SUMMARY OF TOXIC CHEMICAL AND PESTICIDE RESIDUE PROGRAMS AND NUTRIENT/ CHEMICAL ANALYSES

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The Current Status and Risks of Pesticide Residues in the US Agricultural and Food Industries

To gain a perspective of the current status and risks of pesticide residues in the US agricultural and food industries, there are four main considerations that must be addressed. These four considerations include: (1) the goals of a pesticide residue program, (2) the magnitude and complexity of the problems of persistent chemicals in the environment, (3) the regulatory policies of EPA, FDA and USDA, and (4) the approaches that can be taken to address the first three considerations.

The Goals of a Pesticide Program:

The first consideration, the goal, is relatively straight forward: to insure that all products will be below the tolerance levels for all pesticides and PCBs. As discussed later, it is clear that there is greater concern for the organochlorine pesticides than the organo-phosphate pesticides. The organochlorine or chlorinated pesticides are more persistent, bioconcentrate to a greater degree and are likely to exhibit carcinogenic (cancer causing) and other chronic (long term) effects on human health. By contrast, the organo-phosphate pesticides are metabolized and excreted by animals and humans more rapidly, have lower bioconcentration rates and tend to cause neurotoxic effects only at relatively high levels of intake by humans. Therefore, although the goal is to insure that all pesticides are below tolerance levels, the highest risks and, therefore, the major focus has always been on the organchlorine pesticides.

Magnitude and Complexity of Chemicals in the Environment:

The second consideration complicates the picture considerably since there are literally hundreds of chlorinated pesticides and their derivatives (isomers, metabolites and degradation products) that can enter the food chain and bioconcentrate in poultry and livestock tissues. For each chlorinated chemical used as a pesticide or insecticide, structural isomers may be present that were inadvertently formed during chemical synthesis. Other isomers, as well as metabolites, can be formed <u>in vivo</u> by poultry and livestock who ingest the original pesticide from feed and water. Other isomers and partial degradation products can be formed in the environment (i.e., on plants, in soil or in water) through oxidation, hydrolysis, photolysis and microbial metabolism. Finally, existing chlorinated pesticides that may be unfamiliar presently may be introduced unintentionally into the food chain through their use in a new application. Examples of old pesticides that became popular for new uses and therefore appeared in agricultural products at a later date are mirex and chlorpyrifos. Hence, the number of chlorinated compounds and their derivatives that are of concern to the regulatory agencies total in the hundreds and perhaps in the thousands.

Regulatory Policies for Pesticides:

The third consideration, the USDA/FDA/EPA regulatory policies, further complicate the picture by literally transferring all burden of responsibility for any and all chlorinated residues to companies in the agricultural industry who sell products at all levels of the food chain. The regulatory agencies do not publish a finite list of substances that are regulated. Rather, they publish tolerance levels for certain chlorinated pesticides and state that these tolerance levels include the contributions made by the metabolites or partial degradation products of various pesticides without specifically identifying those metabolites. Further, their authority permits them to take action on any chlorinated residue found in poultry or livestock irrespective of whether it is included in lists published by the agencies. For chemical substances that show up in feed or food products and have no published tolerance levels, the tolerance is established as "zero" (the limit of detectability). A tolerance level (or "action level") above zero may be recommended to FSIS if EPA/FDA consider it to be a situation of "unavoidable contamination". However, regulatory agencies are not inclined to make a determination of "unavoidable contamination" and establish tolerance levels above zero in instances where the contamination occurs within one company or a limited number of companies within an industry.

These statutory authorities and regulatory policies provide a clear mandate to the agricultural industry. Both suppliers and food producers are responsible for maintaining residue levels of all listed pesticides and metabolites below established tolerance levels and, more importantly suppliers and producers are responsible for residue levels of any unlisted chlorinated pesticide, isomer or metabolite. Our past experience indicates that unless the residue levels of substances with no published tolerance levels can be demonstrated to be present in all materials of the same type throughout the industry and the ongoing contamination cannot be avoided or eliminated, the consideration of "unavoidable contamination" is not valid and no temporary action level will be established. This simply means that companies producing and selling products in the food chain are liable and can be held accountable for hundreds of chlorinated substances that exist in the environment as pesticides, structural isomers, metabolites or other chlorinated degradation products inadvertently produced in the environment through oxidation, photolysis, hydrolysis or microbial degradation.

