Present were:

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Nancy Thiex opened the meeting. After introductions the following issues were covered:

Approval of Agenda – The agenda was approved with the following additions:

- Uncertainty Calculations
- AAFCO Strategic Planning inquiry
- Ankom Fat and Fiber methods.

Approval of Minutes from the September Meetings – Minutes were approved.
Response form Dr. Vaughn, FDA – Thiex reported that she had received a copy of Dr. Vaughn’s response to Judy Thompson, dated January 13, 2006. Linda Benjamin explained that FDA is interested in working with AOAC and AAFCO on upgrading methods and outlined the approach for the cooperation as follows:

AAFCO and AOAC determine and prioritize the methods needing to be updated. A “Method Needs” summary is developed for high priority analytes and sent to interested parties including the Sponsors of the drugs in question for input.

- A method coordinator (MC) is selected to coordinate the update of a method for a specific analyte. The MC
  - Identifies one of the Sponsors for the drug in question that is willing to assist in the process. The FDA recommended working with an original Sponsor rather than a “generic” sponsor. Due to the FDA’s sizeable generic submission queue, a faster review cycle is possible with an original Sponsor versus a generic Sponsor.
  - Surveys the methods available
  - Drafts a method based on the inputs received. Perform an in-house evaluation and validation of the method.
  - Writes a validation protocol that includes the in-house validation data. Submits the protocol to FDA (ONADE, Mr. Marmane) and AOAC for review. The submission to FDA should include a letter from the Drug Sponsor asking FDA to review the protocol located in the AAFCO Masterfile (referencing the file number provided by FDA) in connection with the Sponsor’s NADA for the purpose of the method becoming either a replacement of the current official method or an alternate method.
  - Updates the protocol in response of feedback received
  - Manages the collaborative study
  - Writes the Study report and submits it to AOAC and FDA for approval
  - Makes the requested changes and submits the report to AOAC for final approval

- The validated method will be stored in an AAFCO Masterfile

Dr. Benjamin explained that FDA will NOT review any other aspects of a Sponsor’s packet as part of the method update process. She said that following Mary Ledbetter’s retirement the Feed/Topical team’s responsibilities will be covered by current staff on a rotating basis – first Linda Benjamin, then Mike Oehlsen followed by Becky Owen and Charles O’Brien. Dr. Benjamin finally recommended that questions about this program as well as suggestions should be directed to Mr. William Marnane.

It was agreed to initiate the process with the method for sulfamethazine validated by Bob Smallidge. Dr. Mark Coleman, Elanco offered to be the participating Sponsor. Howard Campbell’s method for salinomycin, narasin and monensin was also discussed. The decision was made to delay initiating the process on this method since it would require the cooperation of multiple sponsors.

**ACTION ITEM:**

- Victoria Siegel to attempt to locate the original raw data associated with the sulfamethazine study

**Pyrrolizidine alkaloids** – Rob Jeppsen summarized the situation surrounding the pyrrolizidine issue. This group of alkaloids represents the toxins present in comfrey but also the alkaloids present in
approx. 460 other plants. Pyrrolizidine alkaloids can be quantified by GC/MS using fused silica columns. Dr. Larry Walker with University of Mississippi specializes in this subject and would be a good source of information about the specific alkaloid “fingerprint” for comfrey. However, there are also simple spot tests for determining the presence of the targeted alkaloids.

Ben Jones offered to put together a “Needs Statement” for a method.

**ACTION ITEM:**
- Ben Jones to put together a “Needs Statement” for a method.

**Carbohydrate characterization** – Lars Reimann discussed the interest of the equine feed and pet food industry in determining the amount of easily available carbohydrates in feeds and feed ingredients as part of a process to modulate the physiological response to a given feed (similar to estimating the “glycemic index” for a given food). Some labs claim to have proprietary methods addressing this issue and Megazyme is developing a test kit as well. Within the two industries there is not yet agreement as to what parameters should be monitored (e.g., total starch, gelatinized starch, Non-Structural Carbohydrates etc.)

