

North Carolina. The northerly distribution for these species may be indicative of larval transport patterns via currents, i.e., Gulf Stream (Williams 1974). Another significant element, as proposed by Norse (1977), is the presence of summer temperatures in excess of 20°C required for the hatching of eggs and larval development.

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ANALYSIS OF CHLORINATED HYDROCARBON POLLUTANTS: A SIMPLIFIED EXTRACTION AND CLEANUP PROCEDURE FOR FISHERY PRODUCTS

Fishery scientists wishing to quantitate chlorinated hydrocarbons find a multitude of methods only marginally appropriate to the routine analysis of marine fishery products. This paper is a laboratory manual, delineating details of a simple, rapid, and reliable method for the extraction and cleanup of samples of fish, fishery products, and paper for analysis of chlorinated hydrocarbons, such as PCB,¹ dieldrin, and DDT and its

¹Abbreviations used in this paper: DDE—*p, p'*-dichlorodiphenyldichloroethylene; DDMU—*p, p'*-dichlorodiphenylchloroethylene; DDT—*p, p'*-dichlorodiphenyltrichloroethane; IPA—*i*-isopropyl alcohol; PCB—polychlorinated biphenyls (Aroclor 1254 was used as the standard for PCB); and TDE—*p, p'*-dichlorodiphenyldichloroethane. The *o, p'*-isomers of DDT and

metabolites TDE and DDE. The procedures can be adapted to a great variety of sample types. The method is economical since small amounts of solvents are used and the equipment and glassware are relatively inexpensive and readily available.

Chlorinated hydrocarbon analysis in marine fishery products is an extremely complex procedure requiring extensive knowledge and many years of experience to perfect. A number of specialized problems, uncommon in the preparation of foodstuffs and freshwater fish for analyses of chlorinated hydrocarbons by the established methods, occur during the isolation of such materials from marine fish and fishery products. For example, the official method of the Association of Official Analytical Chemists (Horowitz 1970; Porter et al. 1970) often requires 1½ days for the initial extraction of marine fish oil because of intractable emulsions. After purification, the final extracts still contain substances which cause the rapid loss of sensitivity of the electron-capture detector and decomposition of the column packing in the gas-liquid chromatographic system. The procedure described in this paper eliminates extraction of fish oil and provides final extracts free of extraneous substances. In the course of analyzing over 2,000 samples we have found it suitable for routine analysis of marine fishery products.

The method was first developed by Robert Reinert (Reinert 1970; Snyder and Reinert 1971). We have refined it to maximize recovery of chlorinated hydrocarbons and have adapted it to new types of samples, such as fishmeal and carbonless carbon paper. We have described the procedure in detail because of the ultimate purity and freedom from unanticipated contaminants required of extracts for gas chromatography with an electron-capture detector. Data from samples of typical fishery products analyzed by us using the methods described in this paper were comparable to the data obtained by a number of other laboratories² following their usual procedures.

Preliminary Information

The accuracy and precision of analyses for chlorinated hydrocarbons are assured only if careful attention is given to the procedural details and the time factors involved in the various steps.

its metabolites act similarly to the more common *p, p'*-isomers used here.

²See Acknowledgments for the list of laboratories.

Time

For a set of eight samples: Extraction of oil, none; extraction of other samples, 4 h (or less); cleanup, 1 h; separation of DDT and PCB, 4 h; and transfer and dilution of samples for gas-liquid chromatography, 1-2 h.

Reagents

Analytical standards: U.S. Environmental Protection Agency, Health Effects Research Laboratory, Environmental Toxicology Division, Research Triangle Park, N.C.

Aroclor[®]1254: Monsanto Company, St. Louis, Mo. (Aroclor 1254 was used as the analytical standard for PCB because it represented the PCB most typically found in our samples. Snyder and Reinert (1971) tested both Aroclor 1254 and Aroclor 1260. The suitability of the procedure for PCB of other degrees of chlorination would need to be verified.)

Sodium sulfate: anhydrous, reagent grade. If the blank indicates the sodium sulfate is contaminated, rinse with petroleum ether or hexane and dry before use.

Solvents: distilled-in-glass grade, Burdick & Jackson, Muskegon, Mich. (The suitability of other "pesticide" or "nanograde" solvents would need to be determined.)

Florisil: 60/100 mesh P.R. grade, Floridin Company, Pittsburgh, Pa. Store in the dark in tightly closed, screw-cap, flint glass jars, and use without further activation.