Approaches Diversified Laboratories has Taken to Reduce Risk Under the Status Quo:

Given the background previously described, it is important to consider how the agricultural industry has in general coped with these circumstances. First of all, the USDA maintains a certification program which continually addresses analytical laboratory proficiency for a few limited pesticides and continuously checks and rechecks laboratories for these same compounds. Based on this, most commercial laboratories have adopted a list of 18 to 22 pesticides that are relatively easy to analyze from both a technical and economic point of view. These laboratories generally consider everything else present in a GC chromatogram to be nothing more than interfering substances. In most cases, overly simplistic methodologies are employed for even this limited group of pesticides in order to avoid larger capitol investments for necessary equipment and the ongoing expense of R & D and highly trained chemists. Under these circumstances it is believed the agricultural industry is subject to continual incidents that will prove to be expensive and even disastrous for individual companies on a random basis.

Some time ago, Diversified Laboratories adopted a program, and using Ph.D. level chemists, developed the necessary methodologies to substantially reduce the potential risks associated with the status quo. First, methodologies were researched and developed to increase the likelihood of observing the presence of unfamiliar chlorinated compounds in our routine pesticide screen. Over time unknown substances present at significant levels in the pesticide screens were systematically identified in samples from our clients who range from feed ingredient suppliers to food companies. All compounds which are considered likely to persist and bioconcentrate in the food chain and would be of concern to regulatory agencies are integrated into the routine pesticide screen through an inhouse R & D program. Many of these substances have been listed on our standard certificate of analysis with the balance of these compounds being routinely monitored but not currently listed on the certificate. Both the R & D efforts and the ongoing program require extensive expertise in chemistry and sophisticated equipment such as gas chromatography/mass spectrometry (GC/MS). As a result of these efforts, we offer the most comprehensive pesticide screen available (42 listed and 13 unlisted compounds), or more than twice the conventional screen.

The program is both cumulative in that it includes the chemicals from all past residue contaminants and dynamic in that it addresses all new unidentified substances present at significant levels in current samples. This approach enables the optimum protection of companies by eliminating residue problems caused by unfamiliar substances before they become a regulatory problem. Since it is impossible to develop an economically feasible pesticide screen for hundreds or perhaps thousands of chemicals, it is believed that the risk is substantially reduced by focusing on compounds that have been demonstrated to be present in feeds, feed ingredients or poultry and livestock tissues somewhere in the United States. That is, if it has been found once in a feed or food product, it is reasonable to assume that it has the potential for again finding its way into agricultural and food products at some future date. Diversified Laboratories' program has taken years and a substantial investment in dollars and technical expertise to develop and, more importantly, it is a program that is continually expanding the analytical screen. This approach to substantially reducing the risk of contamination requires a specific scientific focus and an ample sample volume to identify new contaminants and enable the program to be economically feasible.

In summary, it is clear that the task of minimizing the risk of pesticide residue

contamination is formidable. However, it is also clear that a common sense approach and the effective utilization of science and technology can dramatically reduce the risk without sacrificing the economic and logistical goals of companies in the agricultural and food industries.

SUMMARY OF TOLERANCES FOR CHLORINATED PESTICIDES AND OTHER COMPOUNDS

(Currently Included in Diversified Laboratories, Inc. Routine Organochlorine Screen)

	TOLERANCE OR ACTION LEVEL[(1),(2)]						
Pesticide, Isomer, Metabolite or Industrial Chemical	**Fat from Swine**	**Meat ByProduct from Swine**	**Fat from Cattle**	**Meat ByProduct from Cattle**	**Fat from Poultry**	**Meat ByProduct from Poultry**	**Notes**
BHC, alpha isomer[(3)]	0.3	---	0.3	---	0.3	---	action level
BHC, beta isomer[(3)]	0.3	---	0.3	---	0.3	---	action level
BHC, delta isomer[(3)]	0.3	---	0.3	---	0.3	---	action level
Lindane, (which is the gamma isomer of BHC).	4	---	7	---	4	---	action level (poultry)
p,p'-DDT[(4)]	5	---	5	---	5	---	action level
p,p'-DDD[(4)]	5	---	5	---	5	---	action level
p,p'-DDE[(4)]	5	---	5	---	5	---	action level
o,p'-DDT[(4)]	5	---	5	---	5	---	action level
o,p'-DDD[(4)]	5	---	5	---	5	---	action level
	TOLERANCE OR ACTION LEVEL[(1),(2)]						
Pesticide, Isomer, Metabolite or Industrial Chemical	**Fat from Swine**	**Meat ByProduct from Swine**	**Fat from Cattle**	**Meat ByProduct from Cattle**	**Fat from Poultry**	**Meat ByProduct from Poultry**	**Notes**
o,p'-DDE[(4)]	5	---	5	---	5	---	action level
Aldrin[(5)]	0.3	---	0.3	---	0.3	---	action level
Dieldrin[(5)]	0.3	---	0.3	---	0.3	---	action level
Endrin	0.3	---	0.3	---	0.3	---	action level
Endrin aldehyde	---	---	---	---	---	---	a metabolite of Endrin

