NOAA TECHNICAL MEMORANDUM NMFS SEFC - 265

BIOMEDICAL TEST MATERIALS PROGRAM: STANDARD OPERATING PROCEDURES OF THE QUALITY ASSURANCE PROJECT

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September 1990

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1 INTRODUCTION

The Biomedical Test Materials Program was initiated in 1986 by a memorandum of understanding (MOU) between the National Oceanic and Atmospheric Administration (NOAA) and the National Institutes of Health (NIH)/ Alcohol, Drug Abuse, and Mental Health Administration (ADAMHA). The purpose of the program is to facilitate evaluation of the role of omega-3 fatty acids, prevalent in marine fishes, in health and disease. The role of the National Marine Fisheries Service in the program is to produce an analytically well defined and consistent supply of refined fish oil and omega-3 derivatives for use as test materials in biomedical and clinical research. The NIH/ADAMHA provides the review and approval mechanism for the distribution of test materials to researchers who are funded by NIH, or other peer reviewed organizations.

Production, chemical analysis, and distribution of the test materials are carried out at the Charleston Laboratory of the National Marine Fisheries Service. At Charleston, the Program is thus administratively and functionally divided into three Projects: Production, Quality Assurance/Quality Control (QA/QC), and Distribution Management. In addition, the Lipid Analytical Services (LAS) Project at Charleston Laboratory performs many of the lipid related analyses as a service to the QA/QC Project. Microbiological analyses are carried out by the Microbiology section at Charleston Laboratory as a service to the QA/QC Project.

Quality assurance (QA) entails the analysis of a completed product for a battery of tests in order to assure its safety and identity, based on established specifications. *Quality control* (QC) ancompasses all activities which verify the quality and identity of raw materials and of the test materials throughout their processing, packaging, and storage.

The responsibilities of the QA/QC Project are thus to (1) certify that the test materials produced by the Program meet specifications for product identity and safety as determined by 29 different analytical tests, (2) assist in the development of new production procedures and product delivery forms, using analytical results to determine product characteristics, (3) carry out storage stability studies on all product forms produced by the Program, (4) provide analytical service to researchers using the test materials as requested, and (5) develop new analytical procedures for the analysis of new product forms as necessary. Many of the standard methods for analysis of oils published by the American Oil Chemists' Society and the Association of Official Analytical Chemists were modified for analysis of fish oils in the first two years of the Program.

The purpose of this manual is to define the role of the QA/QC Project and to detail the administrative procedures used by the Project for quality assurance and quality control of test materials. A separate manual describing the analytical methods used by the Project has been published (Van Dolah and Galloway, 1988). Manuals describing the standard operating procedures used by the Production Project (Joseph, 1989) and Distribution Management (Fair, 1989) have also been published. A comprehensive description of the Biomedical Test Materials Program is presented by Galloway (1989) in the Drug Master Files submitted to Food and Drug Administration in support of the use of the test materials for human clinical trials.

2 SAMPLE NUMBERING SYSTEM

2.1 Batches and Lots

Sample numbers are assigned by the Production Project according to the following system. The daily output from a given production step is assigned a "batch" number consisting of the last two digits of the year followed by three digit day of the calendar year, a code letter indicating the starting material, followed by a code letter indicating the current stage of production of the material. The letter codes for various stages of production are presented in Table 2-1. For example, *88109BF* is a batch of n-3 ethyl ester concentrate, produced from vacuum deodorized menhaden oil, on day 109 (April 18) of 1988.

Table 2-1. Letter codes for the numbering system according to production stage.

Production Stage	Letter Code
Steam Deodorized Menhaden Oil (SDMO)	A
Vacuum Deodorized Menhaden Oil (VDMO)	B
Vacuum Deodorized Oil, with TBHQ only	T
Crude Ethyl Ester	C
Crude Concentrate of Ethyl Ester	D
Short-Path Still, First Stage Distillate	E
Ethyl ester, Second Stage Distillate	F
Ethyl ester, Second stage distillate, with TBHQ only	Q
CO_2 Fractionated Esters	G
HPLC Fractions (in ethanol)	H
HPLC Fractionated EPA (neat)	I.4
HPLC Fractionated DHA (neat)	I.5
Corn Oil	V
Olive Oil	W
Safflower Oil	Y
Fish flavoring added to placebo oils	.99

"Batches" are combined at a later date to make a "lot" of material. The size of a lot is dictated by the amount of material requested by an individual researcher or by storage container size constraints. A lot is given a new number, beginning with "L" to indicate "lot", followed by the year, the three-digit day of the year on which is was combined, the starting material and the current production stage. For example, *L88168BF* is a lot of n-3 ester concentrate combined on day 168 (June 16) of 1988, and produced from vacuum deodorized menhaden oil. *L88333WW* is a lot of olive oil combined on day 333 (November 28) of 1988. *L90160YF.99* is a safflower oil ester with fish flavoring.

2.2 Storage Study Samples

Storage stability studies are carried out on all product forms (see Section 5.5). A sample number is assigned to a storage study sample upon submission of the sample by a member of the QA Project. The sample number consists of the lot number followed by a decimal point, a one-digit treatment code, and a two digit code for the number of months the sample has been stored. For example, *L86339AA.012* is a sample of steam deodorized oil stored for 12 months. *L88216BB.312* is a sample of vacuum deodorized oil stored 12 months under a specific treatment code, 3. The treatment code is specific to a given storage study and is detailed in the protocol for that storage study.

2.3 Miscellaneous Samples

All other types of samples submitted are assigned a sequential Miscellaneous number by the individual submitting, using the following format: *Misc-#-##*, where the first number is a notebook number and the second set of numbers is a page number on which the sample is entered. For example, *Misc-3-65* is a sample entered on page 65 of the third "Miscellaneous" notebook. The notebooks are ordered sequentially, with the first beginning at the start of the program in 1986. When a sample is logged in, the date of log-in, the sample identity, what analysis are to be carried out, and any other pertinent information are indicated on that page.

3 RECORDS

3.1 Sample Request Inventory

QA or MISC Analysis Request Sheets When a sample is submitted for analysis, an analysis request form is filled out by the submitter. If the request is a QA or Misc request, a QA request form is used (Figure 3-1). The form is placed in the "QA Request" or "Misc Request" box in Rm 242. The request form remains in the box until all analyses are completed and the QA Report is produced, at which time it is transferred to a "Sample Request Sheets" notebook. These sheets serve as permanent records of sample requests. One notebook is used for each calendar year. The contents are organized according to date of submittal, with the earliest at the back of the notebook.

QC Request Sheets When a QC request is submitted, a blue QC Request sheet is used (Figure 3-2). The request form is placed in the "QC Request" box in Rm 242. The results of QC analyses are recorded directly onto the request sheet (as well as the analysis notebook) and the request sheet is returned directly to the requestor. The request form is also photocopied and stored in a notebook in Rm 242 for permanent record.

Request Inventory Board In addition to placing the request sheet in the appropriate box, the individual submitting the sample for analysis copies the information from the request sheet to a "request inventory" board on the wall outside of Rm. 242 (Figure 3-3). Request for a given analysis is indicated by a check in the appropriate column. As the analyses are performed, the analyst crosses the check. This enables an individual to note at a glance the stage of analysis for a given sample.

3.2 Notebooks

Analytical Notebooks Each analysis performed by QA is recorded in a notebook designed specifically for that assay. The formats of the notebooks are shown in Appendix 1. The notebooks are stored in Rm 242. Each page of the notebook is dated and initialed by the analyst at the time of analysis. Each page of the notebooks is initialed by the QA/QC Project Leader as an indication that the results and quality control checks have been verified prior to entering the results into the database (below).

Method Development Notebooks Separate notebooks are kept to document the development of methods by the QA/QC Project. These notebooks are held by the analyst working on the method. Because the requirements for notebook format vary with each task, a standard form is not used. Standardized information is included on each page, such as the purpose of a given experiment, procedures, data, sample number and comments. A detailed objective statement appears on the first page of each new method that is being developed, with the stated approach outlined.

3.3 QA/QC Database

All QA/QC data obtained on biomedical test materials produced at Charleston Laboratory are stored in a database established using an IBM PC compatible 286 computer and Lotus Symphony Database software Version 2.1 located at the Project Leader's desk. A back-up copy is made on a 1.2 MB floppy disk each time data is entered and is stored separately in the computer room in order to insure the long term safety of the QA records.