Silica gel: grade 950, 60/200 mesh, desiccant, Davidson Division, Grace Chemical Company, Baltimore, Md. (The performance of other silica gels would have to be evaluated.)

Carborundum chips: 16 mesh. Use as a boiling aid in all evaporation steps. (Whenever ebullition is interrupted, new chips are necessary.)

Aluminum foil: domestic grade.

Teflon wash bottles: Nalgene Co., Rochester, N.Y. Use for adding small amounts of solvents in the boildown and chromatographic procedures. Discard the contents of the wash bottle if left longer than a day.

Glassware

Glassware: borosilicate (DDT and TDE decompose on contact with flint glass).

³Mention of trade names or companies does not imply endorsement by the National Marine Fisheries Service, NOAA.

Glass wool: borosilicate fiber glass, Corning 3950 or equivalent. Use directly from the package.

Graduated centrifuge tubes: 50-ml, Kimax 45164 or equivalent, and stoppered 13-ml, K-410550, Kontes Glass Co., Vineland, N.J., or equivalent.

Modified micro Snyder column: Kontes K-569251 or equivalent.

Chromatographic columns: 7 mm i.d. \times 150 mm and 9 mm i.d. \times 150 and 250 mm glass tubing. (All tubes are without stopcocks.)

Storage bottles: 23-ml screw-cap, borosilicate specimen bottles, Prescription Containers, Inc., Brooklyn, N.Y., with Teflon cap liners, 2390-H62, Arthur H. Thomas Co., Philadelphia, Pa.

Pipettes: Class A. Use for preparation of standards.

Pipettes: disposable glass. Use for quantitative dilution and transfer of samples.

Pipettes: disposable glass, Pasteur capillary, Kimax 72000 or equivalent. Use for sample transfer. (Disposable pipettes are preferred for handling samples in order to eliminate cross contamination.)

Equipment

Virtis homogenizer: Macro, Model 23 or 45, with stainless steel blades, Teflon cap, and 10-50 ml glass homogenizer flasks (No. 16-207), Virtis Co., Gardiner, N.Y., or equivalent. (Bearing lubricants contaminate samples.)

Tube heater: Kontes K-720000. (It is convenient for evaporating solutions in 13-ml centrifuge tubes.)

Plastic and Rubber

Avoid materials, such as gloves, flexible tubing, and polyethylene bags, which often contain PCB or substances which interfere with quantitation by gas chromatography with an electron-capture detector. (Teflon is the only plastic suitable for these analyses.) Wrap samples in aluminum foil for shipment to the laboratory.

Washing Procedure

Chlorinated hydrocarbons and some of the substances in samples adhere tenaciously to glassware. To prevent contamination of future samples, rinse glassware with an organic solvent immediately after use, and then soak in water until washing.

Wash all glassware (and spatulas) in the hottest water practical and rinse thoroughly also with very hot tapwater. (Unless an all-glass still and piping system is available, tapwater is preferable to distilled water because stills often contain plastic tubing and sealing compounds containing PCB.) Bake glassware overnight at 225°-250°C to reduce residual contamination. Cover glassware with aluminum foil. Rinse 3-5 times with petroleum ether or hexane immediately before use. Keep containers covered with foil or stoppers except during evaporation or chromatography to exclude dust, etc.

Procedure For DDT and PCB

The details in time and quantity of chemicals are given only as general guidelines. The exact parameters of these procedures vary with ambient temperature, humidity, altitude, etc. They must be adjusted in each laboratory to optimize recovery, cleanup, and separation of desired components.

Caution

All procedures involving benzene must be performed in a well-functioning hood.

Blank

Carry out the complete procedure as described below without any test sample (omit step 1 under Extraction) to assure that the blank is substantially below the level to be determined. Evaporate the final solutions to 2 ml and quantitate the residues, typically <0.001 ng/μl DDT, TDE, and DDE and <0.01 ng/μl PCB. (Incompletely washed glassware and new batches of reagents are common sources of high blanks.)

Recovery

Carry out the complete procedure with solutions of standards of known concentration in the range anticipated for the samples to assure quantitative recovery. (Some loss of chlorinated hydrocarbons always occurs in the absence of proteins and lipids, which act as keepers. A minimum recovery of 80% is essential. Doping samples originally containing very low levels of chlorinated hydrocarbons gives results which better reflect the accuracy of the method: 85% or higher recovery of DDE, TDE,

DDT, and PCB, and 80-85% recovery of dieldrin and endrin.)