Endrin ketone	---	---	---	---	---	---	a metabolite of Endrin
Hexachlorobenzene (HCB)	0.5	---	0.5	---	0.5	---	action level
Heptachlor[6]	0.2	0.2	0.2	0.2	0.2	0.2	action level
Heptachlor Epoxide[6]	0.2	0.2	0.2	0.2	0.2	0.2	action level

	TOLERANCE OR ACTION LEVEL[(1),(2)]						
Pesticide, Isomer, Metabolite or Industrial Chemical	Fat from Swine	Meat ByProduct from Swine	Fat from Cattle	Meat ByProduct from Cattle	Fat from Poultry	Meat ByProduct from Poultry	Notes
Toxaphene	7	---	7	---	7	---	action level
Strobane	---	---	---	---	---	---	
Chlordane, alpha isomer[(7)]	0.3	---	0.3	---	0.3	---	action level
Chlordane, gamma isomer[(7)]	0.3	---	0.3	---	0.3	---	action level
Chlordene, alpha isomer[(7)]	0.3	---	0.3	---	0.3	---	action level
Chlordene, gamma isomer[(7)]	0.3	---	0.3	---	0.3	---	action level
cis-Nonachlor[(7)]	0.3	---	0.3	---	0.3	---	action level
trans-Nonachlor[(7)]	0.3	---	0.3	---	0.3	---	action level
Oxychlordane[(7)]	0.3	---	0.3	---	0.3	---	action level

	TOLERANCE OR ACTION LEVEL[(1),(2)]						
Pesticide, Isomer, Metabolite or Industrial Chemical	Fat from Swine	Meat ByProduct from Swine	Fat from Cattle	Meat ByProduct from Cattle	Fat from Poultry	Meat ByProduct from Poultry	Notes

Methoxychlor	**3**	**---**	**3**	**---**	**3**	**---**	**action level (poultry)**
Mirex	**0.1**	**0.1**	**0.1**	**0.1**	**0.1**	**0.1**	**action level**
Perthane	**---**	**---**	**---**	**---**	**---**	**---**	
PCNB (pentachloronitrobenzene)	**---**	**---**	**---**	**---**	**---**	**---**	
Chlorobenside	**---**	**---**	**---**	**---**	**---**	**---**	
Chlorothalonil	**---**	**---**	**---**	**---**	**---**	**---**	
Chlorpyrifos and metabolite[8]	**0.5**	**0.5**	**2**	**2**	**0.5**	**0.5**	
Methyl Chlorpyrifos[9]	**0.5**	**0.5**	**0.5**	**0.5**	**0.5**	**0.5**	

	TOLERANCE OR ACTION LEVEL[1],[2]						
Pesticide, Isomer, Metabolite or Industrial Chemical	**Fat from Swine**	**Meat ByProduct from Swine**	**Fat from Cattle**	**Meat ByProduct from Cattle**	**Fat from Poultry**	**Meat ByProduct from Poultry**	**Notes**
Chlorpyrifos Methyl Oxygen Analog (metabolite of Methyl Chlorpyrifos)[9]	**0.5**	**0.5**	**0.5**	**0.5**	**0.5**	**0.5**	
Carbophenothion (Ethyl Trithion)	**0.1**	**---**	**0.1**	**---**	**0.1**	**---**	
Ethion	**0.2**	**0.2**	**2.5**	**1.0**	**0.2**	**0.2**	
Parathion[10]	**---**	**---**	**---**	**---**	**---**	**---**	
Ronnel	**3**	**2**	**10**	**4**	**0.01**	**0.01**	
Endosulfan I and metabolite	**0.2**	**0.2**	**0.2**	**0.2**	**---**	**---**	
Fonophos (Dyphonate)	**---**	**---**	**---**	**---**	**---**	**---**	
PCB's (polychlorinated biphenyls)	**---**	**---**	**---**	**---**	**3**	**---**	**action level**

FOOTNOTES:

1. **Note - the absence of a specific value indicates that a "zero tolerance" currently exists for the pesticide in the material listed. A zero tolerance (or no tolerance) is the usual enforcement level for FDA where no official tolerance has been set and no action level exists.**

2. **Action level is an administratively established level set by FDA for purposes of regulatory action where an official tolerance has not been established through the "Code of Federal Regulations" (CFR) and where a zero tolerance is not deemed feasible for economic reasons.**

