The never ending story of polychlorinated paraffins: New proposals to overcome the persistent quantification problem and new environmental surveys

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Physical properties of CP

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Chloroparaffines (CP) are complex technical mixtures containing thousands of different isomers, congeners and homologues.

- Chlorine content varies between 30 and 70%.
- Technical CP-products are divided into 3 groups: short-chained (sCP, C_{10-13}), mediu-chained (mCP, C_{14-17}) and long-chained CP (lCP, $C_{>17}$).
- Colourless to yellowish, low to highly viskose liquids or waxy solids.
- Thermally stable up to 200-300 °C.
- Can contain additives/stabilisors.

Lit. Muir D. *et al.* in: The Handbook of Environmental Chemistry (Springer) **2000**, 3.

Physical properties of CP, continued

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- Water solubility between 20-1000 μg/l (10-100 times higher than for PCB).
- Vapour pressure comparable to PCB, toxaphene etc. ($1.5\ 10^{-3}$ to $4\ 10^{-6}$ Pa for C_{10} to C_{17} chain)
- Henry constants comparable to PCB and other POPs (0.8-15 Pa m^3 mol^{-1}): Transfer water → air and long range transport possible.
- Highly lipophilic (log K_{OW} 5-7).

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Toxicology

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- Low acute toxicity for mammals.
- Short-chained CP toxic for aquatic organisms (fish).
- Formation of liver, thyroid and kidney carcinomas at mice.
- Teratogenic effects

Furthermore:

- Persistent and high bioaccumulation (BCF ca. 5000 for sCP (rainbow trout))

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Applications of CP

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- High pressure additives in cutting oils.
- Plasticisers (z.B. PVC) and flame retardands.
- Additives in paints, surface coatings, sealants, glues.

Estimated CP-production worldwide ca. 380 000 t/year (2000).

Lit. OSPAR Commission, OSPAR Background Document on Short Chain Chlorinated Paraffins, 2001, No. 41, www. ospar.org.

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Regulation of application

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- Production of sCP banned in Germany since 1. Juni 2003.
- On action list of OSPAR (Convention of the protection of the marine Environment of the North-East Atlantic).
- On priority list of dangerous compounds of the European Water Directive.
- On HELCOM-list of immediate action.

Lit. OSPAR Commission, OSPAR Background Document on Short Chain Chlorinated Paraffins, 2001, No. 41, www. ospar.org.

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Quantitative analysis of CP: Status

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Standard procedure:
Mass spectrometry with electron capture negative ion detection: ECNI-MS

- CP only separatable into a few humps by high resolution gas chromatography (thousands of compounds)
- Detection of the $[M\text{-}Cl]^-$-fragment of each congener- and formula group, respectively.

Combined with: **- high resolution MS (HRMS)**
- low resolution MS (LRMS)

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Limitations and problems of ECNI-MS

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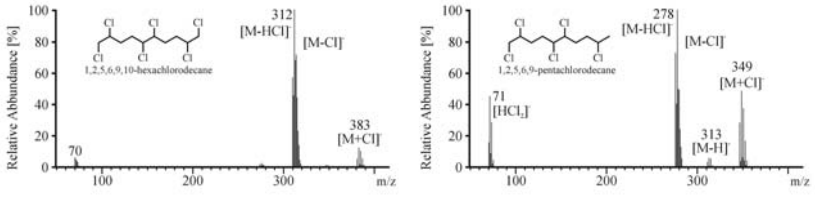
- Simultaneous formation of $[M\text{-}Cl]^-$, M^- and $[M+Cl]^-$ results in disturbances between congeners of different degree of chlorination.
- Other polychlorinated compounds can interfere.
- Response factors depend strongly on degree of chlorination, gives systematic errors. No suitable reference standards.



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Quantification of CP so far

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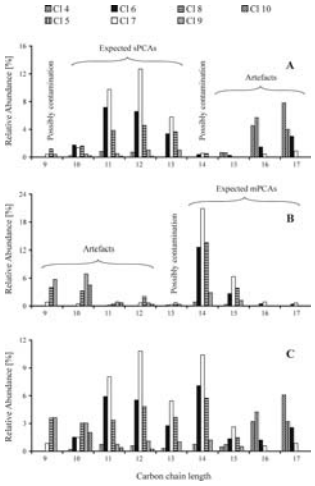
- Quantification of 32 congener groups for sCP (C_9-C_{13}) and 28 for mCP (C_{14}-C_{17}), require 5-10 injections per extract!
- Mass overlaps between sCP and mCP at LRMS.