The Symphony database program is based on a spreadsheet format. The program allows the user to view the database in the "spreadsheet" mode or in a "form" mode. The entire spreadsheet range is called the "QA/QC Report" window and is divided up into numerous smaller "windows", each containing a database for a separate analytical test (Figure 3-4a, 3-4b). The form mode is used for data entry. Each analytical test thus has its own data entry form.

Each database consists of seven "ranges" or categories of information, including the name range, entry range, definition range, report range, criterion range, database range, and the output range (Figure 3-5). The name range is used only when designing the database. The Entry range defines the format of the entry form, and is thus not normally used once the database is established. The definition range defines the type (alpha or numeric) and length of allowable entries for each data point. The criterion range is utilized by the program during data retrieval. The database range stores all entered datapoints. The output range contains data retrieved during a search.

3.3.1 Accessing the Database

The QA/QC database is stored in the Northgate 286 computer at the Project Leader's desk. A password which allows access to the database is held by the QA/QC Project leader. To access the database, turn on the computer. A menu will appear. The password will be requested at this time. After entering the password, press F1 for Symphony. Press "1" for Symphony again. Press F9 for file retrieve. Press F9 for a vertical listing of files. Press "End" to switch to the \symph\QA\ subdirectory. Key down to highlight "QAREPORT.WR5". Press enter. The database will come up in "QA Report" window, in the SHEET mode.

3.3.2 Data Entry

Data is entered using the FORM mode of Symphony. All windows except the QA Report window are stored in FORM mode. In order to enter data, press F9, "Window", "Use", F9 (for a vertical listing). Select the desired window. (For example, to enter PV data, highlight PV and press Enter). The appropriate data entry form will appear (Figure 3-6). If the screen does not state "New Entry" at the top, press "End". This will bring up the last entry. Press "page down". This should bring up the "new entry" sheet.

Type the data into the entry sheet. Press "Insert" to insert the completed data form into the database.

To then enter data for a different assay, press F9, "Window", "Use", F9 (for a vertical listing). Select the desired window. (For example, to enter PV data, highlight PV and press Enter).

After entering all of the desired data, return to the QA Report Window (F9, "Window", "Use") and save the file by pressing F9, "File", "Save", "\symph\qa\QAREPORT.WR5". The screen will ask "Overwrite existing file?". Type "y" for yes. Save the file to both C:\symph\qa and to a floppy disk each time data is entered.

3.3.3 Report Generation

Data is retrieved from the database in the form of QA/QC Reports, generated using a Symphony MACRO "Macro QB", which is stored in the \Symph subdirectory (Appendix 2). The macro searches each database (e.g., PV, FFA etc.) for data relevant to a specific sample number entered by the user. To use the macro, select "QA Report" window by pressing F9, "Window", "Use" and selecting "QA Report". The QA Report window automatically comes up at position A1000. This is the report location (Figure 3-7). Column A contains the names of the analyses. Column B contains the data which is retrieved by the macro. Before beginning generation of a report, erase the contents of column B by pressing F10, "Erase", Pagedown, Pagedown, Pagedown, Enter.

In order to use the macro, press F9, "Application", "Attach", "Macromanager", "Load", "QB", "Quit". Move to location B999, and enter the sample description (i.e., "vacuum deodorized menhaden oil"). Move the cursor to B1000. Enter the desired sample number (Lot No. of Misc-#-##). Then press Alt and B simultaneously. The macro will generate the report in the B column.

To store the QA Report, press F9, "File", "Extract", key over and down to highlight the desired area to extract (i.e., the entire QA report). Press "Enter". All QA Reports are stored in the \Symph\QA subdirectory; therefore, when prompted for the name of the file to be extracted, the file should be named "QA\LxxxxxXX.wr1" (for a lot) or "QA\M-#-##.wr1" (for a misc sample). Editing and printing can then be done from a newly created file. All reports generated by the QA/QC Project are stored in this fashion.

To edit or print the report, the user must leave the database program. Therefore, it is convenient to generate all desired reports using the macro and save each to a new file name before leaving the database. Once all are generated and stored, they can then individually be edited and printed. To retrieve the desired file, press F9, "File", "Retrieve", F9, "End" (to go the \QA subirectory), then key down to the desired file. To print the file, press F9, "Print", "Align, "Go". Make sure the printer is on "compressed print" mode for the most attractive output.

Complete details of the use of Lotus Symphony databases are given in the manuals supplied with the software.

Figure 3-1. Analysis request form used to analyze QA or MISC samples.

QA/QC ANALYSIS REQUEST FORM

DATE: ____________ REQUESTER: ______________________________

BATCH No.: 88__ __ __ __ __ . __

SUB-LOT No.: S88 __ __ __ __ __

LOT No.: L88 __ __ __ __ __

MISCELLANEOUS ____ No.: __ __ - __ __

STORAGE STUDY No.: L8 __ __ __ __ __ __.__ __ __

COMMENTS: ____

ANALYSIS REQUESTED

____ Complete QA
____ Routine QC

____ TLC
____ TLC-FID
____ Fatty Acid Profile
____ Free Fatty Acids
____ Conjugated Fatty Acids
____ Trans Fatty Acids
____ Iodine Value
____ Cholesterol
____ Peroxide Value
____ Anisidine value
____ Polar Oxidation Products
____ Volatile Oxidation Products
____ PCBs - Pesticides
____ Residual solvents
____ Urea
____ TBHQ
____ Tocopherols
Metals
____ a. As,Cd,Hg,Pb,Se
____ b. Ca,Cr,Cu,Fe,K,Na,Ni,Sn,Zn
____ Moisture
Sensory Attributes
____ a. Odor profile
____ b. Flavor profile
____ c. Color
____ Bacteria

Figure 3-2. Analysis request form used to analyze QC samples.

** IN HOUSE QC REQUEST FORM **

DATE: _____________ REQUESTER: ___________________________________

BATCH No.: 90__ __ __ __ __ . __

SUB-LOT No.: S90 __ __ __ __ __

MISCELLANEOUS ____ No.: __ __ - __ __

SAMPLE DESCRIPTION OR INFORMATION NEEDED: ______________________________
____
____

ANALYSIS REQUESTED		DATE COMPLETED	RESULTS and COMMENTS
____	TLC	___________	______________________
____	Fatty Acid Profile	___________	______________________
____	Free Fatty Acids	___________	______________________
____	Peroxide Value	___________	______________________
____	Trans Fatty Acids	___________	______________________
____	Iodine Value	___________	______________________
____	Cholesterol	___________	______________________
____	Anisidine value	___________	______________________
____	Polar Oxidation Products	___________	______________________
____	PCBs - Pesticides	___________	______________________
____	Residual solvents	___________	______________________
____	Urea	___________	______________________
____	TBHQ	___________	______________________
____	Tocopherols	___________	______________________
	Metals		
____	a. As,Cd,Hg,Pb,Se	___________	______________________
____	b. Ca,Cr,Cu,Fe,K,Na,Ni,Sn,Zn	___________	______________________
____	Moisture	___________	______________________
	Sensory Attributes		
____	a. Odor profile	___________	______________________
____	b. Flavor profile	___________	______________________
____	c. Color	___________	______________________
____	Bacteria	___________	______________________

* These data are for in house use only. Results may not have been reviewed or verified by the QA Project Leader.

Figure 3-3. Example use of the sample inventory board to track progress of samples in the process of being analyzed.

SAMPLE	DATE SUBMITTED	LIPID CLASS	FATTY ACID PROFILE	PV	pAV	FFA	MOISTURE	UREA	PCB/Pest	TOCOPHEROLS	TBHQ	CHOLESTEROL	METALS	TRANS FA	IV
L89304BF	12/5/89	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓
L89258BF	12/1/89	✗	✗	✗	✗	✗	✗	✗	✗	✗	✗	✗	✗	✗	✗
L88333BB.012	12/30/89		✓	✓		✓				✓					
Misc-4-12	12/15/89		✓	✓						✓	✓				
89302BF	12/3/89		✓	✓											

Figure 3-4a. Location of databases for individual assays, within the Symphony spreadsheet.