3. **The tolerance for BHC is the sum of the isomers alpha-BHC, beta-BHC, and delta-BHC.**

4. **The tolerance for these isomers is applied on a Total DDT basis which is the sum of all of the six isomers and metabolites listed.**

5. **Aldrin and Dieldrin have been enforced in the past as 0.3 ppm action level for the combined concentrations of these two compounds.**

6. **Heptachlor and Heptachlor Epoxide are enforced as 0.2 ppm action level for the combined concentrations of these two compounds.**

7. **The tolerance for chlordane is 0.3 ppm for the sum of all of the following isomers: alpha-Chlordane, gamma-Chlordane, alpha-Chlordene, gamma-Chlordene, cis-Nonachlor, trans-Nonachlor, and Oxychlordane.**

8. **Chlorpyrifos and it's metabolite are enforced as action levels for the combined concentrations of these two compounds. Note that due to the unavailability of stable standards for the metabolite, the only means currently available for the confirmation of the presence of the metabolite is through Gas Chromatography/Mass Spectrometry analysis.**

9. **Methyl Chlorpyrifos and it's metabolite Chlorpyrifos Methyl Oxygen Analog are enforced as 0.5 ppm action level for the combined concentrations of these two compounds.**

10. **Parathion has been regulated on an action level in the past but recently a zero tolerance has been adapted.**

SUMMARY OF TOLERANCES FOR ORGANO-PHOSPHATE PESTICIDES

(Currently included in Diversified Laboratories, Inc. routine organophopshate screen)

	TOLERANCE OR ACTION LEVEL[(1),(2)]						
Pesticide, Isomer, Metabolite or Industrial Chemical	**Fat from Swine**	**Meat ByProduct from Swine**	**Fat from Cattle**	**Meat ByProduct from Cattle**	**Fat from Poultry**	**Meat ByProduct from Poultry**	**Notes**
Parathion	---	---	---	---	---	---	
Methyl Parathion	---	---	---	---	---	---	
Ethion	0.2	0.2	2.5	1.0	0.2	0.2	
Carbophenothion	0.1	---	0.1	---	---	---	
Malathion	4	4	4	4	4	4	
Ronnel	3	2	10	4	0.01	0.01	
Diazinon	---	---	0.7	0.7	---	---	
Disulfoton	---	---	---	---	---	---	
Phorate	0.05	0.05	0.05	0.05	0.05	0.05	
Meta Systox R	---	---	---	---	---	---	

FOOTNOTES:

(1) Note - the absence of a specific value indicates that a "zero tolerance" currently exists for the pesticide in the material listed. A zero tolerance (or no tolerance) is the usual enforcement level for FDA where no official tolerance has been set and no action level exists.

(2) Action level is an administratively established level set by FDA for purposes of regulatory action where an official tolerance has not been established through the "Code of Federal Regulations" (CFR) and where a zero tolerance is not deemed feasible for economic reasons.

(3) Parathion has been regulated on an action level in the past but recently a zero tolerance has been adapted.

Summary of Quantitative Reporting Limits and Detectability Limits in Routine Chlorinated Pesticide and PCB Screens for Food Grade and Feed Grade Products (ppm)

	Adipose Tissue		Fats & Oils		Feed & Ingredients		
	Reporting Limit(1)	Detect. Limit(2)	Reporting Limit(1)	Detect. Limit(2)	Reporting Limit(1)	Detect. Limit(2)	
BHC isomers	0.01	0.005	0.01	0.005	0.002	0.001	
Lindane	0.01	0.005	0.01	0.005	0.002	0.001	
HCB	0.01	0.005	0.01	0.005	0.002	0.001	
Heptachlor	0.01	0.005	0.01	0.005	0.002	0.001	
Heptachlor Epoxide	0.01	0.005	0.01	0.005	0.002	0.001	
Aldrin	0.01	0.005	0.01	0.005	0.002	0.001	
Dieldrin	0.01	0.005	0.01	0.005	0.002	0.001	
Endrin	0.01	0.005	0.01	0.005	0.002	0.001	
Endrin Aldehyde	0.05	0.025	0.05	0.025	0.010	0.005	
Endrin Ketone	0.05	0.025	0.05	0.025	0.010	0.005	
DDT isomers	0.01	0.005	0.01	0.005	0.002	0.001	
Ronnel	0.02	0.01	0.02	0.1	0.005	0.002	
Chlordane (Technical)(3)	0.10-0.20	0.05-0.10	0.10-0.20	0.05-0.10	0.05-0.10	0.02-0.05	
Toxaphene (3)	2.5-5.0	1.0-2.50	2.5-5.0	1.0-2.50	0.5-1.0	0.2-0.5	
Strobane (3)	2.5-5.0	1.0-2.50	2.5-5.0	1.0-2.50	0.5-1.0	0.2-0.5	
Methoxychlor		0.10	0.05	0.10	0.5	0.025	0.01
Mirex		0.05	0.02	0.05	0.02	0.010	0.005

