Inter-calibration summary for PCBs, OCPs and PAHs in marine environmental samples

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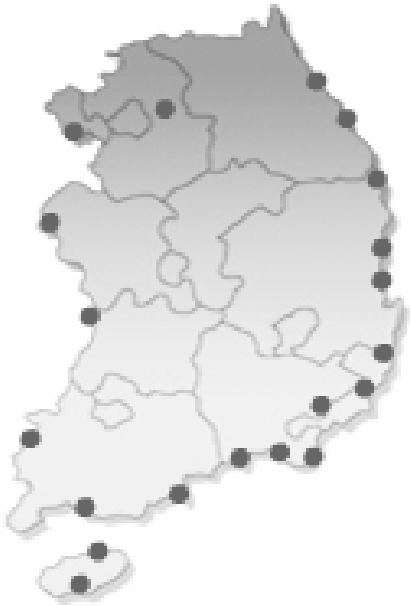
- Polychlorinated biphenyls (PCBs)
- Organochlorine pesticides (OCPs)
- Polycyclic aromatic hydrocarbons (PAHs)
- Summary

NFRDI Introduction

- **National Fisheries Research & Development Institute (NFRDI)**

  - Fisheries Resources
  - Aquaculture
  - Marine Environment



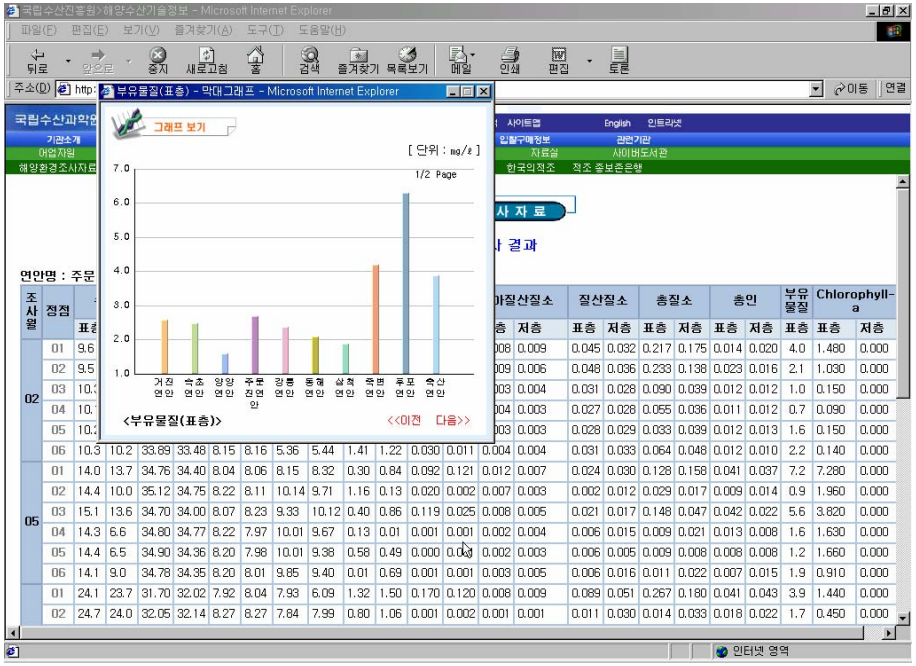
- **Marine Environment Headquarter**

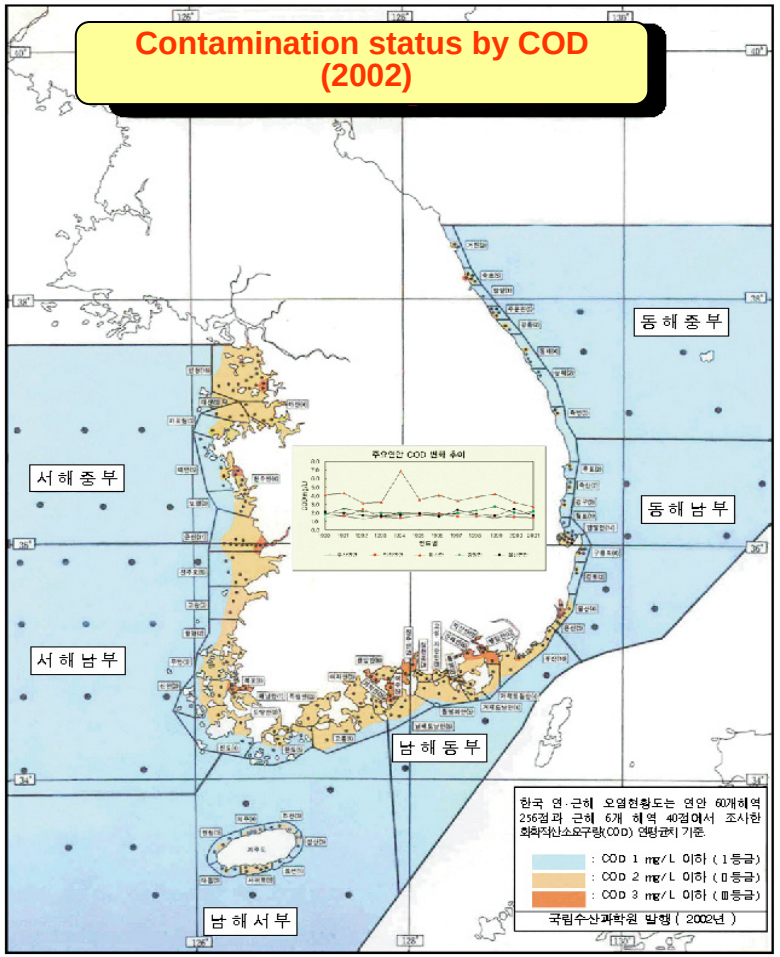
  - Marine Environment Research Team
  - Ocean Research Team
  - Marine Ecology Research Team

Marine Environment Research Team

- **Environment Analysis Laboratory**
  - National Marine Environment Monitoring Network (NMEMN)
  - Trace metals in marine environment
  - Establishment of water quality guideline
- **Endocrine disrupting chemicals (EDCs) Laboratory**
  - POPs monitoring
  - Endocrine disrupting chemicals (EDCs) studies
  - Risk assessment of EDCs by fish consumption
- **Coastal Management Laboratory**
  - Carrying capacity
  - Benthic health assessment
  - Ecological modeling

National Marine Environment Monitoring Network (NMEMN)





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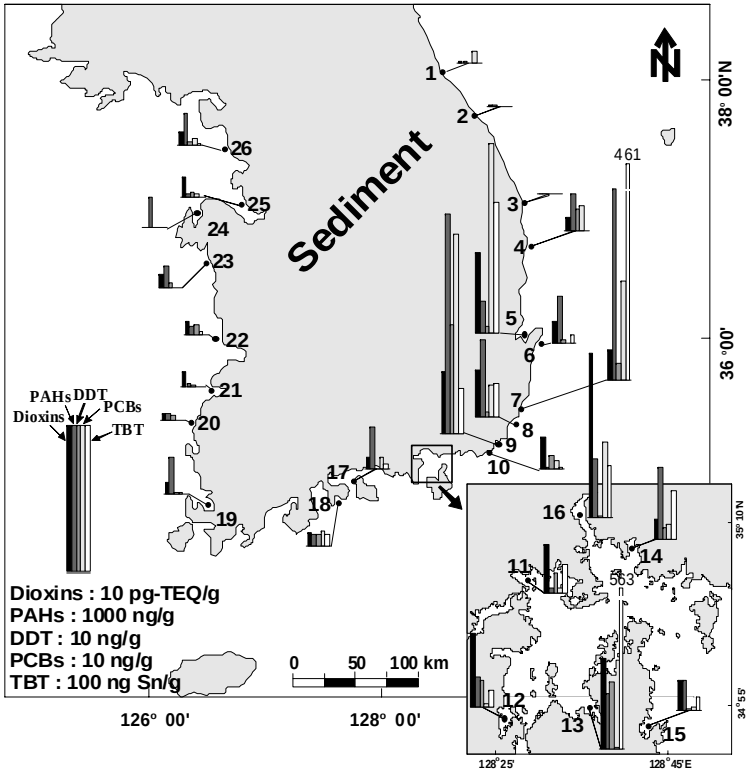
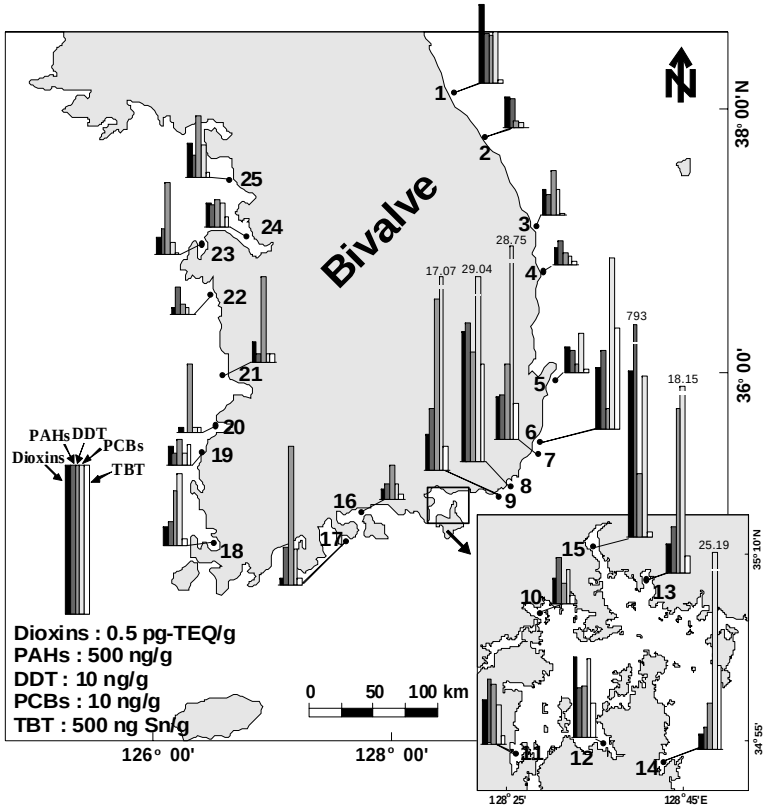
POPs monitoring in Korean coasts