Extraction

1) Weigh approximately 10 g of the material (see Procedure Variations for exceptions), to be analyzed into a Virtis flask and record exact weight to the desired degree of accuracy.

2) Add 20 ml of a 1:1 IPA/benzene mixture to the flask.

3) Homogenize at about 23,000 rpm for 5 min.

4) Rinse homogenizer blade and Teflon cap with hexane so that the hexane drips into the Virtis flask. Fill the flask with hexane nearly to the bottom of the flask neck.

5) Place the Virtis flask in a hot-water bath (ca. 85°C) or sand bath. (An electric fry pan with a layer of sand covered with water provides an economical heating bath.) Boil moderately for at least 45 min, adding hexane whenever the level falls to about one-third of the flask capacity. (If the solution boils too rapidly, material will be lost in the spray. The rate of boiling must be adequate to distill all the H₂O, IPA, and benzene from the flask, because they interfere with the cleanup.) When adding hexane, do it so as to rinse down the sides of the flask as well. Keep the water level in the bath and the hexane level in the flask adjusted so that the flask does not become buoyant and tip over. After 45 min of boildown, reduce the volume of the solution to about 20 ml (ca. 1 cm from the bottom of the flask). Cool. If a layer of water separates, add Na₂SO₄ and allow to stand 1-3 min.

6) Filter through a funnel plugged with glass wool into a 50-ml graduated centrifuge tube. Rinse the flask with three or four 5-ml portions of hexane and pour the rinse solutions through the filter into the centrifuge tube.

7) Concentrate the extract in a hot-water bath to desired volume (20-25 ml), record volume, and pour most of extract into a 23-ml borosilicate screw-cap bottle (with Teflon-lined cap) containing 1 g Na₂SO₄. (The extract can be stored in this condition for extended periods.) (If the extract, prior to concentration, still contains traces of H₂O or IPA, as indicated by cloudiness, add hexane while concentrating in order to remove the H₂O or IPA.)

Oil Determination

8) Pipette exactly 1 ml of the extract into a tared aluminum weighing dish and allow it to

evaporate 4-6 h at room temperature to minimum weight. (Since marine oils oxidize, the weight of oil begins to rise again after a few hours.) Weigh the residue, which contains the oil in 1 ml of extract.

Cleanup

9) Prepare a Florisil column by filling a 9 mm i.d. \times 150 mm glass tube, plugged with glass wool, with ca. 5 cm of Florisil. Wash the column with at least 15 ml of hexane added 1 ml at a time. Allow the hexane level to drop to 1-2 mm, but not to dryness. Pipette 1 ml of the extract onto the column. For samples with very high oil content (refer to step 8 for the amount of oil in 1 ml of the extract), adjust the volume placed on the Florisil so that no more than 0.1 g, and preferably no more than 0.08 g, of oil is placed on the Florisil. Elute with 1-ml portions of hexane; collect the first 12-13 ml of the eluate in a 13-ml graduated centrifuge tube. (Note: Once the Florisil has been wetted, it must always have solvent above it.)

10) If DDT and PCB are not going to be separated, concentrate (in a tube heater) the eluate to an appropriate volume for gas-liquid chromatographic analysis.⁴ If separating DDT and PCB, evaporate the eluate to slightly less than 1 ml.

Separation of DDE, TDE, and DDT from PCB

Quantitation of TDE and DDT is often difficult and quantitation of the PCB is usually impossible unless the DDT family is separated from the PCB. Separation is achieved by chromatography on silica gel. The behavior of DDT and PCB during solid-liquid chromatography is very similar, and obtaining optimal separation requires careful control of all the parameters of the procedure. Even so, DDE does not separate entirely from PCB. Therefore, the DDE in the PCB fraction is quantitated⁵ and included with that in the DDT fraction.

Evaluate the degree of separation of DDE, TDE, and DDT from PCB by chromatographing standard solutions of these compounds according to the procedure described below. Adjust the time and

temperature of activation, the degree of rehydration, the amount of silica gel, and the volume of the pentane fraction to obtain the optimum separation of DDT and TDE from PCB: that is, to maximize the amount of PCB in the pentane fraction and the amount of DDT and TDE in the benzene fraction.