All C_9-C_{11} (sCP) and C_{14}-C_{16} (mCP) congener groups

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Short chain PCAs			Medium chain PCAs		
PCA congener	Most abundant isotope (100%)	Second abundant isotope	PCA congener	Most abundant isotope (100%)	Second abundant isotope
$C_9H_{14}Cl_6$	298.9 (X+2)	300.9 (X+4, 64%)	$C_{14}H_{26}Cl_4$	299.1 (X)	301.1 (X+2, 96%)
$C_9H_{13}Cl_7$	332.9 (X+2)	334.9 (X+4, 80%)	$C_{14}H_{25}Cl_5$	335.1 (X+2)	333.1 (X, 78%)
$C_9H_{12}Cl_8$	366.9 (X+2)	368.9 (X+4, 96%)	$C_{14}H_{24}Cl_6$	369.0 (X+2)	371.0 (X+4, 64%)
$C_9H_{11}Cl_9$	402.8 (X+4)	400.8 (X+2, 89%)	$C_{14}H_{23}Cl_7$	403.0 (X+2)	405.0 (X+4, 80%)
			$C_{14}H_{22}Cl_8$	436.9 (X+2)	438.9 (X+4, 96%)
			$C_{14}H_{21}Cl_9$	472.9 (X+4)	470.9 (X+2, 89%)
			$C_{14}H_{20}Cl_{10}$	506.9 (X+4)	504.9 (X+2, 78%)
$C_{10}H_{18}Cl_4$	243.1 (X)	245.1 (X+2, 96%)			
$C_{10}H_{17}Cl_5$	279.0 (X+2)	277.0 (X, 78%)			
$C_{10}H_{16}Cl_6$	312.9 (X+2)	314.9 (X+4, 64%)	$C_{15}H_{28}Cl_4$	313.1 (X)	315.1 (X+2, 96%)
$C_{10}H_{15}Cl_7$	346.9 (X+2)	348.9 (X+4, 80%)	$C_{15}H_{27}Cl_5$	349.1 (X+2)	347.1 (X, 78%)
$C_{10}H_{14}Cl_8$	380.9 (X+2)	382.9 (X+4, 96%)	$C_{15}H_{26}Cl_6$	383.0 (X+2)	385.0 (X+4, 64%)
$C_{10}H_{13}Cl_9$	416.8 (X+4)	414.8 (X+2, 89%)	$C_{15}H_{25}Cl_7$	417.0 (X+2)	419.0 (X+4, 80%)
$C_{10}H_{12}Cl_{10}$	450.8 (X+4)	448.8 (X+2, 78%)	$C_{15}H_{24}Cl_8$	451.0 (X+2)	453.0 (X+4, 96%)
			$C_{15}H_{23}Cl_9$	486.9 (X+4)	484.9 (X+2, 89%)
			$C_{15}H_{22}Cl_{10}$	520.9 (X+4)	518.9 (X+2, 78%)
$C_{11}H_{20}Cl_4$	257.1(X)	259.1 (X+2, 96%)			
$C_{11}H_{19}Cl_5$	293.0 (X+2)	291.0 (X, 78%)			
$C_{11}H_{18}Cl_6$	327.0 (X+2)	329.0 (X+4, 64%)	$C_{16}H_{30}Cl_4$	327.1 (X)	329.1 (X+2, 96%)
$C_{11}H_{17}Cl_7$	360.9 (X+2)	362.9 (X+4, 80%)	$C_{16}H_{29}Cl_5$	363.1 (X+2)	361.1 (X, 78%)
$C_{11}H_{16}Cl_8$	394.9 (X+2)	396.9 (X+4, 96%)	$C_{16}H_{28}Cl_6$	397.0 (X+2)	399.0 (X+4, 64%)
$C_{11}H_{15}Cl_9$	430.9 (X+4)	428.9 (X+2, 89%)	$C_{16}H_{27}Cl_7$	431.0 (X+2)	433.0 (X+4, 80%)
$C_{11}H_{14}Cl_{10}$	464.8 (X+4)	462.8 (X+2, 78%)	$C_{16}H_{26}Cl_8$	465.0 (X+2)	467.0 (X+4, 96%)
			$C_{16}H_{25}Cl_9$	500.9 (X+4)	498.9 (X+2, 89%)
			$C_{16}H_{24}Cl_{10}$	534.9 (X+4)	532.9 (X+2, 78%)