EDCs laboratory introduction

- **Monitoring compounds**
  - Polychlorinated dibenzo-*p*-dioxins and dibenzofurans (PCDD/Fs)
  - PCBs, OCPs, PAHs
  - Phenolic compounds (such as nonylphenol)
  - Butyltins (such as TBT)
  - Fecal sterols (such as coprostanol)
  - Polybrominated diphenyl ethers (PBDEs)
  - Polychlorinated naphthalenes (PCNs)
  - Synthetic musks (HHCB, AHTN), triclosan
- **Facilities**
  - HRGC/HRMS (1), GC (3), GC/MS (3)
  - VOC analytical systems with GC/MS (1)
  - Various extractor (Soxhlet, ASE), Clean room system
- **Staffs**
  - Regular staff: 4
  - Students or technicians: 12

Analytical methods of PCBs and OCPs

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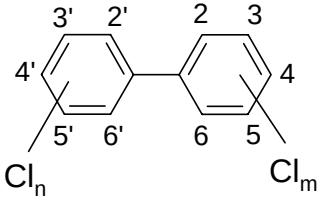
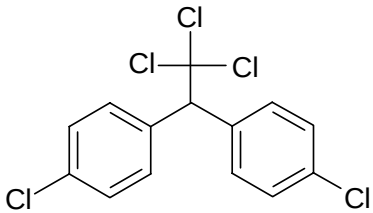
National Fisheries Research & Development Institute

Target compounds of PCBs and OCPs

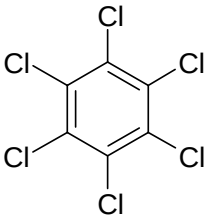
PCBs



- Di-CB: PCB 8
- Tri-CBs: PCB 18, PCB 28, PCB 29
- Tetra-CBs: PCB 44, PCB 52
- Penta-CBs: PCB 87, PCB 101, PCB 105, PCB 110, PCB 118
- Hexa-CBs: PCB 128, PCB 138, PCB 153
- Hepta-CBs: PCB 170, PCB 180, PCB 187
- Octa-CBs: PCB 194, PCB 195, PCB 200, PCB 205
- Nona-CB: PCB 206
- Deca-CB (PCB 209)



OCPs



- Hexachlorobenzene (HCB)
- α-HCH, β-HCH, ɣ-HCH
- *o*,*p*'-DDE, *p*,*p*'-DDE, *o*,*p*'-DDD, *p*,*p*'-DDD, *o*,*p*'-DDT, *p*,*p*'-DDT
- Heptachlor, Heptachlor epoxide
- Aldrine, Endrin, Dieldrine

PCBs and OCPs analyses in sediments

Sediment samples (~ 10 g)

Freeze-drying at -40°C

Spike of surrogates (PCB 103 and dibromooctafluorobiphenyl)

Soxhlet extraction

200 mL (1:1) of hexane and acetone for 8 h

Acid-activated copper treatment

Activated florisil column chromatography

10 g of florisil (60-100 mesh) activated at 450°C for 8 h

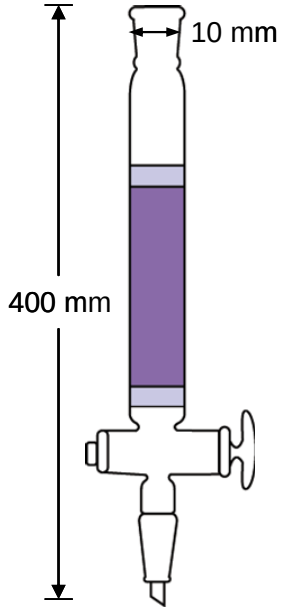
1st Fraction: 100 mL of hexane

2nd Fraction: 100 mL of 20% DCM in of hexane

1st Fraction	**2nd Fraction**
PCBs, HCB, *p,p'*-DDE	**OCPs**

Spike of tetrachloro-m-xylene (TCMX) as internal standard

GC-ECD analysis



PCBs and OCPs analyses in biota

Biota samples (~ 5 g)

Freeze-drying at -40°C

Spike of surrogates (PCB 103 and dibromooctafluorobiphenyl)

Soxhlet extraction

200 mL (1:1) of hexane and acetone for 8 h

Activated florisil column chromatography

10 g of florisil (60-100 mesh) activated at 450°C for 8 h

1st Fraction: 100 mL of hexane

2nd Fraction: 100 mL of 20% DCM in of hexane

1st Fraction

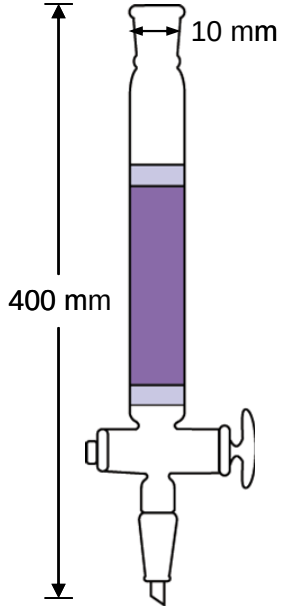
PCBs, HCB, *p*,*p*'-DDE

2nd Fraction

OCPs

Spike of tetrachloro-m-xylene (TCMX) as internal standard

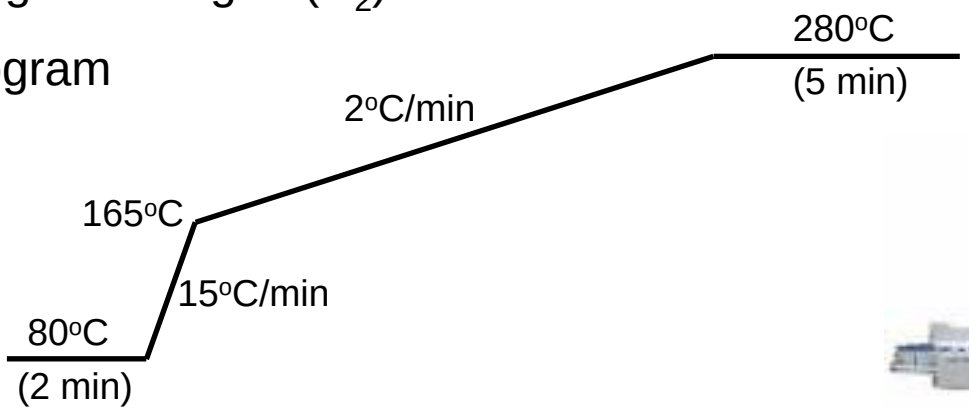
GC-ECD analysis



Instrumental analysis for PCBs and OCPs

- Instrument: GC-ECD (Agilent 6890 Series)
- Capillary column: DB-1701

  (60 m length, 0.25 mm ID, 0.25 μm film thickness, J&W Sci)
- Carrier gas: Helium (He)
- Make-up gas: Nitrogen (N_2)
- Oven program



- Injector temperature: 320°C
- Detector temperature: 300°C

Quality control (QC) activities for PCBs and OCPs

- **Background contamination**
  - Procedural blanks have been processed every 10 samples as real samples
  - Solvent blanks have been used to check carry-over
  - Interference check of surrogates spiked into the samples

- **Recovery test**
  - Spike of surrogate standards (PCB 103 and DBOFB) before extraction
  - PCBs recoveries: 64–103% for sediments; 68–118% for biota
  - OCPs recoveries: 77–102% for sediments; 56–102% for biota

- **Method validation**
  - Analysis of certified reference materials (CRMs)
  - Marine sediments: NIST 1944; Biota: NIST 2978 (mussel tissue)
  - PCBs: Sediment: 71–118%; Biota: 57–98%
  - OCPs: Sediment: 74–100%l Biota: 60-106%