WINDOW	WINDOW NAME
A1...IV8192	QA/QC REPORT
A1...N350	KF (Karl Fischer)
L1...V350	PV (Peroxide Value)
W1...AK350	UA (Urea Analysis)
AI1...AW350	Conjugated FA
AX1...BK350	ATX (Antioxidants)
BL1...BW350	AV (p-Anisidine)
BX1...CJ350	IV (Iodine Value)
CK1...CW350	FAP (Fatty Acid Profile)
CX1...DH350	CHOL (Cholesterol)
DI1...EC350	(MET_TR) (Metals-Trace)
ED1...FA350	SENSOR_OD (Odor)
FB1...FT350	COP (Conjugable Oxidation Products)
FV1...GR350	PCB (PCBs/pesticides)
GT1...HP350	SENSOR_FL (Flavor)
HQ1...IA350	FREE_FA (Free Fatty Acids)
IB1...IV350	MET_MACRO (Macro Elements)
AA351...AM700	BACT (Bacteria)
AN351...AX700	TRANS (Trans Fatty Acids)

b. The QA/QC REPORT window encompasses the entire spreadsheet, with the databases for individual assays being subsections within it.

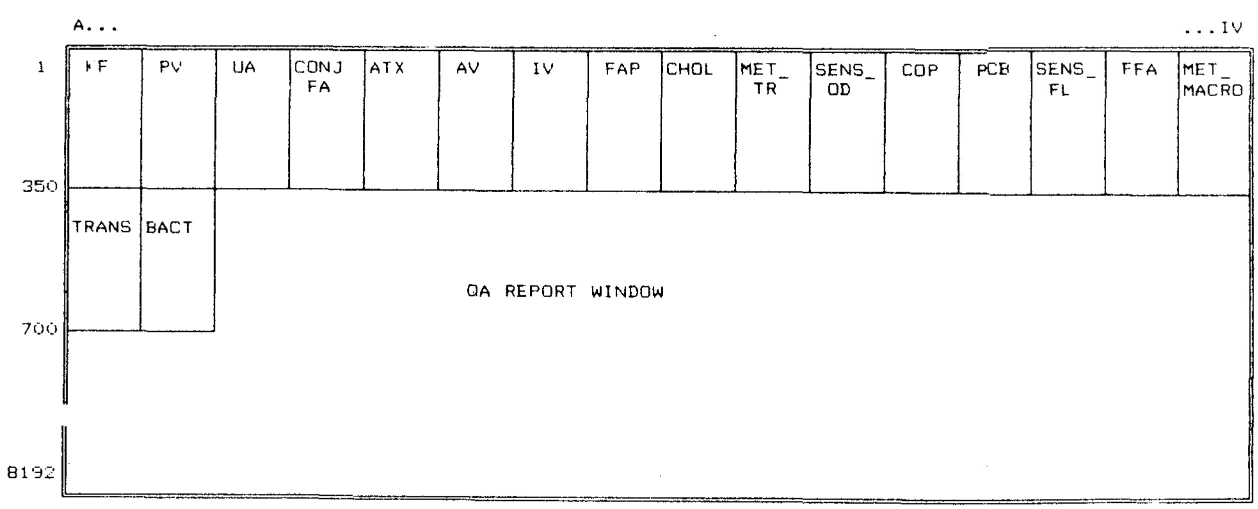


Figure 3-5. Each database is made up of six component ranges which can be viewed in the "sheet" mode.

OUTPUT RANGE

```
ANALYSIS DATE 2 SAMPLE No.2   SAMPLE Wt, g 2 PEROXIDE VALUE COMMENTS 2
```

ENTRY RANGE

```
ANALYSIS DATE ________
SAMPLE No. __________
SAMPLE Wt, g ____
PEROXIDE VALUE _____
COMMENTS: ______________________________________________________________
________________________________________________________________________
________________________________________________________________________
________________________________________________________________________
________________________________________________________________________
________________________________________________________________________
ANALYST INITIALS: ____________________
```

DEFINITION RANGE

Name	Value	Type	Default Formula
ANALYSIS DATE 2	4/13/90	L:17	
SAMPLE No.2	L90103BF	L:20	
SAMPLE Wt, g 2	5	N:18	5
PEROXIDE VALUE	6.16	N:16	nd
COMMENTS 2		L:67	
A2		L:74	
B2		L:74	
C2		L:74	
D2		L:74	
E2		L:74	
ANALYST INITIALS	YT	L:20	YT

REPORT RANGE

ANALYSIS DATE 2	SAMPLE No.2	SAMPLE Wt, g 2	PEROXIDE VALUE	COMMENTS 2
10/28/86	86301BBI.100	5.02	1.39	PB1-11

CRITERION RANGE

ANALYSIS DATE 2	SAMPLE No.2	SAMPLE Wt, g 2	PEROXIDE VALUE	COMMENTS 2
	L90060BB			

DATABASE RANGE

ANALYSIS DATE 2	SAMPLE No.2	SAMPLE Wt, g 2	PEROXIDE VALUE	COMMENTS 2
10/28/86	86301BBI.100	5.02	1.39	PB1-11
10/29/86	86301BCI.100	5.03	0.42	PB1-13
10/30/86	86301BEI.100	5.02	2.98	PB1-15
11/03/86	86301BEI.200	5.02	2.73	PB1-18
11/13/86	86301BEI.300	5.04	11.19	PB1-28
12/19/86	86301BEI.400	5.02	7.37	PB1-71
11/03/86	86307BBI.100	5.03	1.79	PB1-18
11/13/86	86307BEI.100	4.95	6.08	PB1-28
11/13/86	86307BEI.200	5	11.68	PB1-28
11/13/86	86307BEI.300	4.98	8.28	PB1-28

Figure 3-6. An example entry form. When working in the FORM environment of Symphony, only one entry is visible at a time. When working in the SHEET environment, this data would appear in the "database range" as in Figure 3-2.

```
Editing Record 782 of 783                                          FORM
Enter ANALYSIS DATE

 ANALYSIS DATE 4/13/90_
 SAMPLE No. L90103BF____
 SAMPLE Wt, g 5___
 PEROXIDE VALUE 6.16_
 COMMENTS: ____________________________________________________________
 ______________________________________________________________________
 ______________________________________________________________________
 ______________________________________________________________________
 ______________________________________________________________________
 ______________________________________________________________________
 ANALYST INITIALS: YT__________________

PEROXIDE VALUE---------------------------------------------PEROXIDE VALUE
14-May-90  03:57 PM                                   Calc      Num
```

Figure 3-7. QA Report generation area of the QA Report Window. When the QA Report window is brought up, it automatically goes to position A1000. The sample identity in B999 and sample number in B1000 are from the last generated report and must be overwritten to generate a new report. The date in B1001 will be recalculated automatically by the Symphony macro "macro QB".

```
B998:                                                                SHEET
--------------------A-----------------B-----------C------------D----------
992
993 The information contained in this QA/QC REPORT is believed to be accurate
994 and is offered in good faith for the benefit of the investigator.  NMFS,
995 however, cannot assume any liability or risk involved in the use of this
996 test material since the conditions of use are beyond our control.
997
998
999  IDENTIFICATION                    VDMO
1000 BATCH/LOT No.                 L90060BB
1001 REPORT DATE                   05/14/90
1002
1003
1004 EPA, mg/g                        138.0
1005 DHA, mg/g                        102.4
1006 TOTAL n-3, mg/g                  323.9
1007 FREE FATTY ACIDS, %               0.05
1008 TRANS FATTY ACIDS, %                <5
1009 CHOLESTEROL, mg/g                 1.94
1010 PEROXIDE VALUE, meq/kg            0.83
1011 IODINE VALUE                     187.4
-------------------------------------------------------------QA_REPORT
14-May-90  04:08 PM                                   Calc     CapsNum
```

4 QA PROCESS

Quality Assurance (QA) for the Biomedical Test Material Program entails analysis of the complete battery of analytical assays (Figure 4-1) carried out on a "lot" of Charleston Laboratory biomedical test material to assure its safety and identity, and the incorporation of that data into a QA Report. The signature of the QA/QC Project Leader on the report signifies that the material meets specifications for safety and identity. The signature of the QA/QC Project Leader is required for release of the material by Distribution Management. The time allotted from time of sample submission to production of a complete QA report is three weeks.

4.1 Sample Log-in

The QA process begins with the assignment of a lot number by the Production Project (see section 2) for bulk products or by the Distribution Project for encapsulated products. A 100 ml aliquot of test material in a 125 ml Nalgene polyethylene bottle plus a 15 ml aliquot in a glass culture tube with a teflon-lined cap are submitted for QA/QC analysis. An analysis request form (Figure 3-2) is filled out indicating "Complete QA" as the desired analysis. This form is placed in a QA request form box in the Production facility. The samples are labeled with the lot number and placed in a QA sample submittal box in the walk-in cooler in the Production facility.

