility existed that they might be used on the same product. U.S. tolerances for both chlordane

and heptachlor are now revoked, and most residues now occur in fish as a result of lingering environmental contamination, but the procedures developed during the previous period still apply if needed.

Section 504 outlines the prescribed method for quantitating chlordane residues, either against a technical standard or against individual reference standards, depending on the residue pattern. It also specifies that peaks at the retention times of heptachlor and heptachlor epoxide can be included in quantitation of chlordane against the technical standard, if those peaks are relatively small.

If chlordane residues are calculated as individual terminal residues, they are added together to determine total chlordane. If measured against a technical chlordane reference standard, the calculated value is considered total chlordane. In either case, heptachlor and heptachlor epoxide peaks are included as part of total chlordane if they are relatively small and in reasonable proportion to the rest of the residue. If heptachlor and/or heptachlor epoxide are much out of proportion, as shown in Figure 504-d, they are considered as separate residues. Appropriate regulatory action is decided based on these calculations.

PCBs. Regulations related to PCB residues establish tolerances for "PCBs," so compliance is based on total PCBs calculated, regardless of which Aroclor(s) was used as reference standard.

Residues of More Than One Pesticide. In certain cases, pesticides "that cause related pharmacological effects [are] regarded, in the absence of evidence to the contrary, as having an additive deleterious action" [3]. Special rules for adding together residues in these categories may apply.

Residues Calculated From a Derivative. As described above under reporting, some residues can be quantitated only by methods that form a derivative prior to the determinative step. Compliance with regulations in these cases depends on the precise statement of the regulation and on investigatory evidence related to the particular sample. For regulatory purposes, the residue level must be calculated in terms of the chemical(s) specified in the tolerance; if necessary, a conversion is made.

References

[1] Food and Drug Administration (1993) *J. AOAC Int.* **76**, 127A-148A, and previous annual reports

[2] *Compliance Policy Guide*, Section 7141.02, Food and Drug Administration, Rockville, MD

[3] 40 CFR 180.3

105: ANALYTICAL LIMITS OF QUANTITATION

105 A: DEFINITION

FDA defines limit of quantitation (Lq) as the lowest level of residue that can be quantitated by a given method and whose identity can be confirmed in regulatory laboratories operating under routine conditions. Levels less than the Lq are defined as trace.

When MRMs are used, a separate Lq applies to each residue determined by the method because each represents a different analytical situation.

The following factors must be specified in order to define the analytical situation; only then can an Lq be calculated:

1) Analytical method used

2) Sample (matrix) type

3) Sample weight equivalent introduced to the determinative step

4) Sensitivity of the determinative step to the analyte; sensitivity is dependent on the following instrumental conditions:

   a) Determinative technique (In MRMs, the determinative step is usually GLC or HPLC; operational parameters must be defined as part of the method description.)

   b) Range of analyte weight that produces a linear detector response

   c) Overall condition of the system

   d) Amplification and/or attenuation of the detector signal

   e) Characteristics of the signal processing or recording device

   f) Chromatographic elution characteristics of the analyte

105 B: CALCULATION

FDA Lqs for each method are arrived at by (1) specifying a sample weight equivalent to be examined by the determinative step (the amount chosen must be compatible with long-term instrument stability); (2) establishing a recommended determinative step sensitivity that is stable, reproducible, and achievable by all laboratories; and (3) establishing a response equivalent to 10% of full scale deflection (FSD) on the signal-processing device as the minimum considered quantifiable and confirmable. FDA methods applied according to these guidelines are capable of analyzing for most residues at levels well below established tolerances.

Determinative step sensitivity is established by reference to a "marker compound"; *i.e.*, the instrumental parameters are adjusted to cause a specified response to a specified quantity of the marker compound. This approach makes it possible for different laboratories to achieve approximately the same Lq even though the instrument settings may be different for each. Lq for the marker compound can then be calculated with the formula below for any particular method. Lqs for all other compounds recovered through the method will vary according to the determinative step sensitivities for each.

▶ With these guidelines established, Lq for a method is calculated thus:

$$\text{ng 50\% FSD} = \text{ng analyte injected} \times \frac{\text{ng marker specified}}{\text{ng marker injected}} \times \frac{\text{marker peak height}}{\text{analyte peak height}}$$

$$\text{ng 10\% FSD} = \text{ng 50\% FSD}/5$$

$$\text{Lq} = (\text{ng 10\% FSD})/(\text{mg sample injected})$$

▶ Round the Lq result following the guidance for significant figures and reporting analytical results in Section 104, page 104-3. For general purposes, results at or below 0.010 ppm are deemed to have an Lq of 0.010 ppm.

105 C: IMPLEMENTATION

Guidelines for applying analytical methods are required to provide consistency among laboratories performing regulatory analyses. Otherwise, variations in the amount of sample equivalent injected and/or the sensitivity of the determinative step can cause different Lqs in different laboratories. Lqs that result from following FDA guidelines are adequate for the enforcement of tolerances and, in most cases, are sufficient to determine residues below the tolerance level so that data on incidence and levels of residues in foods and feeds can be collected.