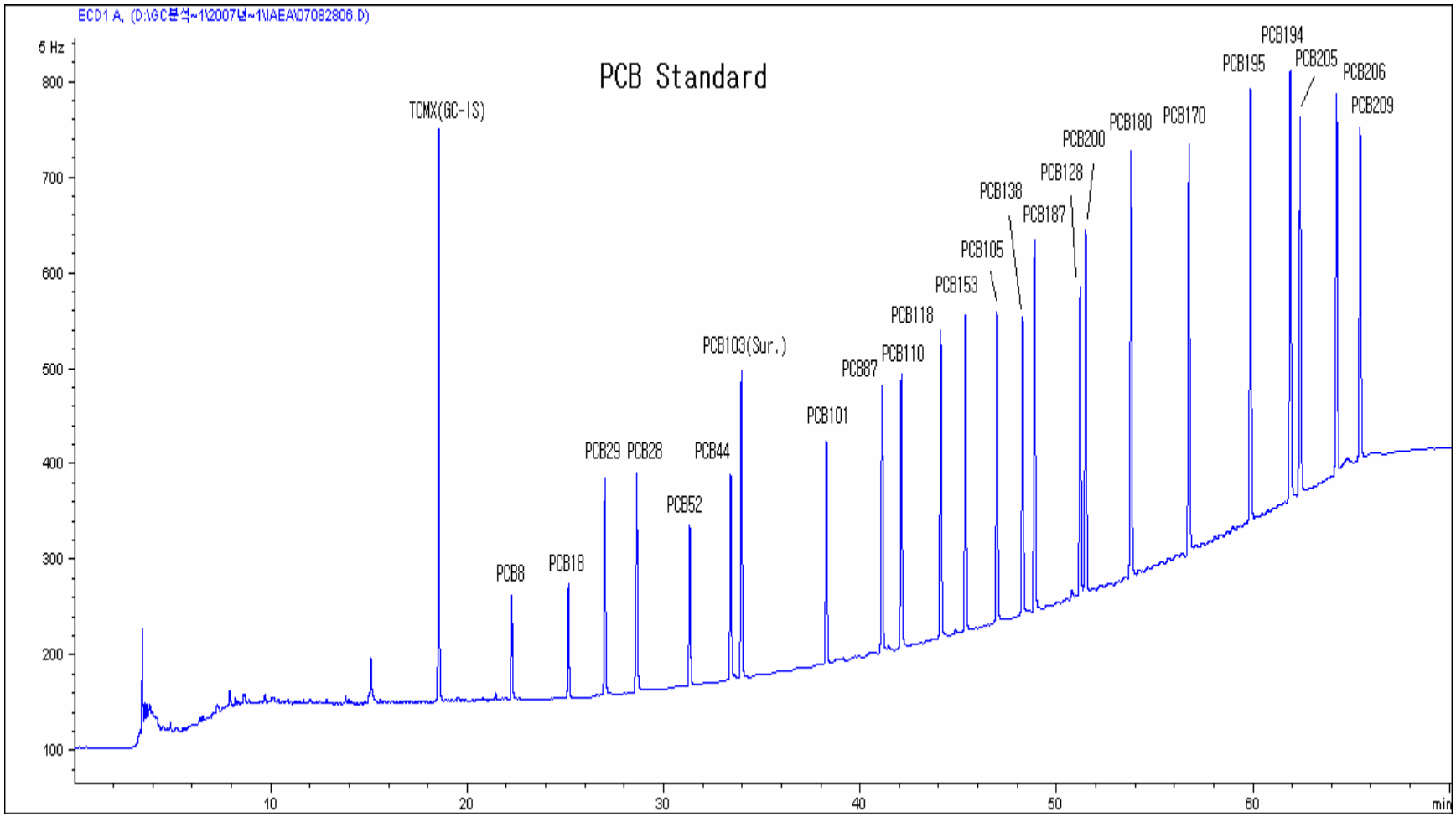
Quality control (QC) activities for PCBs and OCPs

- **Instrument variations**
  - Spike of GC internal standards (tetrachloro-m-xylene, TCMX)
  - Confirmation of PCB and OCP compounds using GC/MDS

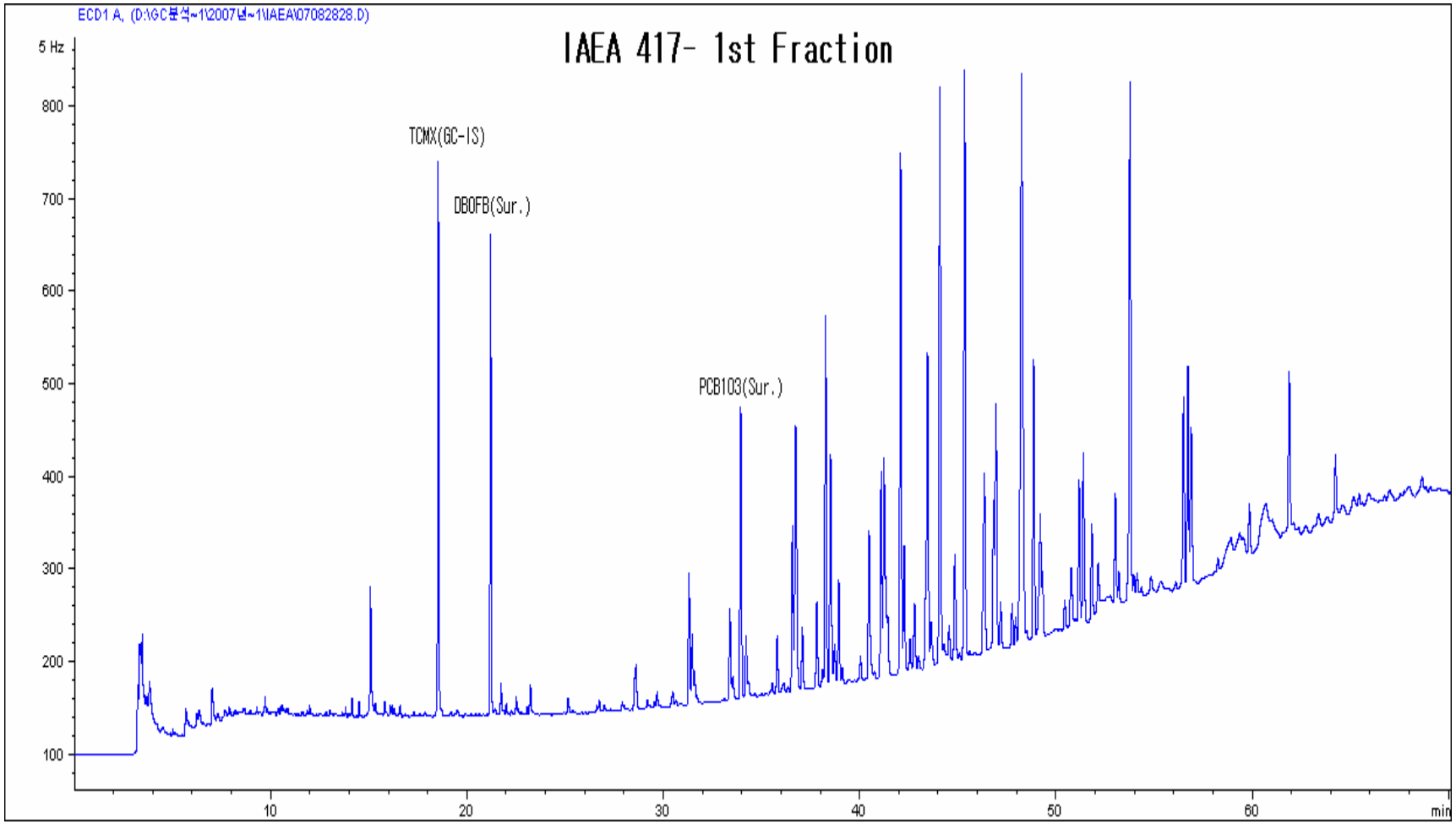
- **Method detection limit (MDL)**
  - Method: 3 times of standard deviation (SD) of repeatability for 7 samples
  - Total PCBs: 1.89 ng/g dw for sediments; 2.75 ng/g dw for biota
  - Total OCPs: 1.2 ng/g dw for sediments; 1.69 ng/g dw for biota

- **Acceptance criteria**
  - Individual concentrations higher than MDLs
  - Recoveries of surrogate standards higher than 50%
  - Repeatability, reproducibility and replicate test are not usual