Each morning a designated member of the QA/QC staff checks for samples and picks them up, along with their accompanying request forms. The request form is placed in the QA request box in Rm. 242. Samples are placed in the refrigerator in Rm. 242. The same individual then copies the lot number, date, and analyses requested to a sample log-in board in the hall outside Rm. 242.

Each QA staff member is assigned primary responsibility of a different array of assays. For most analyses, staff members have been cross-trained to provide coverage of those analyses when the primary analyst is absent. Each individual is responsible for checking the log-in board daily to note any new sample submissions.

4.2 Sample Handling

All samples are stored in the QA/QC refrigerator in Rm. 242 until analyzed. Before opening the bottle, samples are brought to room temperature to avoid absorption of moisture by the cool sample. This can be done by setting the sample out at room temperature for 1 hour or placing the sample bottle in luke warm water for 15-30 min with occasional swirling of the contents to ensure even warming. Contents of the sample bottle must be thoroughly mixed before analysis.

After opening the bottle for sampling the bottle *must* be layered with nitrogen to displace oxygen from the headspace above the sample, since these products are highly susceptible to oxidative degradation, then replaced in the refrigerator for storage.

Peroxide value is generally the first analysis to be performed. In ester samples, the PV must be determined within 5 days of submission in order to ensure that the values represent the sample as it was when submitted. Storage of the sample for longer than that period of time at refrigerator temperatures has resulted in artificially high PVs, relative to the product stored at -40°C. Other analyses do not have a specific time limit other than a standard three-week turn around time for complete QA analysis.

A separate sample aliquot is submitted for PCB and pesticide analysis, in a 15 ml glass culture tube with a teflon-lined cap. The separate treatment of this sample is necessary in order to avoid contamination of the sample with plasticizers from the polyethylene bottles. These samples are stored in the QC refrigerator until analyzed (Rm. 242).

4.3 Sample Analysis

The analytical procedures used by the QA/QC Program (including those used by LAS and Microbiology) are published in the QA methods manual "Biomedical Test Materials Program: Analytical Methods for the Quality Assurance of Fish Oil" (Van Dolah and Galloway, 1988). All analytical procedures include quality control procedures to verify the analyses, which are detailed in that manual.

Data obtained by QA staff is reviewed by the QA Project Leader to ensure analyses meet the established quality control checks. Approval is indicated by the QA Project Leader's initials on each page in the analytical notebooks (see section 3). Validation of data obtained by LAS and Microbiology is the responsibility of their respective project leaders. Once checked, data from each analysis is entered into the QA/QC database by the QA/QC Project Leader and a QA report is generated in the database (see section 3).

4.4 Quality Specifications

Quality specifications (Figures 4-2 and 4-3) for test materials produced at Charleston Laboratory were established as a collaborative decision of BTM and LAS project leaders and the Program Leader. Precedence for some of the specifications established by this program are found in the World Health Organization/Food and Agriculture Organization (WHO/FAO) specifications for edible oils (WHO, 1970) and a WHO/FAO "Compilation of Legal Limits for Hazardous Substances in Fish and Fishery Products" (Nauen, 1983). Other specifications were established for the Charleston Laboratory test materials based on clinical or biomedical relevance (e.g., cholesterol, trans fatty acids) or legal limits imposed by the FDA (e.g., for TBHQ concentration). Standards of identity of fish and placebo oils were derived from a survey of published fatty acid literature.

All lots of material shipped from Charleston Laboratory must meet the established specifications. The QA/QC Project Leader is responsible for determining that specifications are met. Approval of the material for shipment from Charleston Laboratory is indicated by the signature of the QA/QC Project Leader at the bottom of the QA Report. All shipments of material from Charleston Laboratory must be accompanied by a signed QA Report.

If the material tested does not meet specifications, the material must be reprocessed by the Production Project in order to meet specifications. The reprocessed sample is then resubmitted for complete QA analysis. If a test material fails to meet specifications after reprocessing, it is then disposed of as fish oil waste.

TABLE 4-1. Analytical determinations carried out for quality assurance of Charleston Laboratory test materials.

I. Lipid Characterization
  a. Lipid Class Profile
  b. Fatty Acid Composition
  c. Free Fatty Acids
  d. Isolated Trans Fatty Acids
  e. Unsaturated Fatty Acids (Iodine Value)

II. Cholesterol

III. Fatty Acid Oxidation Products
  a. Peroxides (PV)
  b. Aldehydes (pAV)
  c. Polar Oxidation Products

IV. Moisture

V. Organics
  a. PCB's and pesticides
  b. Residual urea
  c. Antioxidants
    i. Tert-butyl hydroquinone (TBHQ)
    ii. Tocopherols

VI. Metals
  a. Arsenic
  b. Cadmium
  c. Lead
  d. Mercury
  e. Selenium

VI. Sensory Attributes
  a. Odor
  b. Flavor
  c. Color - Hellige number

VII. Bacteria (encapsulated products only)
  a. Coliforms
  b. Salmonella

Figure 4-2. Quality specifications for triglyceride test materials.

ANALYSIS TYPE	TRIGLYCERIDE TEST MATERIAL VDMO	CORN	OLIVE	SAFFLOWER
TRIGLYCERIDES, %	>92	>95	>95	>95
EPA, mg/g	>120	na	na	na
DHA, mg/g	>75	na	na	na
16:0, %	na	8-12	9-17	6-7
18:1n-9, %	na	19-49	50-84	9-14
18:2n-6, %	na	34-62	4-18	76-81
FREE FATTY ACIDS, %	<0.2	<0.2	<0.2	<0.2
TRANS ACIDS, %	<5	<5	<5	<5
CHOLESTEROL, mg/g	<5.0	0	0	0
PEROXIDE VALUE, meq/kg	<5.0	<10.0	<10.0	<10.0
IODINE VALUE, g I_2/100g	>160	102-130	79-88	135-150
ANISIDINE VALUE	<50	<20	<20	<20
a-TOCOPHEROL, mg/g	0.5-5.0	0.1-1.0	0.1-1.0	0.1-1.0
g-TOCOPHEROL, mg/g	0.5-5.0	0.05-0.5	0.05-0.5	0.05-0.5
TBHQ, %	0.01-0.02	0.01-0.02	0.01-0.02	0.01-0.02
MOISTURE, ug/g	<500	<500	<500	<500
PCBs, ug/g	<0.5	<0.5	<0.5	<0.5
TOTAL DDT, ug/g	<0.5	<0.5	<0.5	<0.5
TRACE METALS, ug/g:				
Arsenic	<1.0	<1.0	<1.0	<1.0
Cadmium	<1.0	<1.0	<1.0	<1.0
Lead	<1.0	<1.0	<1.0	<1.0
Mercury	<1.0	<1.0	<1.0	<1.0
Selenium	<1.0	<1.0	<1.0	<1.0
SENSORY ATTRIBUTES:				
ODOR (TIO)	<6.0	<4.0	<4.0	<4.0
FLAVOR (TIO)	<6.0	<4.0	<4.0	<4.0
OTHER:				
SPECIFIC GRAVITY	0.93	.914-.921	.910-.915	.919-.924
SOLIDIFICATION RANGE	**	-10 to -6^{o}c	-8 to -3^{o}c	-18 to -16^{o}c
SAPONIFICATION VALUE	191-200	187-193	190-195	186-194
UNSAPONIFIABLE MATTER, %	<1.3	<1.5	<1.5	<1.5

Figure 4-3. Quality specifications for ethyl ester test materials. A. fish oil derived products, B. ethyl esters of placebo oils.