11) Activate the silica gel by heating at 215°C for 16 h. (The time and the temperature are adjusted to obtain an arbitrarily, but consistently activated product with suitable separating characteristics, since complete dehydration occurs over a long period of time.) Cool to room temperature in a desiccator. Rehydrate by placing 98 g silica gel in a glass-stoppered bottle and adding 2 g distilled water. Stopper the bottle and shake and tumble until the water is evenly distributed. Allow the silica gel to equilibrate for 2-4 h before use.

12) Place a portion of this prepared silica gel in a beaker and cover with pentane. Let stand 5-10 min to return to room temperature. (Because the chromatographic columns do not contain stopcocks, a special technique is required for packing.) Quickly transfer silica gel to a glass-wool-stoppered column, 9 mm i.d. \times 250 mm long, wet with pentane. (A disposable transfer pipette with the narrow part of the tip removed works quite well for transferring the silica gel slurry.) Tap the column gently to facilitate packing. Make sure that there is always enough pentane above the column to allow the silica gel to settle slowly in order to eliminate air bubbles and prevent the top of the column from running dry. Pack the column to a height of ca. 8 cm. (Throughout the whole separation procedure the silica gel must always have solvent above it and must be free of bubbles and cracks, which interfere with the desired separation. If the column runs dry or cracks, discard it.) Rinse the column with 15-20 ml of pentane.

13) Allow the pentane level to descend to 1-2 mm (not dry) and place the Florisil eluate (ca. 1 ml) on the column with a Pasteur capillary pipette. Rinse the Florisil eluate tube with three or four 1-ml portions of pentane, and transfer each rinse successively to the column. After the sample and rinses have been adsorbed, fill the tube with

⁴For a detailed description of gas chromatography of chlorinated hydrocarbon pollutants, see the Pesticide Analytical Manual (1977), available from Management Methods Branch, DMS, ACA, HFA-250, 5600 Fishers Lane, Rockville, MD 20857, or National Technical Information Service (NTIS), Springfield, Va. The manual provides extensive background on residue analysis.

⁵Although DDE elutes from the gas-liquid chromatograph at the same time as one of the PCB peaks, measurement of the other five intense PCB peaks provides accurate quantitation of PCB.

Use the amount of PCB in those peaks to determine the size of the peak overlapping the DDE and correct the apparent total DDE (actually DDE plus PCB) to obtain the true DDE concentration. The electron-capture detector is so much more sensitive to DDE than PCB, that the correction affects the accuracy of DDE determination only to a small extent. Consequently the variability in amount of DDE in the pentane fraction does not markedly affect the accuracy of DDE analysis.

pentane. Collect 42 ml of pentane eluate in a 50-ml graduated centrifuge tube. (This fraction contains PCB and some DDE.)

14) After the appropriate volume of pentane eluate has been collected, place a second 50-ml graduated centrifuge tube under the silica gel column. Then fill the tube with benzene. Collect 35 ml of benzene eluate. (This fraction contains most of the DDT complex.) (DDE elutes very rapidly with benzene. If benzene is added to the column before the second centrifuge tube is in place, the DDT complex will often be found in the pentane fraction.)

15) Concentrate each fraction in a boiling water bath to less than the desired final volume and quantitatively transfer with hexane to a volumetric flask of the desired final volume. (The 50-ml centrifuge tubes are not very accurate volumetric containers.) Proceed with gas-liquid chromatographic analysis (see footnote 4).

Notes on DDT/PCB Separation Procedure

1) Elution with hexane instead of pentane during the silica gel chromatography fails to provide the necessary separation of DDT from PCB. Hexane is reported to contain variable amounts of benzene, which would obviously affect an already delicate separation. Use of UV-quality pentane or hexane has been recommended by others, and might allow use of hexane in warm weather.

2) For high residue level samples, evaporation of the Florisil eluate to ca. 1 ml is not necessary; instead an appropriate aliquot is used. However, no more than 1 ml of the eluate should be placed on the silica gel column because the hexane may contain benzene.

Procedure Variations

Plankton and Other High-Moisture Samples

Plankton do not homogenize well using the standard procedure. They also contain water in excess of the amount that 20 ml of 1:1 IPA/benzene can absorb. Addition of Na_2SO_4 before homogenization overcomes both difficulties. Na_2SO_4 not only absorbs water, but also serves as a grinding aid.