Our quantification improvement: Check of isotope signals

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PCA congener	Isotope ratios sPC	mPC	sPCA+mPC	Possible origin of the signal in the s+mPCA mixture
$C_{14}H_{26}Cl_4$	-	-	-	
$C_{14}H_{25}Cl_5$	-	0.75	0.78	mPCA
$C_{14}H_{24}Cl_6$	0.54	1.74	1.56	mPCA
$C_{14}H_{23}Cl_7$	1.19	1.30	1.29	mPCA
$C_{14}H_{22}Cl_8$	0.63	1.07	1.06	mPCA
$C_{14}H_{21}Cl_9$	-	1.09	1.13	mPCA
$C_{15}H_{28}Cl_4$	1.56	-	1.52	sPCA
$C_{15}H_{27}Cl_5$	0.74	0.91	0.75	sPCA
$C_{15}H_{26}Cl_6$	0.60	1.87	1.32	a)
$C_{15}H_{25}Cl_7$	-	1.33	1.27	mPCA
$C_{15}H_{24}Cl_8$	-	1.07	1.09	mPCA
$C_{15}H_{23}Cl_9$	-	1.11	1.23	mPCA

a) Identification not possible

- 1. Exact control of isotope signal to detect and eliminate mass interferences

Our quantification improvement: Retention time ranges

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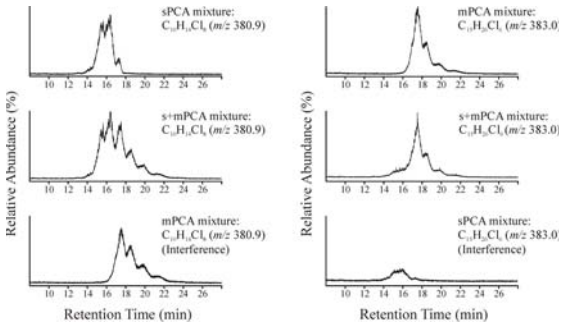

- 2. Exact determination of retention time ranges of the different congener groups.

See: M. Reth, M. Oehme, Anal. Bioanal. Chem. 378, 1741 (2004)

Our quantification improvement: Response factors

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Response factors are influenced by:

- Degree of chlorination of CP-mixture (most important).
- Distribution of chain length (maximum 2-3 % influence).
- Mean molecular weight (maximum 2-3 % influence).

Systematic quantification error of CP-mixtures with 51%, 55% and 63% chlorine content.

	SCCP standard		
Sample	51% Cl	55% Cl	63% Cl
51% Cl	**1500 ng**	616 ng (59 %)	159 ng (89%)
55% Cl	3655 ng (144%)	**1500 ng**	386 ng (74%)
63% Cl	14185 ng (846%)	5822 ng (288%)	**1500 ng**

However: Response factor correlates with degree of chlorination and can be corrected

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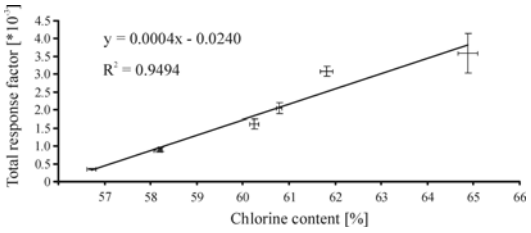

Dependence between total response factor and degree of chlorination for seven sCP-mixtures (51-70 % chlorine content, 5 repetitions).

Remaining maximum errors after correction: typically ca. 5 %

Development of an alternative screening method for CP

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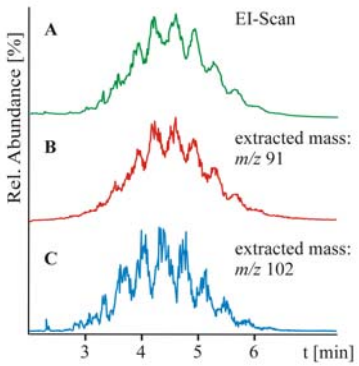
- All CP-compounds have the fragments m/z 102 and m/z 91 in their mass spectra by loss of chlorine/ aromatisation.
- Further selectivity by MS/MS (m/z 105 →65, 91 →53)
- Response factors are nearly structure independent.
- sCP and mCP detectable simultaneously.
- Accuracy <20 %.
- Inexpensive instrumentation (MS-Ion trap for € 60'000.-) or triple-quadrupole.