A. ANALYSIS TYPE	FISH OIL ESTER TEST MATERIAL n-3 CONC	EPA	DHA
ESTERS, %	>90	>98	>95
EPA, mg/g	>400	950	<50
DHA, mg/g	>200	<30	900
FREE FATTY ACIDS, %	<0.2	<0.1	<0.1
TRANS ACIDS, %	<5	<5	<5
CHOLESTEROL, mg/g	<5.0	<0.1	<0.1
PEROXIDE VALUE, meq/kg	<10.0	<5.0	<5.0
IODINE VALUE, g I_2/100g	>320	*	*
ANISIDINE VALUE	<80	*	*
a-TOCOPHEROL, mg/g	0.5-5.0	**	**
g-TOCOPHEROL, mg/g	0.5-5.0	**	**
TBHQ, %	0.01-0.02	0.01-0.02	0.01-0.02
MOISTURE, ug/g	<500	<500	<500
PCBs, ug/g	<0.5	<0.5	<0.5
TOTAL DDT, ug/g	<0.5	<0.5	<0.5
TRACE METALS, ug/g:			
Arsenic	<1.0	<1.0	<1.0
Cadmium	<1.0	<1.0	<1.0
Lead	<1.0	<1.0	<1.0
Mercury	<1.0	<1.0	<1.0
Selenium	<1.0	<1.0	<1.0
SENSORY ATTRIBUTES:			
ODOR (TIO)	<6.0	*	*
FLAVOR (TIO)	<6.0	*	*

B. ANALYSIS TYPE	PLACEBO ESTER TEST MATERIAL CORN	OLIVE	SAFFLOWER
ESTERS, %	>85	>85	>85
EPA, mg/g	<0.05	<0.05	<0.05
DHA, mg/g	<0.05	<0.05	<0.05
16:0, %	8-12	9-17	6-7
18:1n-9, %	19-49	50-84	9-14
18:2n-6, %	34-62	4-18	76-81
FREE FATTY ACIDS, %	<0.2	<0.2	<0.2
TRANS ACIDS, %	<5	<5	<5
CHOLESTEROL, mg/g	<0.05	<0.05	<0.05
PEROXIDE VALUE, meq/kg	<10.0	<10.0	<10.0
IODINE VALUE, g I_2/100g	80-120	60-100	100-150
ANISIDINE VALUE	<20	<20	<20
a-TOCOPHEROL, mg/g	0.5-5.0	0.5-5.0	0.5-5.0
g-TOCOPHEROL, mg/g	0.5-5.0	0.5-5.0	0.5-5.0
TBHQ, %	0.01-0.02	0.01-0.02	0.01-0.02
MOISTURE, ug/g	<2000	<2000	<2000
PCBs, ug/g	<0.5	<0.5	<0.5
TOTAL DDT, ug/g	<0.5	<0.5	<0.5
TRACE METALS, ug/g:			
Arsenic	<1.0	<1.0	<1.0
Cadmium	<1.0	<1.0	<1.0
Lead	<1.0	<1.0	<1.0
Mercury	<1.0	<1.0	<1.0
Selenium	<1.0	<1.0	<1.0
SENSORY ATTRIBUTES:			
ODOR (TIO)	<7.0	<7.0	<7.0
FLAVOR (TIO)	<7.0	<7.0	<7.0

* does not apply
** not enough material to conduct these analyses routinely

5 QC PROCESS

Quality Control (QC) encompasses all activities which verify the quality and identity of raw materials and test materials throughout their processing, encapsulation, packaging, and storage. The majority of QC samples submitted are for the purpose of monitoring changes in quality of a product during a specific stage of processing, requiring immediate turn around of information to the requestor. For this reason, a separate QC request form is used (Figure 3-2), which provides space in which the analyst directly records the analytical results. Results from many of the analyses are available within a few hours of the request. Routinely, QC analyses are performed and results provided within two days.

5.1 Starting Materials

Quality specifications and standards of identity have been established for starting materials, including partially refined menhaden oil, olive oil, corn oil and safflower oil (Figure 5-1). Prior to purchase of a given lot of starting material, a sample is obtained from the vendor by the Production Project and is submitted to QA/QC for analysis of parameters listed in the specifications. Results of those analyses are provided to the Production Project Leader in the form of a QC report prior to purchase.

Upon receipt of a new shipment of raw material (fish or placebo oils), a sample is submitted to QA/QC and is analyzed for the same parameters and, in addition, the following trace metals, As, Cd, Cu, Fe, Hg, Pb, and Se. A QC Report listing results is provided to the Production Project Leader.

Identity of antioxidants (Tenox 20A, Tenox GT-1, Vitamin E-67 (Eastman Chemicals)) are verified upon purchase of a new lot of material for the content of active ingredients indicated on the label.

5.2 Production Process Development

The QA/QC Project is intimately involved in development of Production processes by providing analysis of samples submitted by the Production Group while testing new equipment for processing parameters. These samples are submitted as "miscellaneous" samples (see section 2).

5.3 Inspection of Production Process

Onsite inspection by Production Staff Production staff submit QC samples from various stages of Production as necessary to check the quality or consistency of product obtained.

Independent Inspection by QA Staff On a random schedule, critical stages of Production are spot-checked at least four times per year by a designated member of the QA/QC staff. These stages are: (1) vacuum-deodorized oil, (2) crude concentrate, (3) purified n-3 concentrate, and HPLC fractions of (4) EPA and (5) DHA. Analyses performed on random check samples are fatty acid profile, PV, cholesterol, and PCB/pesticides, and antioxidant content (TBHQ and a- and g-tocopherol).

5.4 QC of Encapsulated Test Materials

Prior to encapsulation of a test material, the material is analyzed for identity (fatty acid composition) and peroxide value to ensure the material is of good quality before encapsulation takes place and for antioxidants to allow balancing of antioxidant levels between test materials and placebos. After encapsulation, the material is assigned a new lot number, and is submitted for complete QA. Failure of the encapsulated lot to meet specifications for PV, fatty acid composition, or microbes are thus attributable to mishandling of the material by the encapsulating contractor, which must be dealt with according to the encapsulation contract.

5.5 Storage Stability Studies

All product forms produced by Charleston Laboratory are subjected to formal storage stability studies. The purpose of the studies is to determine the optimum storage conditions in order to recommend optimum conditions to users of the material. Results of the studies are available to researchers in the form of summary reports. Two summary reports have been published to date (Van Dolah *et al.*, 1988; Van Dolah *et al.*, 1989). The following studies have been completed or are currently in progress:

I. *Bulk BTMs*

Bulk fish oil with antioxidants(I) - L88012BB - at -40°C and 5°C. Quarterly sampling is being done for 12 months at the following periods: 1/12/88, 4/12/88, 7/12/88, 10/12/88, 1/12/89. The following analyses are performed quarterly: Peroxide value, Anisidine value, Free fatty acids, Tocopherols. Fatty acid composition at 0, 6, 12 months. Sensory analysis is done at time 0 and 12 months.

Bulk fish oil without antioxidants(I) - L88118BO - at 5°C. Quarterly sampling is being done for 12 months at 5/1/88, 8/1/88, 11/1/88, 2/1/89, 5/1/89. Samples are analyzed for PV quarterly. AV and fatty acid composition at 0, 6, and 12 months. Sensory analysis is done at time 0 and 12 months.

Bulk fish oil with antioxidants (II) - L88216BB - This study is testing three containers, thick walled polyethylene, thin walled polyethylene, and glass, at 5°C, and one container, thick walled polyethylene at -40°C. Quarterly sampling is done at 8/10/88, 11/10/88, 2/10/89, 5/10/89, 8/10/89, then 6-month sampling at 2/10/90, and 8/10/90. PV is analyzed quarterly. AV, tocopherols, fatty acid composition are analyzed at 0, 6, 12, and 24 months. Sensory analysis is done at time 0, 12, and 24 months.

Bulk fish oil without antioxidants (II) - L88218BO - This study is testing three containers, thick walled polyethylene, thin walled polyethylene, and glass, at 5°C, and one container, thick walled polyethylene at -40°C. Quarterly sampling is done at 8/10/88, 11/10/88, 2/10/89, 5/10/89, 8/10/89, then 6-month sampling at 2/10/90, and 8/10/90. PV, AV, tocopherols, fatty acid composition are analyzed quarterly. Sensory analysis is done at time 0, 12, and 24 months.

Bulk n-3 ethyl ester - L88168BF - at -40°C. Sampled quarterly on 9/30/88, 12/30/88, 3/30/89, 6/30/89, then every six months at 12/30/89, 6/30/90. PV, AV, tocopherol, and fatty acid composition are analyzed quarterly. Sensory analysis is conducted at 0, 12 and 24 months.

Bulk corn oil ester - L89152VF - at -40°C. Samples are analyzed quarterly for one year, then every six months until two years, at 6/89, 9/89, 12/89, 3/90, 6/90, 12/90, 6/91. PV, fatty acid profile, tocopherol are analyzed quarterly, sensory analysis every six months.

Bulk safflower oil esters - L89194YF - at -40°C. Samples are analyzed quarterly for one year, then every six months until two years, at 7/89, 10/89, 1/90, 4/90, 7/90, 1/91, 7/91. PV, fatty acid profile, tocopherol to be done quarterly, sensory analysis is performed every six months.