The following rules are established to maintain consistent Lqs among FDA laboratories:

▶
- Establish the sensitivity recommended in each determinative step method module (*e.g.*, Section 302 DG1-DG12, Section 401 DL1). Note that the requirement for GC determinations to be based on columns of 100% methyl siloxane is in effect as of FY'98 (October 1, 1997); prior to that time, other DG modules may have been used to calculate Lq.

- Inject a volume of extract containing the equivalent sample weight recommended for each method (*e.g.*, Section 302, Determination).

- If one of the recommended specifications above cannot be achieved, or if changing one is advisable for any reason, adjust the other parameter to maintain the targeted limit of quantitation. Section 105 D describes factors that may cause problems in specific situations.

Table 105-a lists examples of Lqs that can be calculated from the recommended sample weight equivalent and determinative step sensitivity for particular PAM I methods. The list is not exhaustive but does illustrate the way in which the Lq for any method in PAM I can be calculated.

105 D: FACTORS AFFECTING TARGET LIMITS OF QUANTITATION

The following factors, individually or in combination, may reduce the certainty of quantitation and/or identification of a residue in any specific analytical situation. They may also cause the Lq to differ from the recommended limit defined by the formula above and by Table 105-a. Measures taken to compensate for one factor may trigger the influence of another.

1) Determinative step sensitivity to any particular residue. A distinct Lq applies to each residue determinable by a particular MRM, because the sensitivity of the determinative step to each compound may be different.

2) Limited detector sensitivity. Not all individual detectors are capable of reaching the sensitivity specified; in such cases, the Lq will be higher than targeted.

3) Greater detector sensitivity. Directions here recommend sensitivity at which detectors should be operated, even though some are capable of greater sensitivity. However, operation at conditions that produce recommended sensitivity may sometimes be precluded by other disadvantages in detector performance. For example, many models of ^{63}Ni electron capture detectors are not linear at conditions that produce sensitivity of 50% FSD to 1.5 ng chlorpyrifos, as is recommended for other detectors; most are linear, however, at conditions that produce 50% FSD to 0.15 ng chlorpyrifos. The rules in Section 105 C specify that, in this situation, the laboratory should operate at the greater sensitivity in order to work in a linear range, then proportionately reduce the weight of sample equivalent injected in order to maintain Lqs consistent with those achieved by other laboratories.

4) Other improvements that affect determinative step. Wide bore capillary GLC columns (Section 502 C) permit analytes to elute in a tighter band than was possible with packed column chromatography. When detector response is measured in terms of peak height, use of capillary columns results in an apparent improvement of response. Injection of a smaller amount of equivalent sample, as directed in Section 105 C, is appropriate and, at the same time, beneficial to the longevity of the column.

5) Excessive interferences from sample co-extractives. Interferences from sample co-extractives raise the Lq of a method by masking the detector response to the residue or by preventing injection of the specified sample equivalent without undesirable damage to the system. Additional procedures to clean up the sample extract prior to determination may improve the Lq by removing these interferences.

Table 105-a: Examples of Method Specifications Used to Calculate Lqs

PAM I Method[1]		Recommended Mg Injected	Recommended Sensitivity[2]	Lq (marker compound)[3]
302E1+DG2 (FPD-P)		20mg	1.5ng chlorpyrifos	0.015ppm chlorpyrifos
302E3+C1+DG3 (E1CD-X)		20mg	1.5ng chlorpyrifos	0.015ppm chlorpyrifos
302+E1+C3+DL1		116mg	10ng carbofuran	0.017ppm carbofuran
303E1+C1+DG1 (EC)	OR	20mg 2mg	1.5ng chlorpyrifos 0.15ng chlorpyrifos	0.015ppm chlorpyrifos 0.015ppm chlorpyrifos
304E4+C2+DG1 (EC)		10mg (cheese with 30% fat)	1.5ng chlorpyrifos	0.03ppm chlorpyrifos, whole product basis
401E1+C1+DL1		200mg	10ng carbofuran	0.01ppm carbofuran
402E1+C1+DG3 (fatty foods)		5mg Eluate 1	1.5ng chlorpyrifos (0.2ng PCP methylether)	0.008ppm PCP methylether
		10mg Eluate 2	1.5ng chlorpyrifos (0.5ng 2,4,5-T methylester)	0.01ppm 2,4,5-T methylester
402E2+C1+DG3 (nonfatty foods)		10mg Eluate 1	1.5ng chlorpyrifos (0.2ng PCP methylether)	0.004ppm PCP methylether
		20mg Eluate 2	1.5ng chlorpyrifos (0.5ng 2,4,5-T methylester)	0.005ppm 2,4,5-T methylester
403E1+C1+DL3		800mg	40ng diuron	0.01ppm diuron
404E1+DL5		125mg	62.5ng MBC	0.1ppm MBC
404E1+DL7		125mg	6.25ng thiabendazole (fluorescence detector)	0.01ppm thiabendazole

[1] Parenthetical codes indicate the detector used in the GLC determinative step.
[2] Ng marker compound that causes detector response of 50% FSD; where residues targeted by the method are different from the marker compound, weight of example target that caused 50% FSD is also listed.
[3] Calculated by formula in Section 105 B; note that sensitivity is divided by 5 to produce ng causing 10% FSD.