Summary of Quantitative Reporting Limits
and Detectability Limits in Routine Chlorinated Pesticide
and PCB Screens for Food Grade and Feed Grade Products (ppm)

Continued:

	Adipose Tissue		Fats & Oils		Feed & Ingredients	
	Reporting Limit(1)	Detect. Limit(2)	Reporting Limit(1)	Detect. Limit(2)	Reporting Limit(1)	Detect. Limit(2)
Endosulfan I	0.02	0.01	0.02	0.01	0.005	0.002
PCBs(4)	0.5-1.0	0.2-0.5	0.5-1.0	0.2-0.5	0.1-0.2	0.05-0.1
Nonachlor isomers	0.01	0.005	0.01	0.005	0.002	0.001
Oxychlordane	0.01	0.005	0.01	0.005	0.002	0.001
Chlorpyrifos	0.01	0.005	0.01	0.005	0.002	0.001
Chlorpyrifos Oxygen Analog	0.01	0.005	0.01	0.005	0.002	0.001
Methyl Chlorpyrifos	0.01	0.005	0.01	0.005	0.002	0.001
Chlordene isomers	0.01	0.005	0.01	0.005	0.002	0.001
Chlordane isomers	0.01	0.005	0.01	0.005	0.002	0.001
Carbophenothion	0.05	0.025	0.05	0.025	0.010	0.005
Ethion	0.05	0.025	0.05	0.025	0.010	0.005
Parathion	0.05	0.025	0.05	0.025	0.010	0.005
Chlorbenside	0.01	0.005	0.01	0.005	0.002	0.001
PCNB isomers	0.01	0.005	0.01	0.005	0.002	0.001
Perthane	0.05	0.025	0.05	0.025	0.010	0.005
Chlorothalonil	0.05	0.025	0.05	0.025	0.010	0.005
Fonophos (Dyphonate)	0.05	0.025	0.05	0.025	0.010	0.005

Summary of Quantitative Reporting Limits and Detectability Limits in Routine Chlorinated Pesticide and PCB Screens for Food Grade and Feed Grade Products (ppm)

Continued:

(1) **Reporting limit - represents lowest value in parts per million (ppm) for reporting quantitative values in routine pesticide and PCB screen.**

(2) **Detectability limit - represents lowest value in parts per million (ppm) normally detected in routine pesticide and PCB screen. Values below levels shown are routinely reported as "none detected" or "N.D.".**

(3) **Range due to multiple-isomer nature of the substance and the characteristic presence of other compounds.**

(4) **Range due to different Arochlor pattern sensitivities and number of isomers present in a given Arochlor or other chlorinated biphenyl product.**

Summary of Quantitative Reporting Limits and Detectability Limits in Routine Organophosphate Screen for Food Grade and Feed Grade Products (ppm)

	Adipose Tissue		Fats & Oils		Feed & Ingredients	
	Reporting Limit(1)	Detect. Limit(2)	Reporting Limit(1)	Detect. Limit(2)	Reporting Limit(1)	Detect. Limit(2)
Parathion	0.08	0.05	0.08	0.05	0.03	0.025
Methyl Parathion	0.08	0.05	0.08	0.05	0.03	0.025
Ethion	0.08	0.05	0.08	0.05	0.03	0.025
Carbophenothion	0.08	0.05	0.08	0.05	0.03	0.025
Malathion	0.12	0.10	0.12	0.10	0.08	0.05
Ronnel	0.02	0.01	0.02	0.01	0.01	0.005
Diazinon	0.08	0.05	0.08	0.05	0.03	0.025
Disulfoton	0.30	0.25	0.30	0.25	0.15	0.12
Phorate	0.30	0.25	0.30	0.25	0.15	0.12
Meta-Systox	3.0	2.0	3.0	2.0	2.0	1.0

(1) Reporting Limit - represents lowest value in parts per million (ppm) for reporting quantitative values in routine pesticide screen values below reporting limit but above detectability limit are reported as "less than ...."
(2) the value of the reporting limit.

(2) Detectability Limit - represents lowest value in parts per million (ppm) normally detected in routine pesticide screen. Values below levels shown are routinely reported as "none detected" or "N.D.".

The Use of Trap Greases in Feed Grade Fats

The use of trap grease as a component of feed grade fats has increased substantially during the past several years. The term "trap grease" has been used to describe a wide range of fat soluble materials that are recovered from waste water streams primarily to comply with the US EPA's standards for oil and grease in water discharged from commercial facilities.