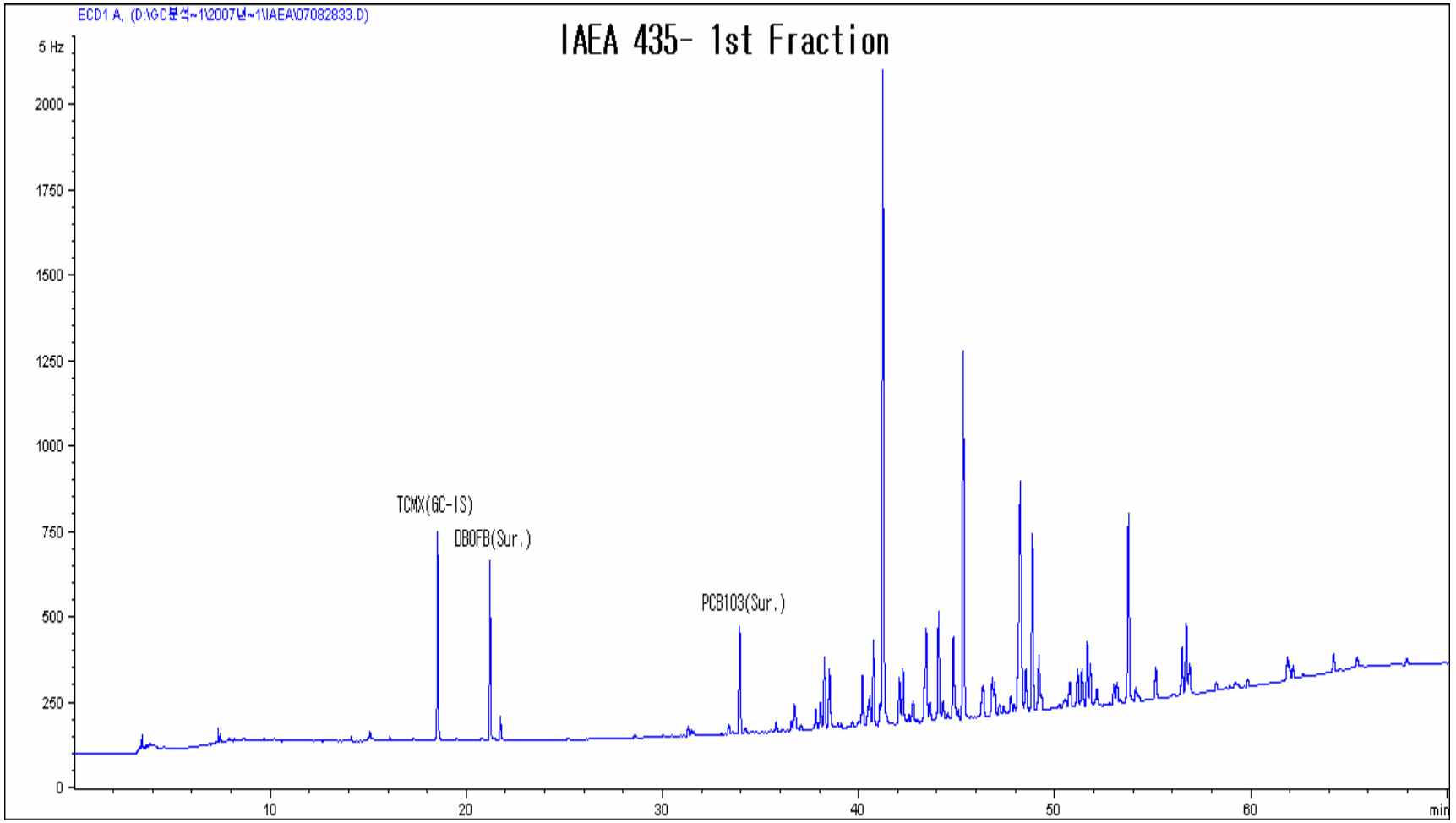
Chromatogram of PCB standards



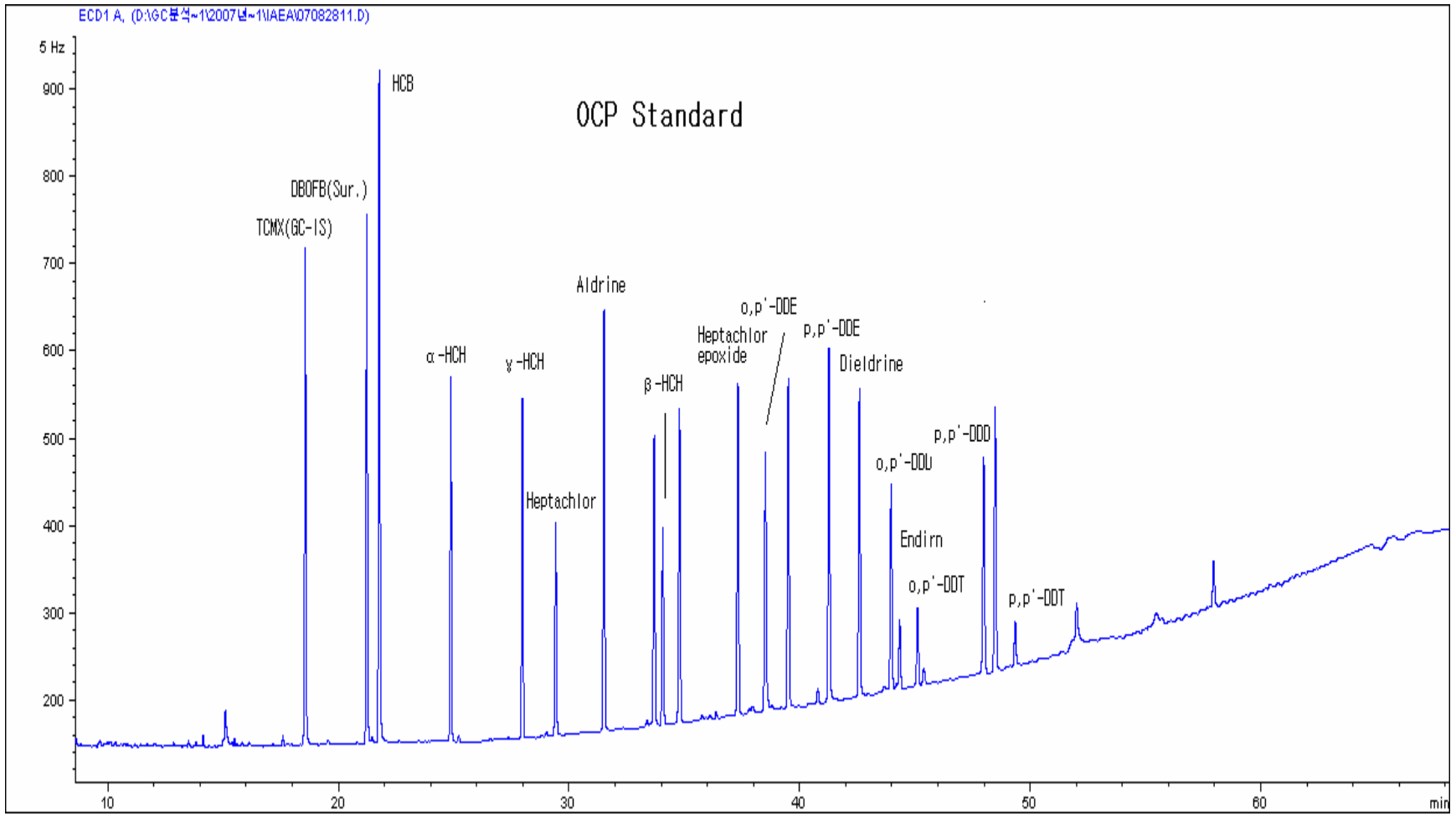
Sediment CRM chromatogram for PCBs



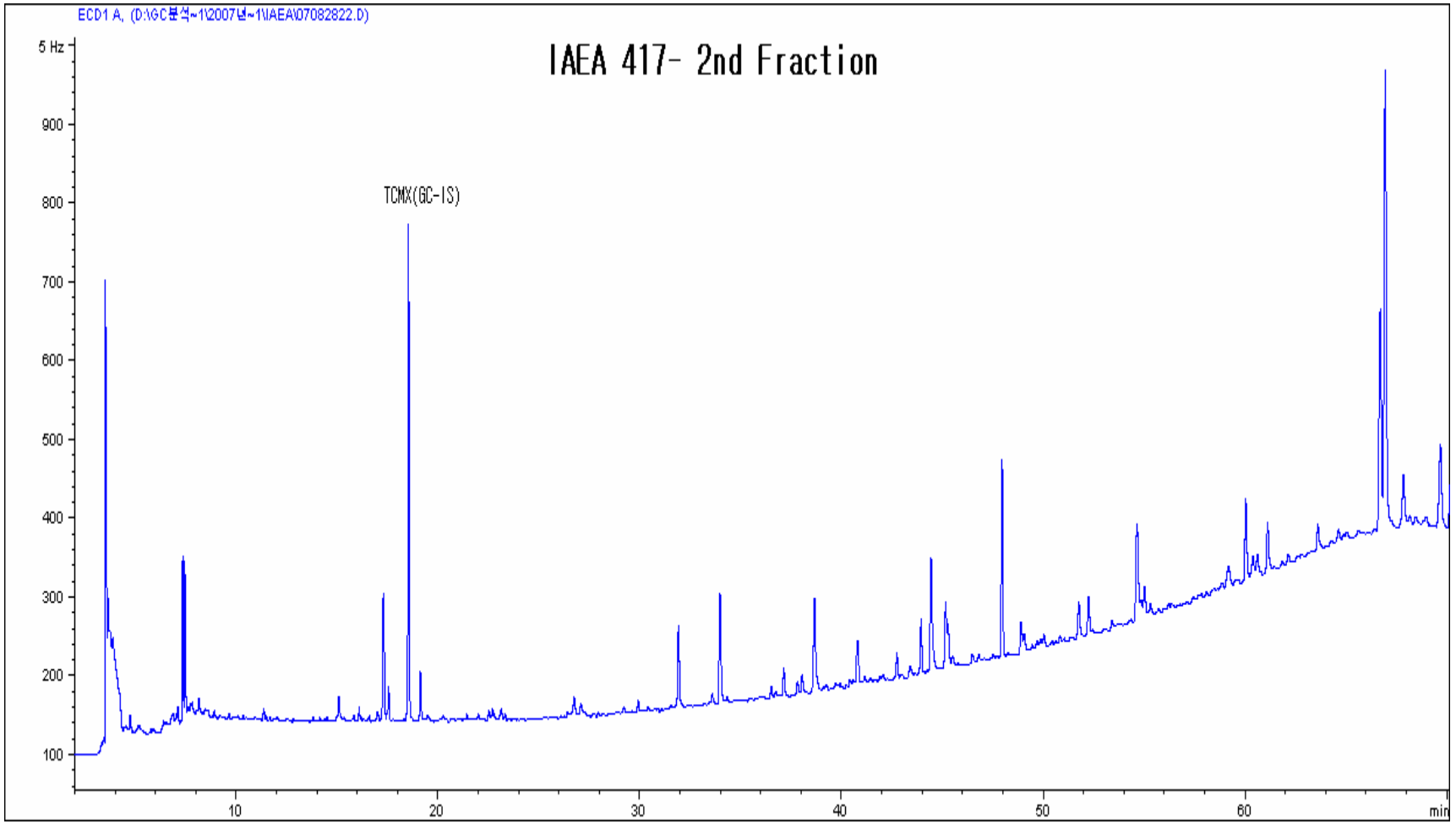
Biota CRM chromatogram for PCBs



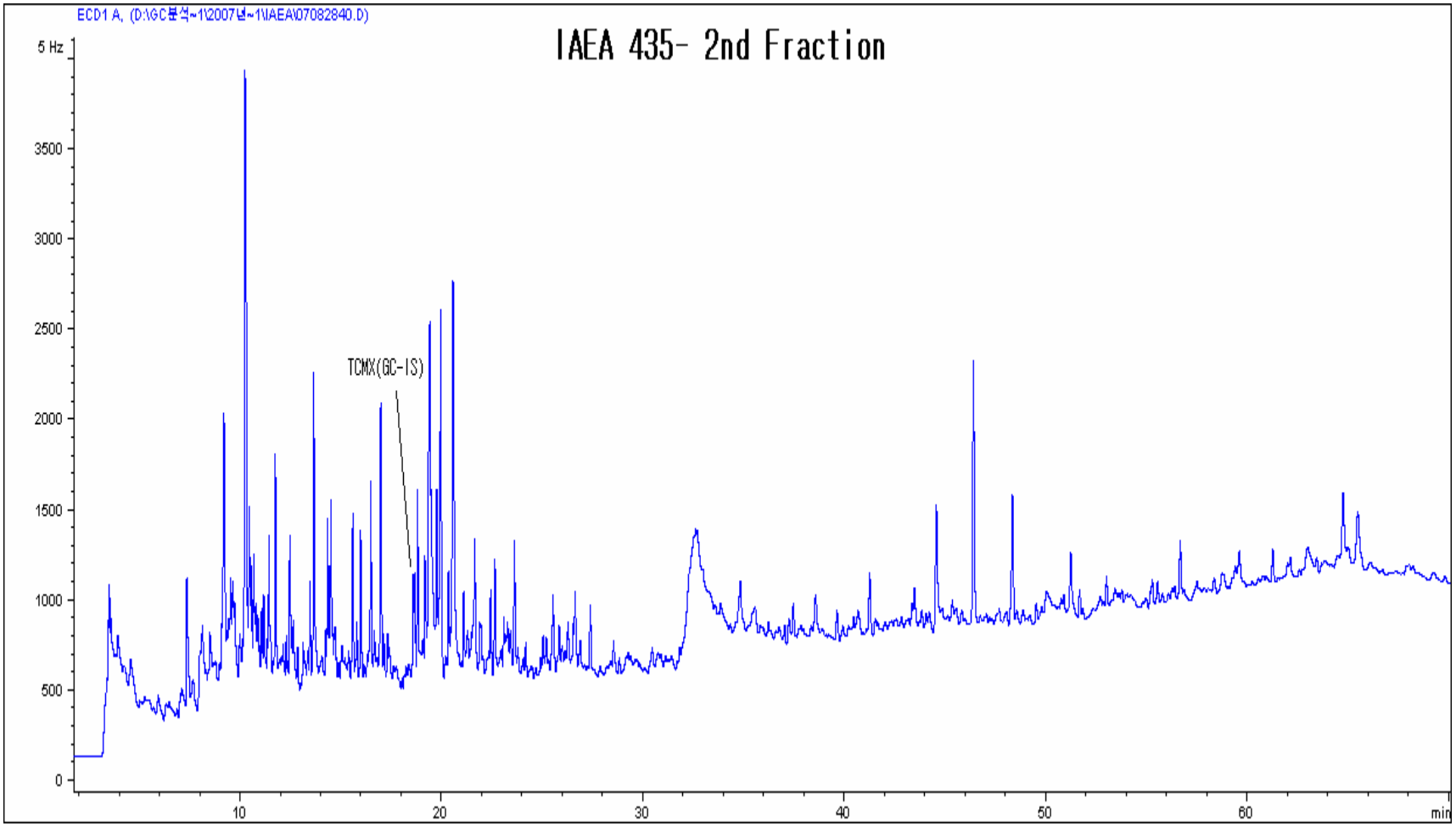
Chromatogram of OCP standards



Sediment CRM chromatogram for OCPs

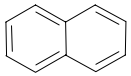


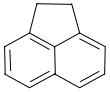
Biota CRM chromatogram for PCBs

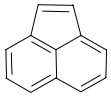


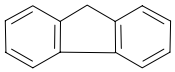
Analytical methods of PAHs

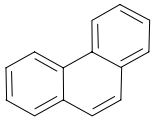
Polycyclic aromatic hydrocarbons

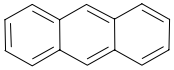
1.Naphthalene (NaP) 

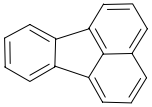
2. Acenaphthylene (AcPy) 

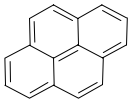
3. Acenaphthene (AcP) 

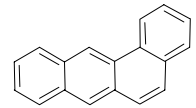
4. Fluorene (Flu) 

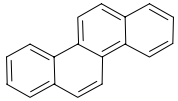
5. Phenanthrene (PhA) 

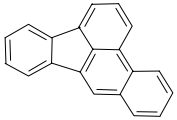
6. Anthracene (AnT) 

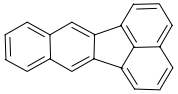
7. Fluoranthene (FluA) 

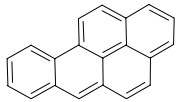
8. Pyrene (Pyr) 

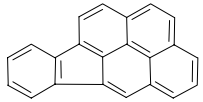
9. Benzo(a)anthracene (BaA) 

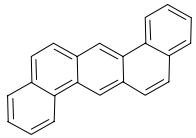
10. Chrysene (Chr) 

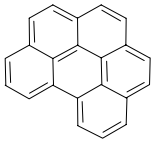
11. Benzo(b)fluoranthene (BbF) 

12. Benzo(k)fluoranthene (BkF) 

13. Benzo(a)pyrene (BaP) 

14. Indeno(1,2,3-c,d)pyrene (InP) 

15. Dibenzo(a,h)anthracene (DbA) 

16. Benzo(g,h,i)perylene (BghiP) 

PAHs analysis in sediments

Sediment samples (~ 5 g)

Freeze-drying at -40°C

Spike of surrogates (NaP-d_8, AcP-d_{10}, PhA-d_{10}, Chr-d_{12}, Per-d_{12})

Soxhlet extraction

200 mL (1:1) of hexane and DCM for 16 h