To the weighed sample of plankton add 25 ml of 1:1 IPA/benzene; then add 25 ml hexane and 10-15 g Na_2SO_4 . Homogenize 5 min and proceed as usual.

During the 45-min boil-down, scrape the material on the bottom of the flask. Pile up the solids to leave areas of the flask bottom in direct contact with the solvent to improve boiling action and prevent bumping. Cool. If water separates, add Na_2SO_4 .

Filter through glass wool and proceed as usual (step 6 under Extraction).

In order to compensate for the low residue level usually found in plankton, place 2 ml of the extract on the Florisil column for cleanup.

Fishmeals or Dry Feeds

If the standard extraction procedure is used for meals and animal feeds, the finely ground meal forms a layer on the bottom of the Virtis flask which causes bumping and loss of solvent during the 45-min boil-down. Extraction with hexane provides as good recovery as IPA/benzene. This substitution allows omission of the boil-down.

1) Homogenize sample with 20 ml of hexane. Wash down Virtis blades and Teflon top with minimal amount of hexane. Add 10 g Na_2SO_4 .

2) Immediately filter through glass wool tightly wadded to remove as much of the solids as possible. Wash flask and funnel with a minimal amount of hexane so that the volume of the centrifuge tube is not exceeded (35-40 ml).

3) Stir to mix and centrifuge at 1,500 rpm for 45-60 min at 10°C. (There should be about 35 ml of clear solution with less than 1 ml of solids.)

4) Record volume, subtracting 50% of the volume occupied by the solids. (Although this involves an approximation, the error involved should be no more than 2%.)

5) Decant the supernatant liquid into a storage bottle containing 1 g Na_2SO_4 . Proceed with Florisil cleanup (step 9 under Cleanup).

Fish Oil

Homogenization and extraction of oil are unnecessary.

1) On an analytical balance weigh accurately about 2 g of oil into a 50-ml graduated centrifuge tube.

2) Dilute to about 20 ml with hexane and swirl to dissolve the oil completely.

3) Record the volume.

4) Place in a storage bottle, containing 1 g Na_2SO_4 , and proceed with the usual cleanup (step 9 under Cleanup).

NOTE: For greater accuracy in oil analysis, weigh accurately about 2.0-2.5 g oil into a 25-ml volumetric flask. Dilute to volume with hexane. Shake thoroughly. Place sample in storage bottle and proceed at step 9 under Cleanup.

Paper

This procedure is included because occasionally fishery samples come in contact with contaminated packaging and labelling materials, such as carbonless carbon paper and cardboard. Although the procedure has not been validated by collaborative studies, it provides guidelines for an analysis relevant to fishery studies.

Cut the paper (or cardboard) into small pieces, approximately 1 cm on a side, with a scissors or office paper cutter, which has been cleaned thoroughly with iso-octane or hexane. Mix the paper thoroughly and weigh ca. 7 g into a Virtis flask. Add 15 ml distilled water and mix thoroughly with the paper. Allow the mixture to stand 2-5 min, stir again, and dilute with portions of 1:1 IPA/benzene to a total of 70 ml. Homogenize the mixture briefly at low speed. Push the paper down with the Virtis blades, then homogenize briefly. Repeat the process until the paper is completely homogeneous, approximately 10 min total homogenization time. Follow the usual boildown and Florisil procedures.

Procedure For Dieldrin and Endrin

Saponification⁶ and Extraction

1) Weigh 10 g of material to be analyzed into a 250-ml Erlenmeyer flask. For oil, use only 2 g.

2) Dissolve 10 g of KOH in 6 ml distilled water. Slowly dilute with 34 ml of ethyl alcohol (95 or 100%). Swirl until clear.

3) Pour the alcoholic KOH over the sample and heat in a water bath without boiling for 20 min; the exact temperature is not critical.

4) Allow the mixture to cool. Pour the liquid portion into a 250-ml separatory funnel and rinse out the Erlenmeyer flask with 50 ml of water, divided into 4 or 5 portions. Avoid pouring any solids into the separatory funnel. For finely powdered samples like meal, filter the sample through

⁶Saponification in the presence of some proteinaceous materials has been reported to cause degradation of dieldrin. As stated above, recovery studies are always important.

a wad of glass wool and rinse the glass wool carefully with each rinse.

5) Add 15 ml hexane to the separatory funnel and shake for 2 min. Open the stopcock several times during shaking to relieve the pressure buildup. Allow the layers to separate completely, usually about 30 min.