There can be significant risks associated with the use of trap greases as a result of their contamination with toxic substances. Moreover, the consistency and variability in the components which may find their way into trap greases presents even greater risks in the absence of a rigorous contamination quality control program. Further, there is the possibility that trap greases will contain significant amounts of fat soluble material other than normal (triglyceride) fat or that the glyceride and fatty acids have been chemically altered. In this case the energy value for poultry and livestock also will be reduced. This may significantly effect the efficacy of a finished feed grade fat product in the absence of an extended quality control program.

To reduce the likelihood of contamination, three important steps should be taken. First, a thorough investigation of the trap grease sources should be undertaken. This includes obtaining information on the type of facility that is providing the trap grease, the processes conducted at the facility and the types of materials that may be introduced into waste water streams either purposefully or inadvertently. Secondly, a quality control program should be established to monitor for the presence of contaminants that may be present and are of concern to regulatory agencies. Although it is impossible to conduct routine testing for all possible substances that may be present, Diversified Laboratories has been working on a group of organic and inorganic substances that are of principal concern to regulatory agencies and that may be present in trap greases separated from waste water streams. It is believed that a generic list of substances can be included in a routine screen and may be customized for particular facilities by supplementing this basic contamination group with chemicals actually used by a facility that are identified during the investigations of sources of trap grease. Third, a quality control program should be established to monitor the efficacy of the material as an energy source for poultry and livestock. This can be accomplished by conducting key analyses to determine the chemical composition and, if necessary, the metabolizable energy level of the material to insure that there are no significant changes among shipments.

The development of analytical groups for both contamination and efficacy has been underway at Diversified Laboratories for the past year. Although many tests for contamination are immediately available more definitive groups specific to trap greases will be available in 1995. It is our objective to provide the most comprehensive trap grease screens possible.

The Presence of "Polymers" in Feed Grade Fats and Oils

For many years it was believed that shortenings used in deep fat frying and other sources of feed grade fats and oils formed polymers that were detrimental to the use of restaurant greases for poultry and livestock. Buyers, particularly in European countries, with a concern for polymers used an analytical methodology that quantified the "non-elutable fraction" to determine the degree of polymerization of fats and oils. Their concerns created problems for U.S. exporters of fats and oils.

Research efforts by Diversified Laboratories resulted in the development of a sophisticated analytical method to identify the "polymeric" material and quantify the levels present. Diversified Laboratories subsequently analyzed restaurant greases supplied by the National Renderers Association under a grant from the Fats and Proteins Research Foundation. Further, in a collaborative effort with the University of Georgia, studies were conducted to determine the biological significance of the various "polymeric substances" present.

The results of these studies clearly demonstrated the nonspecificity and other weaknesses of the "non-elutable" method for estimating polymer content and the nutritional value of fats and oils. The data demonstrated that feed grade fats and oils including used restaurant greases, do not contain true polymers. The "polymerized" material was shown to be oligomers or "baby polymers". Moreover, the studies showed that the oligomers are not always present at high levels. Finally, the studies showed that certain portions of the various oligomer compounds could be utilized as a source of energy by poultry and livestock and that it is necessary to conduct qualitative and quantitative analyses of oligomers to determine their effect on the metabolizable energy level of the fat in question.

It has been recommended that fat suppliers conduct an "audit" of oligomer levels in their raw materials to insure that the oligomers in their finished products are low and consistent. If the oligomer levels are significant, it is recommended that the metabolizable energy value be determined to assess the impact on the finished products. Once a comprehensive audit has been completed, the approach to a low cost quality control program can be developed. The quality control program will be customized to meet the individual needs of the company or its specific plants.