Acid-activated copper treatment

Activated silica gel column chromatography

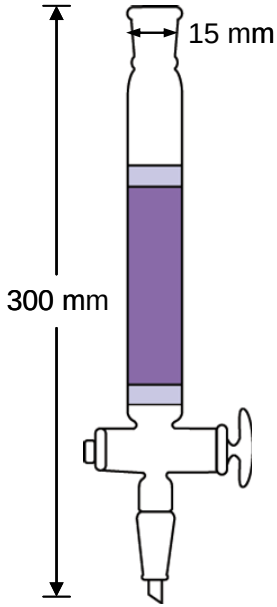
5 g of silica gel (70-230 mesh) activated at 130°C for 4 h

1st Fraction: 16 mL of hexane

2nd Fraction: 50 mL of 10% DCM in of hexane

2nd fraction concentration

GC/MSD analysis (SIM)



PAHs analysis in biota – 1 (by GPC clean-up)

Biota samples (~ 2 g)

Freeze-drying at -40°C

Spike of surrogates (NaP-d_8, AcP-d_{10}, PhA-d_{10}, Chr-d_{12}, Per-d_{12})

Soxhlet extraction

200 mL (1:1) of hexane and DCM for 16 h

Gel permeation chromatography (GPC)

50 g of Bio-beads S-X3 (Bio-Rad Laboratories)

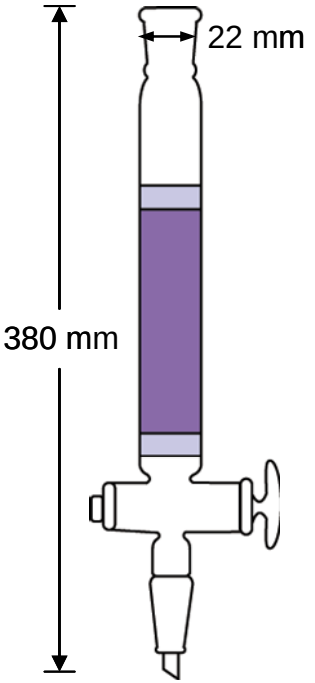
1st Fraction: 100 mL of DCM:hexane (1:1)

2nd Fraction: 150 mL of DCM:hexane (1:1)

Activated silica gel (1.5 g)

2nd fraction concentration

GC/MSD analysis (SIM)



PAHs analysis in biota – 2 (by digestion)

Biota samples (~ 2 g)

Freeze-drying at -40°C

- Spike of surrogates (NaP-d_8, AcP-d_{10}, PhA-d_{10}, Chr-d_{12}, Per-d_{12})

Digestion in 1 N KOH/ethanol

- Water 80 mL + hexane 150 mL for 10 min
- Remove KOH with water
- Remove water with anhydrous Na_2SO_4

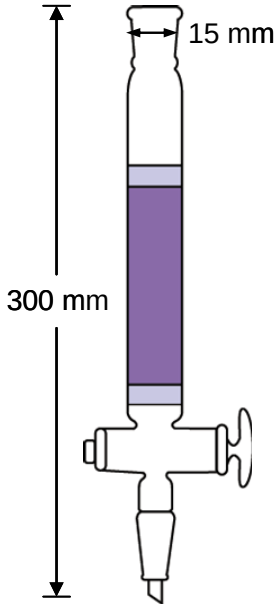
Filteration and concentration

Activated silica gel column chromatography

- 5 g of silica gel (70-230 mesh) activated at 130°C for 4 h
- 1st Fraction: 16 mL of hexane
- 2nd Fraction: 50 mL of 10% DCM in of hexane

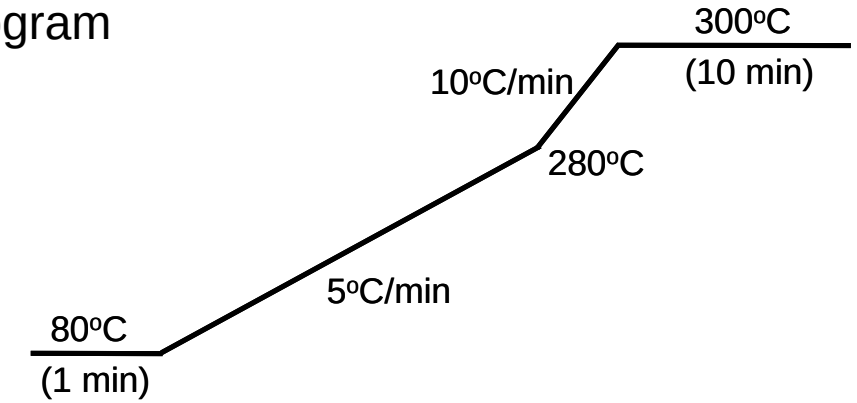
2nd fraction concentration

GC/MSD analysis (SIM)



Instrumental analysis for PAHs

- Instrument: GC/MSD (Agilent 5973N)
- Capillary column: DB-5MS
  (30 m length, 0.25 mm ID, 0.25 μm film thickness, J&W Sci.)
- Carrier gas: Helium (He), 1.0 mL/min (Constant flow)
- Oven program