**ACTION ITEM:**
- “Needs Statement” must be developed.

**Lasalocid Collaborative Study** – Charlie Focht had submitted a written update dated 1/10/06. In light of the difficulties experienced by participating labs in separating the 5 homologs the question was raised if they all had significant biological activity (e.g., could the 4 minor components be ignored).

**ACTION ITEM:**
- Focht to gather information concerning the biological activity of individual homologs.
- Focht to check calculation and submit study for review.

**Oxytetracycline Collaborative Study** – Richard Larson had submitted a written update dated 1/13/06. He reported that he had sent samples to 17 labs, that two labs had withdrawn from the study, and that he was still missing results from 6 labs. EDTA concentration in the mobile phase is an issue (too little leads to poor response, too much leads to poor separation (tailing))

**ACTION ITEM:**
- Larson to follow up with the 6 missing labs
- Larson & Thiex to compile and evaluate the results and, if applicable, write the Study Report and submit it to AOAC for review.

**Multi-Analyte Mycotoxin Methodology** – Laszlo Torma reported that only 1 method had been submitted for consideration. It has been requested that the method be further expanded. Extraction and clean-up are still a challenge. The determination of T2 toxins is a major issue. The goal is to generate methods that can be used by many labs potentially including methods based on ELISA and LC/MS/MS technologies.

**ACTION ITEM:**
• Issue a call for more method suggestions

**Neomycin Collaborative Study** – Thiex reported that the “Method Needs” statement has been reviewed by FDA (Dennis McCurdy), a Sponsor (Alex Macdonald, Phibro) and Harold Campbell, CFIA. MacDonald reviewed the drug chemistry. While the USP reference material contains only a single active ingredient all commercial products contain two active components. The standard offered by Sigma contains both components and can be used as reference material and for system suitability evaluations. He also reported that an ELISA method currently available seems to work well. **Thiex reported that her laboratory is working on validating an HPLC method based on OPA post column derivatization.**

**ACTION ITEM:**

• Thiex will report on the feasibility/progress on SLV for neomycin in feed at the next AAFCO Meeting.

**Decoquinate Collaborative Study** – Thiex reported that the “Method Needs” statement has been reviewed by FDA (Dennis McCurdy) and Harold Campbell, CFIA. Aaron Price reported that the method will likely be accepted in the EU while the method’s status in the US is somewhat uncertain pending the allocation of resources to take it through the FDA/AOAC approval process. Sondra Flick, Alpharma agreed to accept the sponsor role part of the method development. Price reported that Anabil Santhez will be spearheading the validation project within CFIA.

**ACTION ITEM:**

• Anabil Santhez will draft validation protocol for FDA, Alpharma and AOAC review and comments.

**Chlortetracycline Collaborative Study** – Thiex reported that the “Method Needs” statement has been accepted. The current method under development is more prone to back pressure than the OTC method. Reduction in the EDTA concentration leads to peak broadening. While CTC has multiple peaks the components behind the minor peaks are not thought to have any biological activity. Comparability has been established between micro and HPLC based data.

**ACTION ITEM:**

• Jane Sabbatini, Eurofins Scientific will work together with Scott Panagiottis, PennField on the optimization and evaluation of a method and the development of a protocol.

**Non-Nutritive Elements Methodology** – Thiex reviewed a spreadsheet with consolidated information and a very rough draft of a “Needs Statement”. A discussion of the applicability of the title followed with the suggestion to include all elemental analyses. There was agreement that such an approach would require multiple methods differing in sample prep, solubilization and detection technologies. It was suggested to consider the use of performance based methodologies similar to those proposed by EPA.

**ACTION ITEM:**

• Thiex and Brenda Snodgrass to work further on “Needs Statement”.

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• Lars Reimann to provide information about level of interest from sources outside the US.
• Info about levels of concern should be forwarded to Thiex.