OLIGOMERS AND OTHER POLYMERIC SUBSTANCES FOUND IN RESTAURANT GREASES

Free fatty acid dimers

Free fatty acid trimers

Free fatty acid tetramers

Free fatty acid pentamers
and true polymers

Triglyceride dimers

Triglyceride trimers

Triglyceride tetramers

Triglyceride pentamers
and true polymers

Diglyceride dimers and free
fatty acid / triglyceride complexes

SUMMARY OF QUALITY CONTROL/QUALITY ASSURANCE IN ANALYTICAL PROCEDURES

Toxic Hydrocarbon Analysis

With respect to pesticide and other toxic hydrocarbon residues, Diversified Laboratories has established a very stringent set of quality control procedures to ensure the reporting of scientifically valid results both qualitatively and quantitatively. First, an appropriate extraction and cleanup procedure is selected or developed consistent with the analyte(s) in question, the sample matrix and the objectives of the analytical results. Second, to ensure that the extraction and cleanup procedures do not result in a loss of the compounds of concern in a sample, every sample is "spiked" with an appropriate substance not found in the environment or the food chain to serve as a marker and ensure that good recovery of all unknown substances is achieved. These "internal standards" are routinely analyzed and quantified in each sample to ensure no inadvertent loss of the substances of concern. Third, sample matrices identical to those analyzed are also "spiked" with quantitative levels of the analytes in question to monitor the recovery of each compound of interest in each analytical run conducted. Fourth, all analyses are subjected to duplicate analyses to verify qualitatively and quantitively the presence of any substance or in the case of our existing screen, any of the 42 compounds listed as pesticides or the 13 other problem compounds that are also routinely monitored. Fifth, a constant vigilance is maintained regarding any unidentified compounds present and, with the approval of the client, appropriate procedures are implemented to identify and quantify the chemical substance or substances and attempt to determine the toxicological and regulatory significance immediately. Finally, in instances where it is necessary, confirmatory procedures are implemented using chemical techniques, multiple GC analyses, and GC/MS methodologies to confirm suspect chemicals both qualitatively and quantitatively.

Lipid Chemistry and Proximate Analyses

Because of the wide variation in results of individual analyses for many procedures, Diversified Laboratories has utilized the routine practice of conducting the analysis of problem procedures in duplicate. This unprecedented approach to the analysis of agricultural and food products has been in force for over twelve years. Although the other standard QA/QC procedures such as blank samples, external standards and check samples are followed, it is the duplicate analysis that insures the accuracy and precision of the results. This is extremely important in such tests as AOM stability, moisture, insolubles and unsaponifiable matter in fats and oils as well as proximate analyses of byproduct meals for protein, moisture, fat, calcium and phosphorus.

Partial List of Accomplishments in the Area of Pesticide/Hydrocarbon Residue Contamination

1. **Rapid response to pesticide contamination in national integrated turkey operation. Demonstrated significant errors in results by USDA laboratories and significantly reduced the scope of an expensive (approximately $2.5 million) recall of turkey products. Involved over 350 samples in seven day time period.**

2. **Rapid response to U.S. multinational feed company faced with a seven state recall of feed in Northeastern U.S. Demonstrated "false positive" analytical results on feed samples by another commercial laboratory and avoided the expensive seven-state recall. Laboratory work completed within a 48-hour time period over a weekend.**

3. **Rapid development of a special pesticide analytical method to demonstrate the presence of a specific pesticide on two flies (yes two flies). This was only evidence a large national meat-packer had to prove to USDA that an illegal pesticide had not been used in their packing plant. The analytical results demonstrated that USDA's challenge of the company's use of an illegal pesticide was valid.**

4. **Rapid response as an authorized USDA laboratory to identify and contain pesticide contamination in turkey and broiler flocks throughout Pennsylvania, Virginia and North Carolina caused by fat products from an East Coast renderer.**

5. **Rapid response to accidental spillage of a large volume of the toxic substances atrazine and metolachlor in Virginia stream by an international pharmaceutical company. Results were reported within 36 hours, including the development of a method to analyze water and soil samples. The contamination was restricted to a small segment of the stream and adjacent land.**

6. **Conducted special research and development project for multinational food company to evaluate feasibility of the use of preen gland fluid as a marker for chlorinated hydrocarbon residues in turkey tissues.**

7. **Identification of di-isooctyl phthalate, a carcinogenic (cancer-causing) agent, in broiler tissues of a large integrated broiler operation. With the aid of gas chromatography-mass spectrometry, the substance was positively identified and traced to the source of contamination and eliminated from the operation within ten days of the discovery of the unidentified compound. This avoided a potential human health problem throughout the east coast of the United States.**

8. **Prevented a threatened shutdown of a large Canadian rendering operation by the Canadian Government due to contamination of animal by products with chlorinated hydrocarbons (chlorinated dibenzo-p-dioxins). Diversified Laboratories demonstrated erroneous results generated by the Canadian Government Laboratories, avoided shutdown of the rendering plant and established ongoing pesticide/PCB/chlorinated dioxin quality control program approved by the Canadian Government.**

9. **Resolved problem of unidentified compounds which were continuously reported as pesticides by other commercial laboratories and found to be present in certain feed grade fat blends. Comprehensive research project determined that the unidentified substances were not pesticides but five isomeric compounds derived from the antioxidant component, tert butyl hydroquinone (TBHQ) which appear in certain types of fat blends under specific conditions.**

10. **Expeditiously resolved significant problem of organophosphate pesticide contamination of animal byproducts from a major national rendering company. Diversified Laboratories collaborated with the USDA on the part of the company, identified the source of the organophosphate pesticides and the contamination source was removed from the operations with no penalties or business interruption for the company.**

11. During the past decade, Diversified Laboratories has encountered illegal levels of pesticides in various products ranging from feed ingredients to food products. In every instance where the discovery was made through an on-going quality control program, USDA/FDA involvement and any significant cost to the company as a result of the contamination were avoided. In instances where these compounds were unidentified substances not present in the traditional pesticide screen, Diversified Laboratories has conducted research to include them in its routine pesticide screen.