- Ionization mode: EI+
- Injector temperature: 250°C
- Ion source temperature: 230°C
- Quadruple temperature: 150°C
- Quantification method: SIM (Selected ion monitoring)

Mass spectrometer SIM conditions

Native standards (EPA 610, Supelco)					
Compounds	Abbrev.	Selected molecualr ions			Retention time
		M	M+1	Ratio (%)	
Naphthalene	NaP	128.1	127.1	9.9	8.56
Acenapthylene	AcPy	152.1	151.1	19.6	15.12
Acenaphthene	AcP	154.1	153.1	86.6	15.93
Fluorene	Flu	166.1	165.1	80.4	18.32
Phenanthrene	PhA	178.1	176.1	15.2	22.71
Anthrancene	AnT	178.1	179.1	15.3	22.95
Fluoranthene	FluA			19.4	28.27
Pyrene	Pyr	202.1	203.1	25.9	29.26
Benzo(a)anthracene	BaA			19.4	35.00
Chrysene	Chr	228.1	229.1	22.8	35.14
Benzo(b)fluoranthene	BbF				39.71
Benzo(k)fluoranthene	BkF			22.8	39.83
Benzo(a)pyrene	BaP	252.1	253.1	21.4	40.98
Indeno(1,2,3-c,d)pyrene	InP	276.1	277.1	23	44.68
Dibenzo(a,h)anthracene	DbA	278.1	276.1	22.4	44.83
Benzo(g,h,i)perylene	BghiP	276.1	277.1	23	45.54
Surrogate standards (48902, Supelco)					
Naphthalene-d_8	Int-NaP	136			8.49
Acenaphthene-d_{10}	Int-AcP	164	162		15.79
Phenanthrene-d_{10}	Int-PhA	188			22.61
Chrysene-d_{12}	Int-Chr	240	236		35.03
Perylene-d_{12}	Int-Per	264	260		41.21

Quality control (QC) activities for PAHs

- **Background contamination**
  - Procedural blanks have been processed every 7 samples as real samples
  - Solvent blanks have been used to check carry-over
  - Interference check of surrogates spiked into the samples
- **Recovery test**
  - Spike of surrogate standards (5 species of PAHs) before extraction
  - Recoveries: 72–98% for sediments; 69–104% for biota
- **Method validation**
  - Analysis of certified reference materials (CRMs)
  - Marine sediments: NIST 1944; Biota: NIST 2978 (mussel tissue)
  - Sediment: 68–104%; Biota: 74–117%

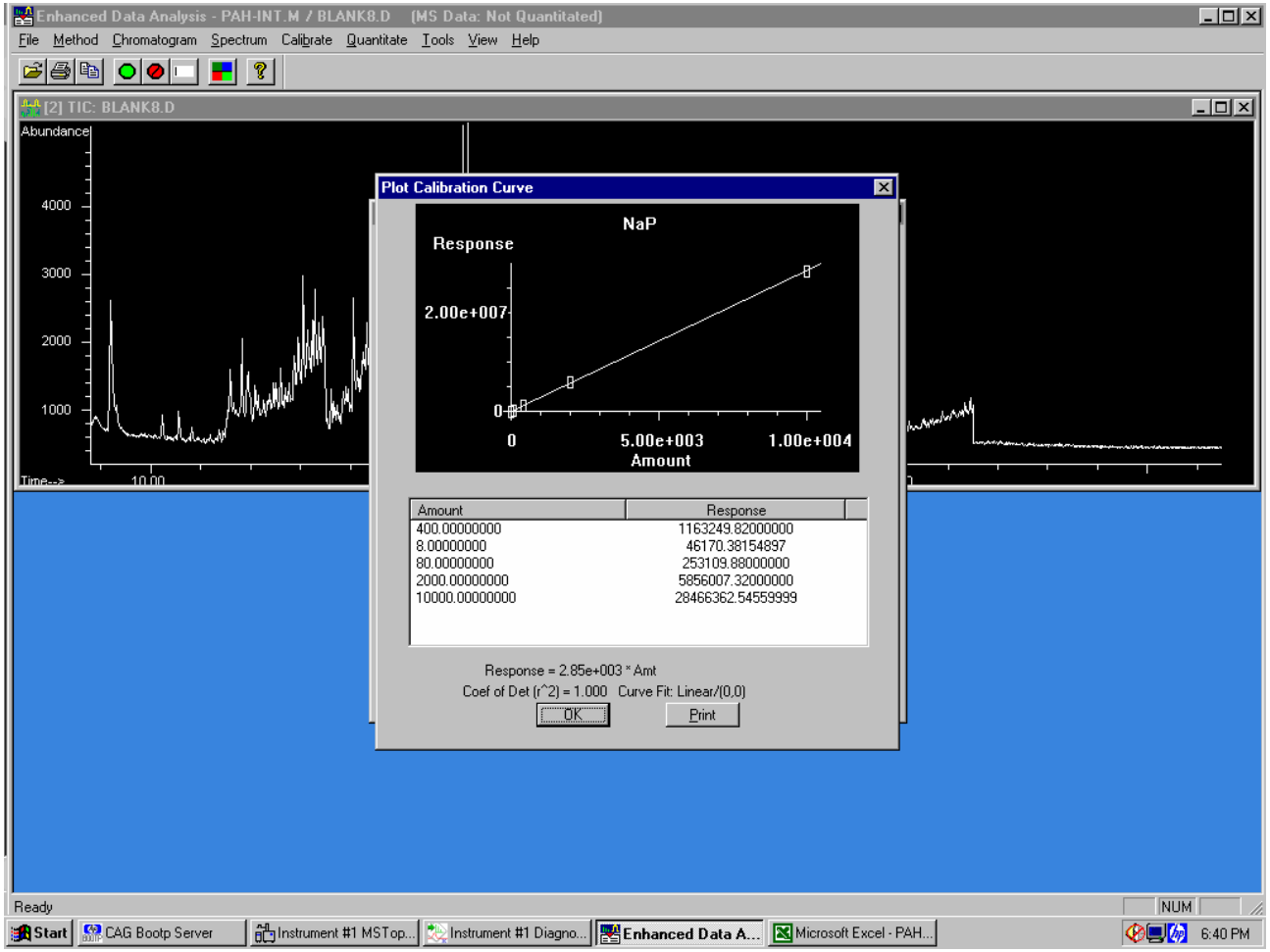
Quality control (QC) activities for PAHs

- **Instrument analysis**
  - Molecular ions confirmation using M and M+1 ions
  - Repeatability sample test (every 10 samples)

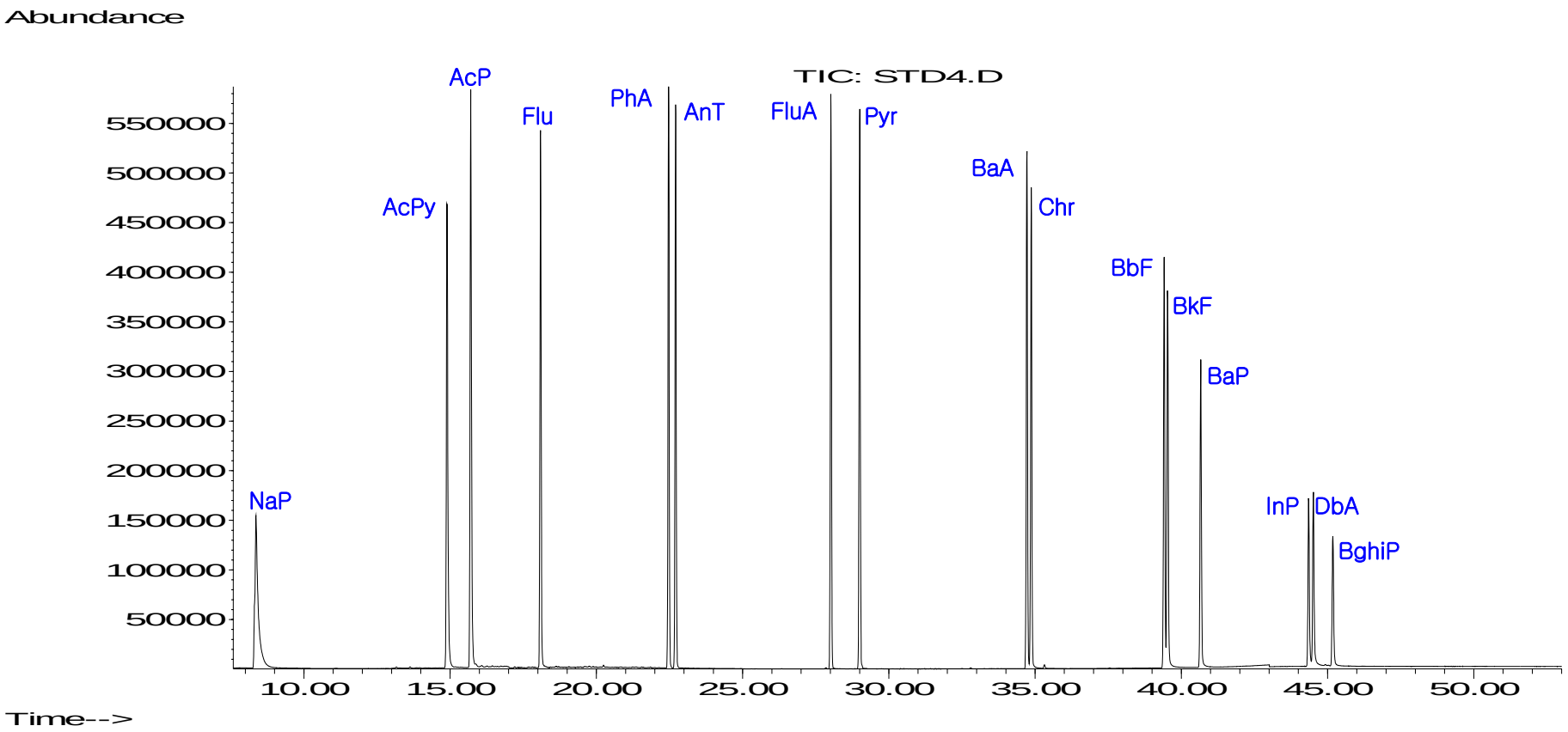
- **Detection limits**
  - Method: three times of signal-to-noise (S/N=3) ratio
  - Individual PAHs: 1 ng/g dry weight for sediment and biota samples