6) Drain off the aqueous layer into the Erlenmeyer flask from which it came. Pour the hexane layer into a 30-ml beaker. Do not let any water escape into the beaker. Cover the beaker tightly with aluminum foil.

7) Pour the aqueous layer back into the separatory funnel and repeat step 5.

8) Drain off the aqueous layer and discard it.

9) Return the hexane extract in the beaker to the separatory funnel. Rinse the sides of the beaker with 1 ml hexane. Add the hexane wash to the extract in the separatory funnel.

10) Wash the hexane extract with 10 ml water by rotating the separatory funnel gently to avoid emulsion formation. *Do not shake.* Allow the layers to separate and discard the aqueous layer.

11) Pour the hexane layer into a 50-ml graduated cylinder. Do not transfer any water to the cylinder. Record the volume. Pour the extract into a 23-ml borosilicate screw-cap bottle (with Teflon-lined cap) containing 1 g Na₂SO₄.

Cleanup

12) Prepare a Florisil column by filling a 9 mm i.d. × 150 mm glass tube, plugged with glass wool, with ca. 4 cm of Florisil. Or use a 7 mm i.d. × 150 mm tube containing ca. 5 cm of Florisil. (The longer column of adsorbent may give slightly better cleanup.) Wet the Florisil with benzene and wash it with 4 to 5 ml benzene added 1 ml at a time. Wash it next with 10 ml hexane (or more) added 1 ml at a time. Pipette 2 ml of the hexane extract onto the column. Put a 12-ml graduated centrifuge tube under the column. Elute with hexane added 1 ml at a time. Collect the first 12 ml of hexane eluate, which contains DDMU (the dehydrochlorination product of TDE), DDE, and PCB. (They may be quantitated if desired.)

Place a second 12-ml centrifuge tube under the column. Change eluant to benzene; add it 1 ml at a time. Collect the first 10 ml of eluate. Place a modified micro Snyder column on the centrifuge tube to prevent loss of residues during evaporation. Concentrate the eluate containing dieldrin and endrin to an appropriate volume (1-3 ml) for

gas-liquid chromatographic analysis (see footnote 4).

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GROWTH OF JUVENILE SPOT PRAWN, *PANDALUS PLATYCEROS*, IN THE LABORATORY AND IN NET PENS USING DIFFERENT DIETS

Floating net pens have been used to culture Pacific salmon, genus *Oncorhynchus*, in the marine waters of the West Coast since 1969 (Mahnken 1975). Although it has been a monoculture effort to date, use of a companion crop species such as the spot prawn, *Pandalus platyceros* Brandt, could diversify and enhance this industry.

In 1975 the National Marine Fisheries Service selected the spot prawn to examine as a potential companion species to net pen-reared salmon. The spot prawn was selected as a candidate for several reasons: 1) it has a rapid growth rate and large size compared with other pandalids (Butler 1964); 2) it can be successfully cultured to maturity in captivity (Prentice 1975); 3) it will reproduce in captivity, often for two consecutive years (Rensel and Prentice 1977); 4) it is gregarious and is normally not cannibalistic; 5) it adapts to vertical or horizontal substrates; and 6) it scavenges for, and accepts, a wide variety of foods (Wickins 1972).

Coincident to investigating the prawn as a companion crop to salmon, several prawn diets were evaluated with prawns held in tanks and net pens at the NMFS Aquaculture Experiment Station on Puget Sound near Manchester, Wash. These experiments were conducted using diets made up of underutilized marine species or fishery byproducts that are available to most salmon farmers.

Materials and Methods

The spot prawns used in the experiments were laboratory-reared progeny of females captured in Hood Canal, Wash. Three concurrent experiments with juvenile prawns (<1 yr of age) began 10 July 1975 (Table 1).

Experiment A was conducted in the laboratory where prawns were held in flowing seawater tanks at 110 animals/m² of immersed substrate. Four diets were evaluated: 1) steamed mussel, *Mytilus edulis*, meat; 2) chopped salmon that had died in nearby net pens; 3) feces and pseudofeces from the Pacific oyster, *Crassostrea gigas* (eight oysters per replicate having a mean weight (total) of 153 g); and 4) no food (control). Diets 1 and 2 were fed every other day while diet 3 was always present in varying amounts. A sample of 10 prawns for each of four replicates was measured during each of