12. To date, Diversified Laboratories has developed the most comprehensive chlorinated pesticide screen available in the United States including 42 individual pesticides, isomers, or metabolites. This represents approximately 2 to 2 1/2 times the number of pesticides included in the typical pesticide screen. The majority of the 42 pesticides are compounds that have been identified in feed ingredients, poultry and livestock tissues or final food products throughout the United States during the past seventeen years.

13. Provided laboratory support for the pesticide contamination problem encountered by Mississippi integrated poultry producers. Problem was responsible for an estimated $20 million dollar loss by these companies.

14. Identified the presence of polychlorinated biphenyls (PCBs) in poultry tissues and traced the contamination back to Peruvian fish meal containing heat exchanger fluids. Contamination, which included large number of poultry operations in the Southeast, resulted in $50 million dollar lawsuit with the manufacturer of the heat exchanger product.

15. Conducted pharmacokinetics research on the quantitative relationship between the oral dose and exposure period of specific pesticides and the corresponding pesticide level in the tissues of poultry. This information enables Diversified Laboratories to provide its clients with guidance on appropriate action in the event of a pesticide contamination problem.

16. Identified and quantified the presence of polychlorinated biphenyls in the poultry tissues of a major integrated turkey producer and traced the source of contamination to a principal supplier of feed grade fat in the southeastern portion of the U.S. Definitive data and rapid response avoided a financially disastrous contamination of large numbers of poultry and livestock throughout the southeast.

Partial List of Accomplishments
in the Area
of Non-Pesticide Research & Development

1. **Collaborative research studies with Kansas State University regarding the effect of dietary fat on the total fat and fatty acid composition of beef cattle fed fats of different chemical composition.**

2. **A presentation of information to the U.S. Food and Drug Administration (FDA) regarding the use of tall oil fatty acids in feed grade fat blends throughout the south and southwest. This project included the development of a analytical method specific for abietic acids, which serve as the "marker" for the presence of tall oils in feed grade fat products and the cessation of its use in feed grade fats and oils.**

3. **A collaborative study with U.S. Department of Agriculture for the development of a high performance thin layer chromatographic (HPTLC) method for a more definitive (i.e., highly specific) analysis for sulfa drug residues in turkey tissues such as muscle, liver and kidney. The high degree of specificity of this methodology provides more precise residue levels for sulfa in the tissues as well as enabling lower cost shipment of the tissue since continual freezing during transit to the laboratory is no longer necessary.**

4. **Conducted research work for the American Fats and Protein Research Foundation (FPRF) for the identification of the "non-elutable fractions" present in restaurant greases which were previously thought to be polymers. Diversified Laboratories' research demonstrated that the materials were not polymers but oligomers, generally present at low levels which paved the way for an improvement in the export of fats to western Europe by the National Renderers Association (NRA) and its members.**

5. **Collaborative study with University of Georgia to demonstrate the impact of the fatty acid oligomers and glyceride oligomers present in restaurant greases on the metabolizable energy value of these materials in poultry and livestock rations. Diversified Laboratories developed special chemical and analytical methods to measure the utilization of oligomers, and, hence, their impact on the metabolizable energy value of feed grade fats and oils.**

6. **Conducted research studies for a large integrated broiler company for comparisons of body composition among competitive chicken products sold for human consumption. The research included the development of analytical methodologies to determine small but statically significant differences in fat percentages and fatty acid composition of various edible tissues. The results of this research project were required to meet the standards of the Federal Communications Commission (FCC) in order to be used as a part of a large regional advertising campaign through various advertising media.**

7. **Development of new analytical methodology for qualitative and quantitative analysis of low levels of the toxic chemical ethylene glycol to evaluate the potential contamination of edible soybean oils for a major food producer.**

8. **Development of proprietary predictive methodology for the estimation of the metabolizable energy values of feed grade fats and oils.**

9. **Development of specialized analytical methodologies to serve as quality control measures in the commercial production of fat/alginate pellets for use as an energy supplement in dairy feeds.**