- **Acceptance criteria**
  - Individual concentrations higher than detection limits
  - Recoveries of surrogate standards higher than 50%
  - Reproducibility and replicate test are not usual

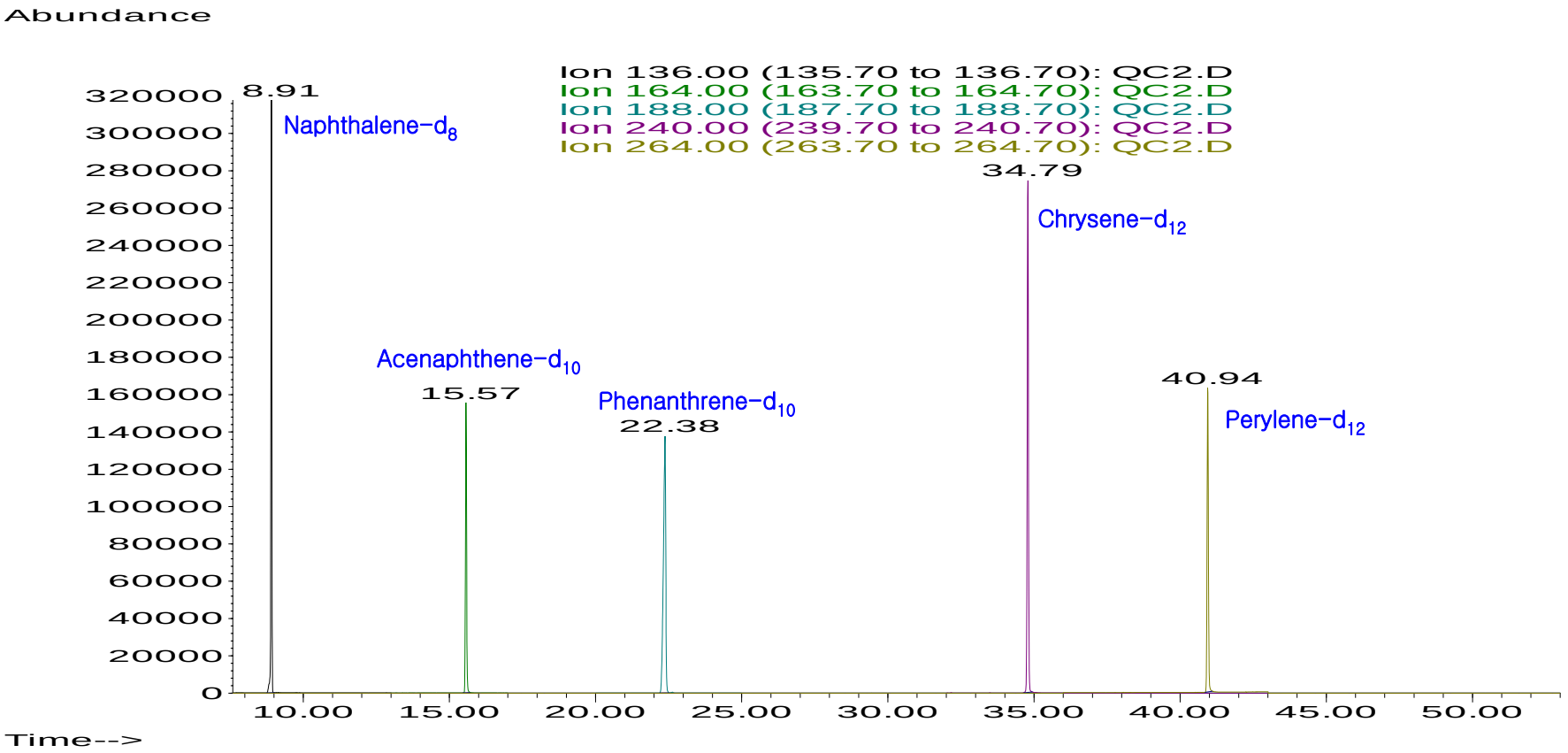
Calibration curve of quantification program for PAHs