II. *Encapsulated BTMs*

Chase steam deodorized menhaden oil. - A86339A - 5°C. Capsules were analyzed monthly for 1 year (Van Dolah *et al*, 1989). Samples will continued to be analyzed quarterly until 24 months: 5/2/88, 8/2/88, 11/2/88, 2/2/89. PV, AV, FFA, Fatty acid composition, moisture, tocopherol analyzed quarterly. Sensory analysis is performed at 18 and 24 months. After 24 months, samples will be analyzed annually for the duration of the BTM Program.

GNP steam deodorized menhaden oil. - A87196A - 5°C. Capsules were analyzed at 6, 12, 18 months to confirm comparability with Chase capsule stability: 2/88, 8/88, 2/89. PV, AV, FFA, Fatty acid composition, moisture, tocopherol and sensory analysis were performed at all timepoints.

GNP vacuum-deodorized menhaden oil. - L88333BB - 5°C. Capsules were analyzed at 0 and 12 months to confirm comparability with SDMO. PV, Fatty acid composition, tocopherol and sensory analysis were performed at both timepoints, 12/88, 12/89.

GNP encapsulated n-3 esters. - L88333BF - 5°C. Capsules are analyzed quarterly for 12 months and every six months till 24 months for PV, AV, fatty acid composition, moisture, tocopherol. Sensory analysis is performed at 6-month intervals: 12/88, 3/89, 6/89, 9/89, 12/89, 6/90, 12/90.

GNP encapsulated corn oil esters - L88165VF - 5°C. Capsules are analyzed quarterly until one year, then at six month intervals until two years. PV, Fatty acid composition, tocopherol and sensory analysis are performed at all timepoints: 6/89, 9/89, 12/89, 3/90, 6/90, 12/90, 6/91.

III. *Other Product forms*

Microencapsulated materials. Storage stability studies were carried out on microencapsulated fish oil and n-3 esters from three different encapsulating companies to determine the feasibility of utilizing microencapsulated materials for animal diets. Monthly sampling was carried out for 6 months, then quarterly until 12 months. Results of the studies were summarized in a feasibility report (Fair *et al, in prep)*.

Omega-whip. Storage stability studies were carried out on formulated edible whips containing fish oil, corn oil, and n-3 ethyl ester in order to determine feasibility of this dosage form for use in clinical trials. The whips were packaged in aerosol cans and stored at room temperature. Sampling regime: quarterly sampling for one year, 1/89, 4/89, 7/89, 10/89, 1/90. Fatty acid composition, PV, sensory analysis are performed at each timepoint.

Aerosol studies. Storage stability studies on fish oil and n-3 ethyl esters packaged in aerosol cans were carried out to determine the feasibility of using this form for clinical trials. The cans were stored at room temperature and sampled quarterly for fatty acid profile, PV and sensory analysis.

5.6 Analytical Method Development

A major activity of the QA/QC Project has been development or modification of existing methods for the analysis of fish oils or esters. Many of the methods used by the program are modifications of standard methods published by the American Oil Chemists' Society (AOCS) or the Association of Official Analytical Chemists (AOAC). Most of the standards methods are designed for analysis of vegetable oils, and required some modification due either to (1) the different behavior of the long chained polyunsaturated fatty acids in the triglyceride fish oils or (2) the very different solubility characteristics of the esters versus the triglyceride oils. Methods developed as of December 1988 are published in the QA methods manual (Van Dolah and Galloway, 1988). Other methods are still being refined.

All methods used by the QA/QC program incorporate quality control checks of the procedure, in the form of either the analysis of known standards or recoveries calculated on spiked samples. These are detailed in the methods manual.

Figure 5-1. Specifications for starting materials purchased by the BTM Program.

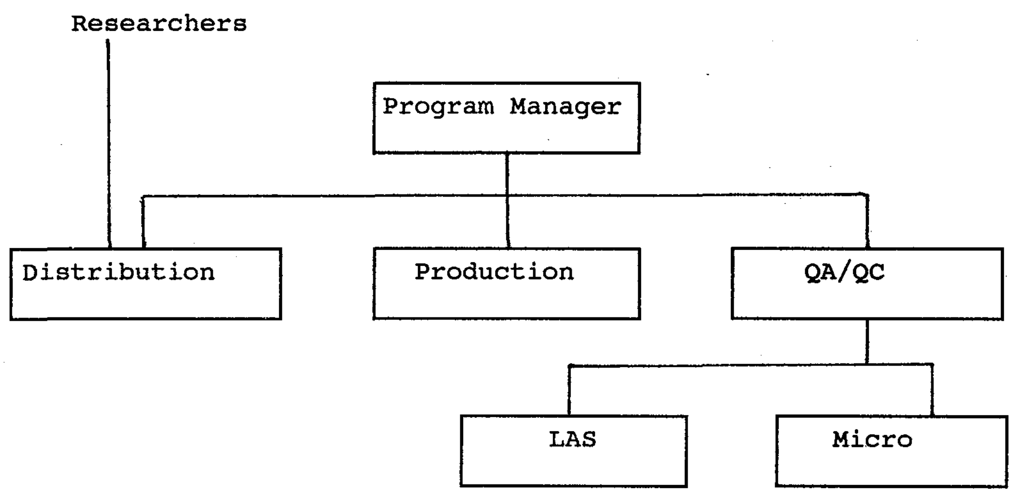
ANALYSIS TYPE	STARTING OILS MENHADEN	CORN	OLIVE	SAFFLOWER
TRIGLYCERIDES, %	>92	>95	>95	>95
EPA, mg/g	>120	<0.05	<0.05	<0.05
DHA, mg/g	>75	<0.05	<0.05	<0.05
16:0, mg/g	*	8-12	9-17	6-7
18:1n-9, mg/g	*	19-49	50-84	9-14
18:2n-6, mg/g	*	46-62	4-18	76-81
FREE FATTY ACIDS, %	<0.2	<0.2	<0.2	<0.2
CHOLESTEROL, mg/g	<5.0	0	0	0
PEROXIDE VALUE, meq/kg	<10.0	<10.0	<10.0	<10.0
IODINE VALUE, g I_2/100g	>160	102-130	79-88	135-150
ANISIDINE VALUE	<50	<20	<20	<20
MOISTURE, ug/g	<500	<500	<500	<500
PCBs, ug/g	<5.0	<0.5	<0.5	<0.5
TOTAL DDT, ug/g	<5.0	<0.5	<0.5	<0.5
TRACE METALS, ug/g:				
Arsenic	<1.0	<1.0	<1.0	<1.0
Cadmium	<1.0	<1.0	<1.0	<1.0
Lead	<1.0	<1.0	<1.0	<1.0
Mercury	<1.0	<1.0	<1.0	<1.0
Selenium	<1.0	<1.0	<1.0	<1.0
SENSORY ATTRIBUTES:				
ODOR (TIO)	<6.0	<4.0	<4.0	<4.0
FLAVOR (TIO)	<6.0	<4.0	<4.0	<4.0

* does not apply

6 COORDINATION WITH OTHER PROGRAM COMPONENTS

The objectives of the Program necessitate close coordination between the component projects. Formal updates from each project are provided at bi-weekly Program meetings with the administrative Program Manager. Written status reports on each project are provided monthly to the Program Manager and copied to all other project leaders. In accordance with (USP) Good Manufacturing Practices, formal written request forms are used to request the service of one project by another. This helps to establish a chain-of-custody of samples as well as to provide permanent records of requests and analyses performed. Figure 6-1 depicts the flow of requests and products between elements of the Program at Charleston Laboratory.

Figure 6-1. Flow of information between Charleston Laboratory components of the Biomedical Test Materials Program.



6.1 Coordination with Distribution Management

A request for the production of a given amount of a specific type of test material is initiated by the Distribution Management Project, as a result of communication with researchers to determine quantities and types of materials needed. At the time a request is submitted to the Production Program, a copy of the request form (Figure 6-2) is copied to the QA Project Leader. This alerts the QA/QC Project staff of dates on which to expect QA sample submission.

Upon completion of QA analysis, a QA Report (Figure 6-3) is written; if the product meets specifications, the report is signed by the QA Project Leader and copied to Distribution Management. The signed report constitutes release of the material, which allows Distribution Management to ship it to researchers. A copy of the signed QA Report accompanies each shipment of test material received by an NIH approved researcher, as well as a copy of the QA/QC Analytical Methods Manual and other technical information.