Virginiamycin Method Update – MacDonald questioned the use of HPLC for virginiamycin based on personal experience and a review published in 1991 in The Analyst. The problem is that the commercial form of virginiamycin consists of many components with the primary ones being M1 and S1 and that the components seem to interact in a synergetic fashion making it very difficult to correlate a chemical analysis by HPLC to a biological response. All current assay methodologies are activity based. (turb, plate). Residues are determined based on the presence of the M1 factor for which there are standards commercially available. A method developed by CFIA has a detection limit in the low ppb range. Assay methods based on Biacore technology may be an option.

ACTION ITEM:
• None – this analyte will be removed from consideration until the time new information becomes available.

Multi-Residue Pesticide Methodology – Thiex reported that she had contacted Randy Lowell, FDA and that he uses PAM methodology and told her to call EPA. Position is open for a project leader to author a “Needs Statement” and drive this project.

ACTION ITEM:
• Identify Leader for project.

New Methods Need Survey – Thiex reviewed a spreadsheet with consolidated information obtained from the last survey.

ACTION ITEM:
• The committee will seek additional project leaders for additional priority methods and potential stakeholders.

Prohibited Material Methodologies – Dragan Momcilovic reported that the research into methods for the presence of prohibited materials (mainly ruminant proteins) is still on-going. FDA is using RealTime PCR together with Microscopy in their enforcement efforts. EU has formed a research group in Geel, Belgium to develop more definite methods. There is an issue with fish meal - early PCR results were poor while microscopy worked but demonstrated poor detection limits.

ACTION ITEM:
• None

CEN and ISO Activities – Harold Campbell had submitted a summary of the activities at the meeting in Bankok, December 13-15, 2005. The ISO/CEN ad-hoc committee was considering using the AAFCO sample preparation guideline. A discussion followed whether AAFCO should ask for royalties since CEN would be selling the book likely for a profit. An amount of $25 per copy was suggested
ACTION ITEM:

- Thiex to ask AAFCO BOD for advice on this issue.

Training Videos— Mark Lee, California Department of Ag is working on this project. No news.
ACTION ITEM:

- None

Next Workshop— Mark Lee, California Department of Ag may be willing to arrange the workshop in 2007 or 2008. Texas may also be a candidate for a 2008 (or later) meeting.
ACTION ITEM:

- None

Web sites— Thiex reported that both http://www.nfalc.org and http://www.aoac.org/ag_materials/additives/main.htm are up and running. Membership should not be required for access. The old aglabs website will be moved to nfalc sometimes this year when the nfalc website is a little more complete. States should still wait to enter information into nfalc since this part of the web site is still under development.
ACTION ITEM:

- None

Uncertainty Calculations— Aaron Price provided updates of the spreadsheets for estimating method uncertainty distributed earlier by Ed Moore, CFIA. The new version included corrections of some errors found in the original version.
ACTION ITEM:

- None

AAFCO Strategic Planning— Thiex will complete the survey sent out by AAFCO on behalf of the committee including working on an AAFCO Lab Committee value statement.
ACTION ITEM:

- None

Ankom Bag Based Methods— Thiex reported that method comparability data were missing and questioned if methods using different solvents could ever be expected to be equivalent but that each method should be considered by itself.
ACTION ITEM:
• Thiex will follow up with ANKOM to see if they wish to submit follow up data on their fat method showing comparability data to existing AOAC fat methods.

AOAC ALACC
The AOAC INTERNATIONAL Analytical Laboratory Accreditation Criteria Committee (ALACC) is revising the *AOAC INTERNATIONAL Accreditation Criteria for Laboratories Performing Food Microbiological and Chemical Analyses in Foods, Feeds, and Pharmaceutical Testing*. They have asked for formal input of the AAFCO Laboratory Committee to carefully review the document to see if it's a good fit (little or no edits) for feed laboratories, a possible fit (lots of edits) or doesn't fit at all. Louise Ogden and Brenda Snodgrass will represent the AAFCO Laboratory Committee. Additional volunteers may be needed.

Respectfully submitted,

Lars Reimann

*Eurofins Scientific, Inc.*