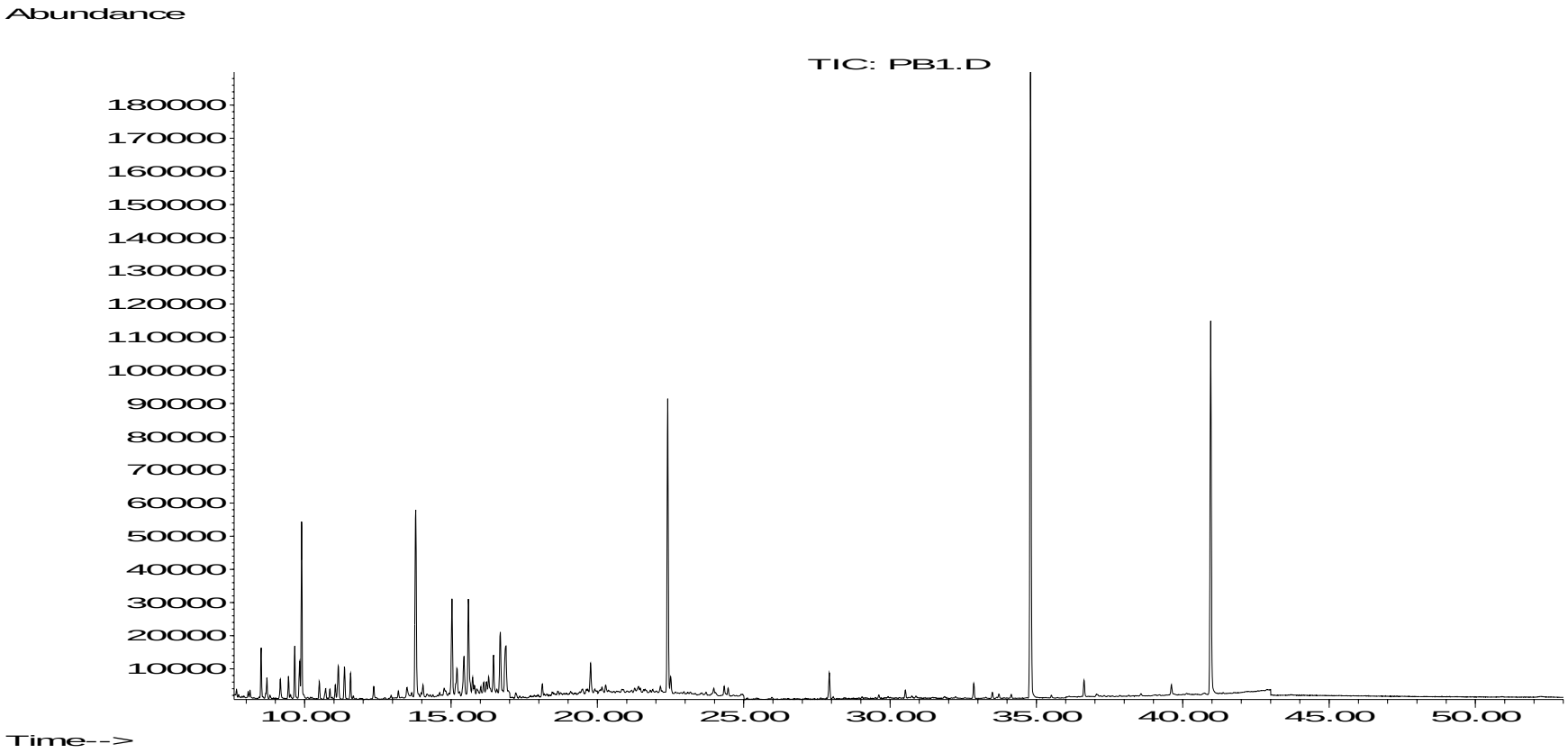
Chromatogram of PAH standards



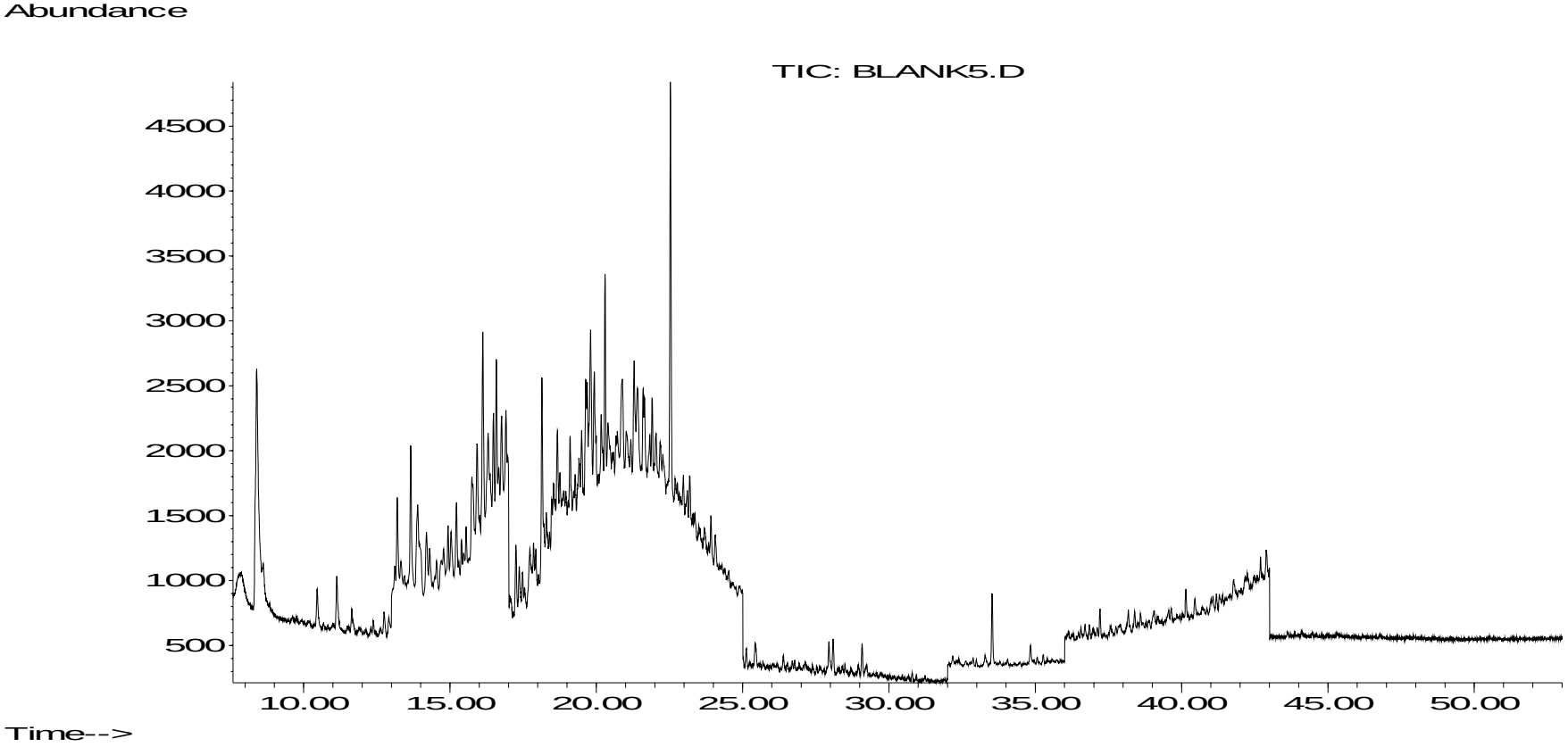
Chromatogram of surrogate standards for PAHs



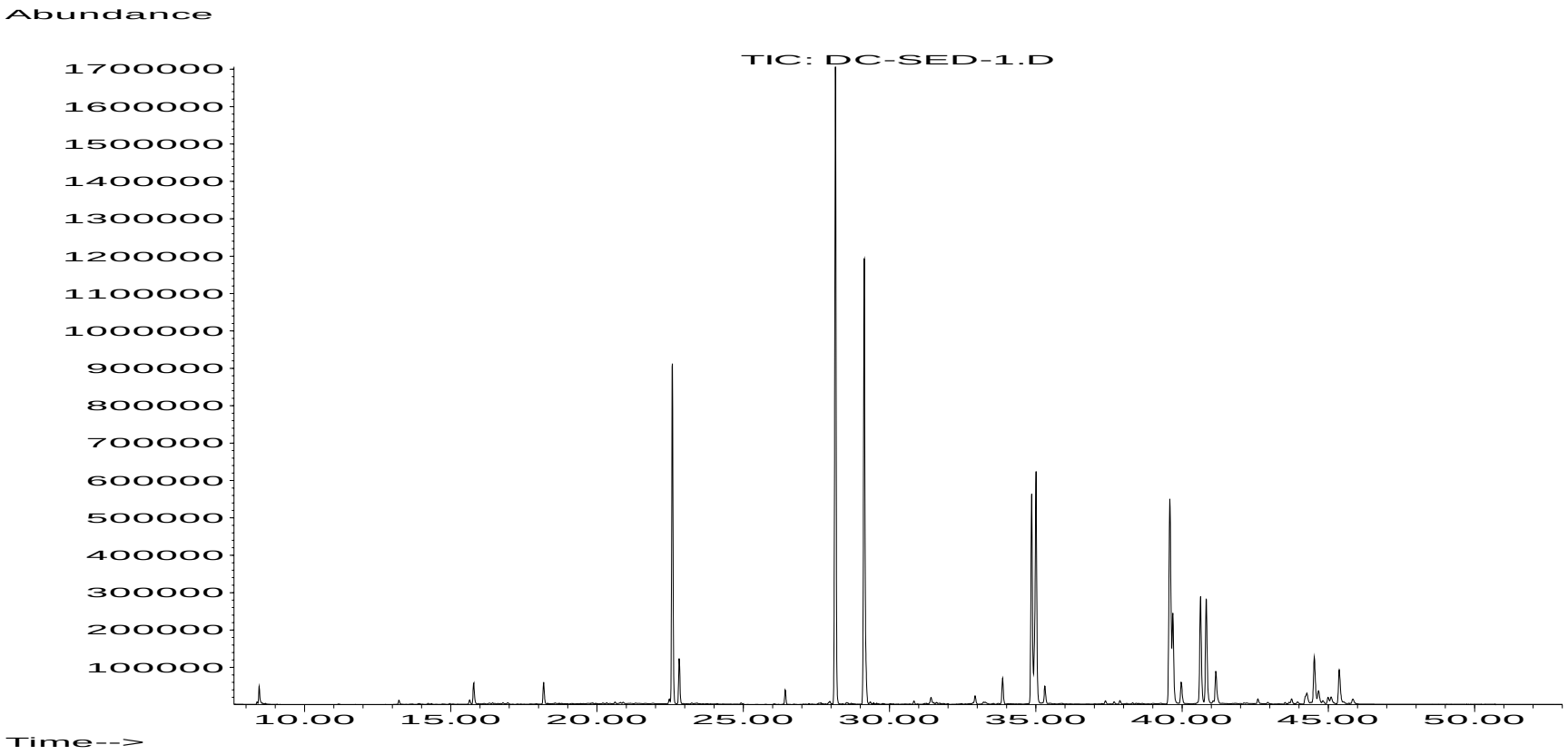
Chromatogram of procedural blank for PAHs



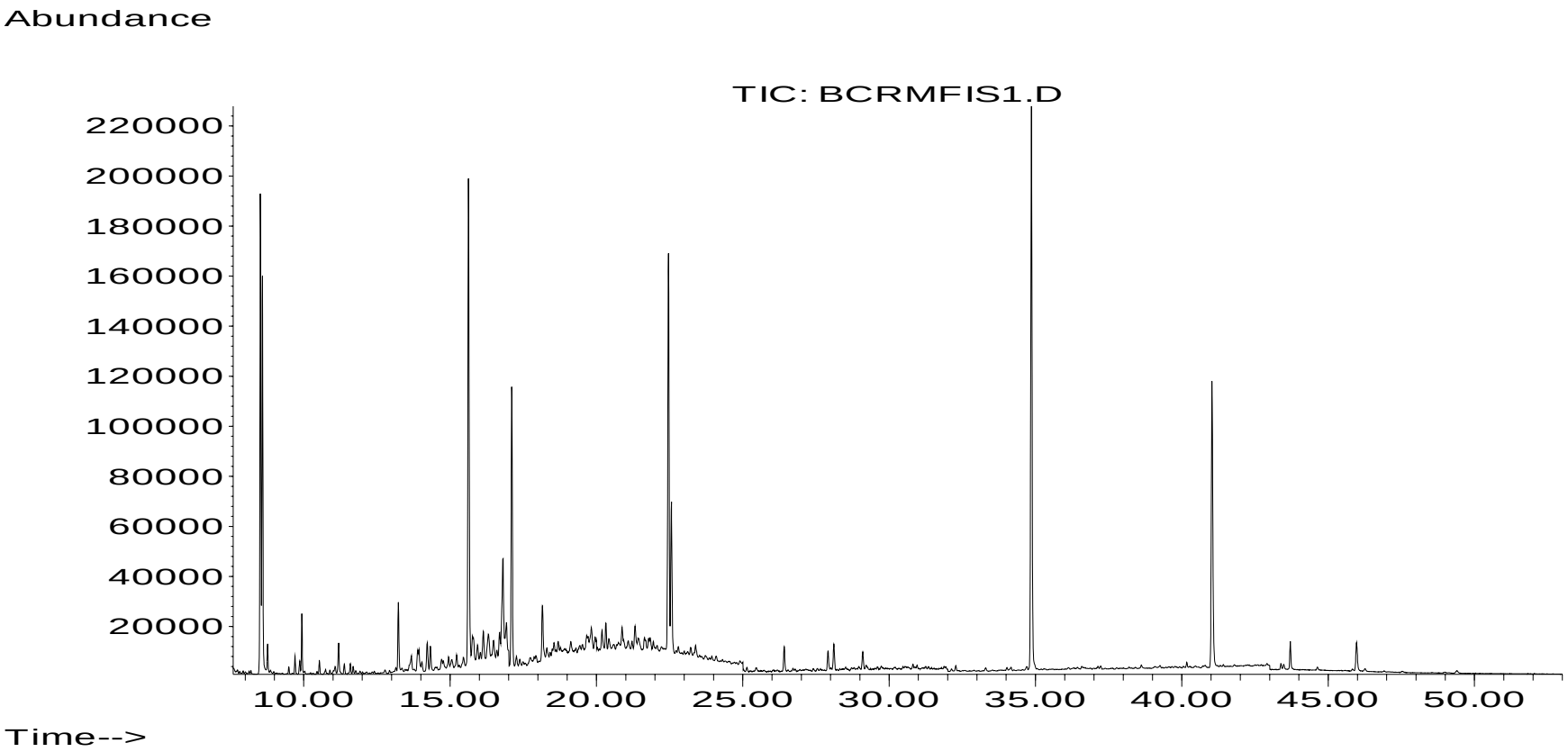
Chromatogram of carry-over blank for PAHs



Sediment CRM chromatogram for PAHs



Biota CRM chromatogram for PAHs



Thank you for your attention.

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