6.2 Coordination with Production

For QA analysis of a "lot" of test material, a member of the Production team fills out an analysis request form (Figure 3-1) indicating a "complete QA" analysis is requested. This is placed in the QA/QC request box in the Production Facility. The box is checked each morning by a designated member of the QA/QC staff and any submitted samples are collected. When a QA analysis is completed, the QA Report on that "lot" is copied to the Production Project Leader.

In order to request QC analysis of a sample, a member of the Production Team fills out an analysis request form, indicating the specific analyses required, and places it in the analysis request box in the Production facility. The requests and the samples are collected each morning by a QA/QC staff member. When the QC analysis is completed, results are provided verbally by the QA/QC analyst, since results are frequently needed immediately. A QC report is written by the QA/QC Project Leader to formally record the results and is distributed to the individual requesting the analysis and to the Production Project Leader.

6.3 Coordination with Lipid Analytical Services

The QA/QC Project works closely with the LAS Project for the production of QA/QC reports, since several of the routine assays are conducted by the LAS group. Samples submitted for QA/QC are automatically analyzed by the LAS Project for those specific assays assigned to that group (see section 5). Upon completion of their analysis, data is verified by the LAS Project Leader and a standardized LAS report (Figure 6-4) is submitted to the the QA/QC Project Leader for incorporation into the QA/QC database and QA/QC reports.

6.4 Coordination with Microbiology

All test materials which are gelatin encapsulated are analyzed for microbial contamination as part of the routine QA analysis. This is carried out by the Microbiology Section as an ad hoc service to QA/QC. Since this is an intermittent requirement, advance warning is provided to the head of the Microbiology Section two-three weeks prior to the encapsulation process to allow scheduling of the analyses. Sampling and analysis of the "lot" of materials is carried out by the Microbiology Section. After completion of the analyses, the results are verified by the head of the Microbiology Section, who submits a standardized written report to the QA/QC Project Leader (Figure 6-5).

Figure 6-2. Example production request form.

PRODUCTION REQUEST FORM

REQUEST DATE: 8-23-90
MATERIAL REQUIRED: VDFO
COMPLETION DATE: ______
LOCATION: ______
COMMENTS: ______

NEED DATE: 9-30-90
AMOUNT REQUIRED: 194 Kg
LOT NUMBER: ______

Figure 6-3. Example QA Report.

QA/QC REPORT

The information containedin this QA/QC REPORT is believed to be accurate and is offered in good faith for the benefit of the investigator. NMFS, however, cannot assume any liability or risk involved in the use of this test material since the conditions of use are beyond our control.

IDENTIFICATION	FISH OIL ETHYL ESTERS - TBHQ ONLY
BATCH/LOT No.	L90148BQ
REPORT DATE	07/13/90

EPA, mg/g	382.8
DHA, mg/g	282.7
TOTAL n-3, mg/g	781.7
FREE FATTY ACIDS, %	0.16
CHOLESTEROL, mg/g	2.74
PEROXIDE VALUE, meq/kg	3.27
IODINE VALUE	365
ANISIDINE VALUE	13.8
ANTIOXIDANT CONTENT:	
a-TOCOPHEROL, mg/g	0.09
g-TOCOPHEROL, mg/g	0.01
TBHQ, %	0.017
MOISTURE, ug/g	774
PCB, ug/g	0.06
TOTAL DDT, ug/g	0.31
SENSORY ATTRIBUTES,	
0-15, 15 MAX INTENSITY:	
ODOR:	
TIF	3.88
BUTTERY	0.23
BEANY	0.11
RANCID	0
PAINTY	0.39
OXIDIZED	0.08
GRASSY	0
FISHY	0
BITTER	0.24
SWEET	0.19
FRUITY/PERFUMY	1.07
BURNT	0
SOLVENT	0.39
LIQUOR	0.61
SOAPY	0.57
RAW GREEN	1.13
SPICE	0.14
CARDBOARD	0.57
BITTER	0.24
FLAVOR:	
TIF	3.77
BUTTERY	0.08
BEANY	0.11
RANCID	0
PAINTY	0.19
OXIDIZED	0
GRASSY	0
FISHY	0
BITTER	0.13
SWEET	0.19
FRUITY/PERFUMY	0.54
BURNT	0
SOAPY	0.93
SOLVENT	0.23
LIQUOR	0.43
CARDBOARD	0.61
RAW GREEN	1.13
COLOR (HELLIGE No.)	9
BACTERIA:	
E. COLI	neg
SALMONELLA	neg

QA/QC SUPERVISOR'S SIGNATURE [illegible]

DATE 7/18/90

* - not determined

Figure 6-4. Typical LAS report to QA/QC for fatty acid composition data. Signature of the LAS Project Leader indicates the data are verified.

VACUUM DEODORIZED FISH OIL Lot# L90159BB

FATTY ACID*	MG/G OIL**	FATTY ACID*	MG/G OIL**
12:0	1.1	16:3n-4	16.3
13:0	0.3	16:3n-3	0.5
14:0	63.0	16:4n-3	0.0
15:0	4.6	16:4n-1	11.8
16:0	155.6	18:3n-6	0.0
17:0	6.7	18:3n-4	3.2
18:0	27.0	18:3n-3	11.7
19:0	0.0	18:4n-3	33.3
20:0	1.8	18:4n-1	3.2
22:0	1.1	20:3n-6	1.5
24:0	0.0	20:4n-6	5.4
TOTAL SATS.	259.8	20:3n-3	1.4
		20:4n-3	13.3
14:1n-7	0.9	20:5n-3	127.7
14:1n-5	0.5	21:5n-3	5.9
16:1n-11?	3.6	22:4n-6	1.0
16:1n-9	1.6	22:5n-6	2.6
16:1n-7	83.4	22:5n-3	19.2
16:1n-5	3.4	22:6n-3	108.7
17:1	0.0		
18:1n-11	0.0	TMTD	0.0
18:1n-9	79.6	PRISTANATE	0.0
18:1n-7	26.5	14:0,ISO	0.0
18:1n-5/2n-9	2.4	14:0,AISO	0.0
19:1	0.0	15:0,ISO	2.4
20:1n-11+13	0.9	15:0,AISO	0.0
20:1n-9	10.7	17:0,ISO	2.2
20:1n-7	1.6	17:0,AISO	0.0
20:1n-5/NMID?	1.8	PHYTANATE/17:1?	2.8
22:1n-11+13	0.0	7MH	0.9
22:1n-9	1.6	7M7H	0.0
22:1n-7	0.4		
22:1n-5	0.0		
24:1	3.6		
TOTAL MONOENES	222.7		
16:2n-7	2.2		
16:2n-6	0.0	TOTAL PUFA	400.1
16:2n-4	11.7	TOTAL n-3	321.7
18:2n-9/1n-5	0.3	TOTAL n-6	24.7
18:2n-7	0.0	n-3/n-6	13.0
18:2n-6	12.5		
18:2n-4/3n-6	5.1		
20:2n-9	0.0		
20:2n-6	1.7		
TOTAL DIENES	33.4		

* Fatty acids were tentatively identified by comparison of their relative retention times with those of primary and secondary standards.
** 0.0 = <0.05 MG/G

8/1/90

Figure 6-5. Microbiology report to QA/QC. Signature of the Microbiology supervisor indicates that the data are verified.

LABORATORY LOT REPORT

Product Description: Capsules

Product Label:
Master case: L90148WQ, 72 per case
Inner case: L90148WQ, 24 per case
Bottles: L90148WQ, Biomedical Test Material, 100 capsules (1000 mg), Keep Refrigerated, Caution: New Drug, Limited by Federal (USA) Law to Investigational Use.

Quantity/Pack Size: Quantity determined by inventory list: 22 cases packed 72 bottles per case, 100 grams per bottle, and one partial case containing 10 bottles.

Storage Location: Refrigerated Environmental Room, Charleston Laboratory

Sampling Date: June 29, 1990

Number Samples Collected: 3 bottles

Date Bacteriological Analyses Initiated: 7-2-90 , Date Completed: 7-7-90

Bacteriological Results Including Isolate Identifications:

	XLD/ID	BGA/ID	MAC/ID
Sample 1	No Growth	No Growth	No Growth
Sample 2	No Growth	No Growth	No Growth
Sample 3	No Growth	No Growth	No Growth

Analyst: Laura F. Webster Reviewed by: Gary R. Richard

Date: 7/9/90 7/9/90

7 REFERENCES

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APPENDIX I. Forms used in QA/QC Project Notebooks

MOISTURE

Date ________

Analyst ___________________

Sample No. ______________

Titration No.	Sample Weight (g)	Moisture (ug/g)
Sample mean (x)		

Titrant Conc (meq/burette vol) _____________
Drift (ug/min) _____________

Instrument maintenance ____
____

Comments ____
____
____
____

ANISIDINE VALUE

Page _______

Date ________

Analyst ___________________

Sample	Weight (g)	A(s)	A(b)	pAV	pAV (mean)
Blank					

Comments ____

____

____

____

IODINE VALUE

Page _______

Date ________

Analyst __________________

Sample No. ______________

Titration No.	Sample Wt.(g)	IV (%)
Blank		
Mean (x)		

Titrant Conc (meq/burette vol) _____________
KI (date) _________

Instrument maintenance ____
____

Comments ____
____
____
____

PEROXIDE VALUE

Page _______

Date ________

Analyst ___________________

Sample No. ______________

Titration No.	Sample Wt. (g)	PV (meq/kg)
mean (x)		

Titrant Conc (meq/burette vol) _____________
KI (date) _________

Instrument maintenance ____
____

Comments ____
____
____
____

FREE FATTY ACIDS

Page _______

Date ________

Analyst ___________________

Sample No. ______________

Titration No.	Sample Wt.(g)	FREE FA (%)
Blank		
Oleic acid		
Sample mean ($\bar{x}$)		

Titrant Conc (meq/burette vol) _____________
KI (date) _________

Instrument maintenance ____
____

Comments ____
____
____
____

PCBs/PESTICIDES

Page ______

GC Date __________ Analyst ______________

Sample ID ________________________________

Column used for quantitation _______ Standard date __________

Compound	Run 1		Run 2		x Amount
	Pk Area	Amount	Pk Area	Amount	(ug/g)
A BHC					
HCB					
B BHC					
LINDANE					
HEPTACHLOR					
A CHLORDENE					
ALDRIN					
HEPT EPX					
G CHLORDANE					
O,P'DDE					
A CHLORDANE					
TRANSNONACLOR					
DIELDRIN					
P,P'DDE					
O,P'DDD					
ENDRIN					
P,P'DDD					
O,P'DDT 4					
P,P'DDT					
PCB					
SUM OF DDT's					

Comments ____

STD PK AREAS	HCB	P,P' DDE	P,P' DDT	PCB
STD 1				
SUM OF DDT's				

METALS

Element ______________

Page ______________
Analyst ______________
Date ______________

Digest Number ____

Lamp Energy ____________ Bkg Lamp Energy __________

Standard Curve (ng/ml): S1 _______ S2 _______ S3 _______
Absorbance (uncorrected): Blank _______ S1 _______ S2 _______ S3 _______

Characteristic Conc: __________________

Sample	Wt.	Vol	Dil	Abs(corr)	ug/L	ug/g	X	% Rec

Comments ____
____
____

APPENDIX II. Symphony Macro "QB"

```
\B              {GOTO}A1000~
                {MENU}C{ESCAPE}B1000~B42~
                {MENU}C{ESCAPE}B1000~M42~
                {MENU}C{ESCAPE}B1000~X47~
                {MENU}C{ESCAPE}B1000~AJ48~
                {MENU}C{ESCAPE}B1000~AY51~
                {MENU}C{ESCAPE}B1000~BM45~
                {MENU}C{ESCAPE}B1000~BY48~
                {MENU}C{ESCAPE}B1000~CL48~
                {MENU}C{ESCAPE}B1000~CY42~
                {MENU}C{ESCAPE}B1000~DJ68~
                {MENU}C{ESCAPE}B1000~EE71~
                {MENU}C{ESCAPE}B1000~FW70~
                {MENU}C{ESCAPE}B1000~GU71~
                {MENU}C{ESCAPE}B1000~HR42~
                {MENU}C{ESCAPE}B1000~AP389~
                {MENU}C{ESCAPE}B1000~AB398~
                {MENU}C{ESCAPE}B1000~IC63~
                {SERVICES}WUKARL FISCHER~{SWITCH}{MENU}QEQ{SUB_KF}
                {SERVICES}WUPEROXIDE VALUE~{SWITCH}{MENU}QEQ{SUB_PV}
                {SERVICES}WUUREA ANALYSIS~{SWITCH}{MENU}QEQ{SUB_UA}
                {SERVICES}WUPCB ANALYSIS~{SWITCH}{MENU}QEQ{SUB_PCB}
                {SERVICES}WUANTIOXIDANTS~{SWITCH}{MENU}QEQ{SUB_ATX}
                {SERVICES}WUANISIDINE~{SWITCH}{MENU}QEQ{SUB_AV}
                {SERVICES}WUIODINE VALUE~{SWITCH}{MENU}QEQ{SUB_IV}
                {SERVICES}WUFA PROFILE~{SWITCH}{MENU}QEQ{SUB_FAP}
                {SERVICES}WUCHOLESTEROL~{SWITCH}{MENU}QEQ{SUB_CHOL}
                {SERVICES}WUMETALS-TRACE~{SWITCH}{MENU}QEQ{SUB_MET_TR}
                {SERVICES}WUMACRO ELEM~{SWITCH}{MENU}QEQ{SUB_MET_MAC}
                {SERVICES}WUSENSOR_OD~{SWITCH}{MENU}QEQ{SUB_SENSOR1}
                {SERVICES}WUSENSOR_FL~{SWITCH}{MENU}QEQ{SUB_SENSOR2}
                {SERVICES}WUFREE_FA~{SWITCH}{MENU}QEQ{SUB_FFA}
                {SERVICES}WUTRANS_FA~{SWITCH}{MENU}QEQ{SUB_TRANS}
                {SERVICES}WUBACTERIA~{SWITCH}{MENU}QEQ{SUB_BACTER}
                {SERVICES}WUQA_REPORT~
                {CALC}

                {MENU}CIC50~B1036~
                {MENU}CIC51~B1037~


SUB_KF          {IF +B42=+B2}{MENU}CD2~B1018~
                {END}{PGDN}
                {RETURN}

SUB_PV          {IF +M42=+M2}{MENU}CO2~B1010~
                {END}{PGDN}
                {RETURN}

SUB_UREA        {IF +X47=+X2}{MENU}CAA2~B1011~
                {END}{PGDN}
                {RETURN}
```

```
SUB_PCB             {IF +FW70=+FW2}{MENU}CGJ2~B1020~{MENU}CGK2~B1021~
                    {END}{PGDN}
                    {RETURN}

SUB_ATX             {IF +AY51=+AY2}{MENU}CBC2~B1014~{MENU}CBD2~B1015~{MENU}CBB
                    {END}{PGDN}
                    {RETURN}

SUB_AV              {IF +BM45=+BM2}{MENU}CBP2~B1012~
                    {END}{PGDN}
                    {RETURN}

SUB_IV              {IF +BY48=+BY2}{MENU}CCC2~B1011~
                    {END}{PGDN}
                    {RETURN}

SUB_FAP             {IF +CL48=+CL2}{MENU}CCN2~B1004~{MENU}CCO2~B1005~{MENU}CCP
                    {SWITCH}{END}{PGDN}
                    {RETURN}

SUB_CHOL            {IF +CY42=+CY2}{MENU}CDA2~B1009~
                    {END}{PGDN}
                    {RETURN}

SUB_METALS_TR       {IF +DJ68=+DJ2}{MENU}CDQ2~B1023~{MENU}CDR2~B1024~{MENU}CDS
                    {END}{PGDN}
                    {RETURN}

SUB_SENSOR1         {IF +EE71=+EE2}
                    {IF +EE71=+EE2}
                    {END}{PGDN}
                    {RETURN}

SUB_SENSOR2         {IF +GU71=+GU2}{SWITCH}{MENU}CU{MENU}CI{RETURN}
                    {SWITCH}{END}{PGDN}
                    {RETURN}

SUB_METALS_MAC      {IF +IC63=+IC2}{MENU}CIE2~B1027~{MENU}CIF2~B1029~{MENU}CIG
                    {IF +IC63=+IC2}
                    {SWITCH}{END}{PGDN}
                    {RETURN}

SUB_BACTER          {IF +AB398=+AB352}{MENU}CAD352~B1008~
                    {END}{PGDN}
                    {RETURN}

SUB_TRANS           {IF +AP389=+AP352}{SWITCH}{MENU}CU{MENU}CI{RETURN}
                    {SWITCH}{END}{PGDN}
                    {RETURN}

SUB_FFA             {IF +HR42=+HR2}{MENU}CHT2~B1007~
                    {END}{PGDN}
                    {RETURN}
```