Technical sCP-mixture, 55.5% Cl

Example: CP in North Sea dab

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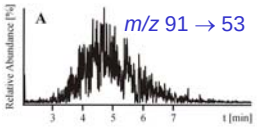
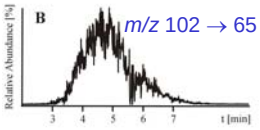
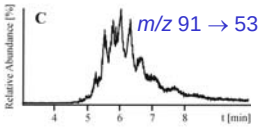
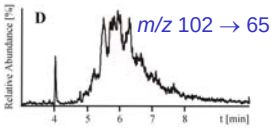
Ion trap (IT)

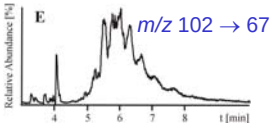




Triple quadrupole (TQ)







➡ Difference between IT and TQ ca. 10%.

A further alternative method: Negative ion detection with CH_2Cl_2+CH_4 as reagent gas

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Addition of CH_2Cl_2 to CH_4 results in:

- Only $[M+Cl]^-$ adducts, therefore no CP interferences
- Response factors very little structure dependent.
- Also low chlorinated Cl_3-congeners detectable.
- Ionisation of polychlorinated interferences suppressed by at leasrt a factor of 20.
- Only method allowing to analyse sediments with a high matrix load.

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CACI-MS and ECNI-MS chromatograms of a sediment sample with high matrix load

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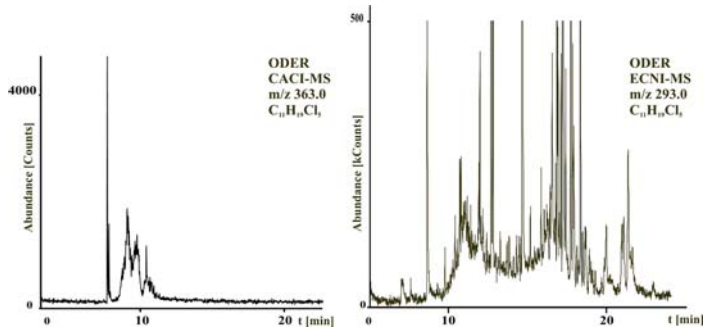

CACI: “Chlorine attachment chemical ionisation”;
ECNI: “Electron capture negative ionisation

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Method comparability?

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	Spiked amount (ng)	ECNI-HRMS (ng)	ECNI-LRMS (ng)	CH_4/CH_2Cl_2-NICI-LRMS (ng)	EI-MS/MS (ng)
sCP	1500	1731 error: 15%	1694 error: 13%	1346 error: -10%	*
mCP	1500	1358 error: -9.4%	1510 error: 0.7%	1454 error: -3%	*
Total amount	3000	3089 error: 3.0%	3204 error: 6.8%	2800 error: -6.7%	2848 error: -5%

* Separate determination not possible

➡ Methods are comparable!

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Which method to apply?

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- **EI-MS/MS:** „Screening“ to detect the presence of CP and determination of total CP concentration.
- **CH_4/CH_2Cl_2-NICI (CACI):** Quantification and determination of congener pattern including $Cl_{3/4}$-congeners, or
- **ECNI-LRMS:** Quantification after response factor correction and determination of congener pattern (>Cl_4).

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Conclusions methodology

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- Alternative methods to ECNI-MS now available.
- Systematic errors of ECNCI by variable response factors minimised.
- Applied methods give comparable results.

CP-burden of North and Baltic Sea

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- Information about CP contamination very scarce worldwide. Comparability of results hardly possible due to quantification problems.
- First study of CP concentration in fish and sediments from North and Baltic Sea. Also first **systematic** study worldwide.

Sample clean-up for CP in sediments

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Extraction
Soxhlet-extraction of 2-50 g dair-dried sediment with
200 ml CH_2Cl_2/*n*-hexane 1+1 (v/v) overnight.

Sulphur removal
during Soxhlet-extraction with aktivated copper powder

Clean-up and removal of interfering chlorinated compounds
1. Silicagel (44 % sulphuric acid conc.)
70 ml *n*-hexane and CH_2Cl_2 (1+1, v/v)
2. Florisil® (deactivated with 1.5 % water (w/w))
Fraction 1 (60 ml *n*-hexane and 5 ml CH_2Cl_2)
Fraction 2 (55 ml CH_2Cl_2, CP fraction)

Screening
Determination of total sCP- and mCP-concentration by HRGC-EI-MS/MS
CID (*m/z*): 91 → 53, 102 → 65, 102 → 67

Determination of sCP- and mCP-concentrations
and congener profiles with HRGC-NICI-LRMS

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Sampling sites and s+mCP concentrations in sediments (ng/g TM)

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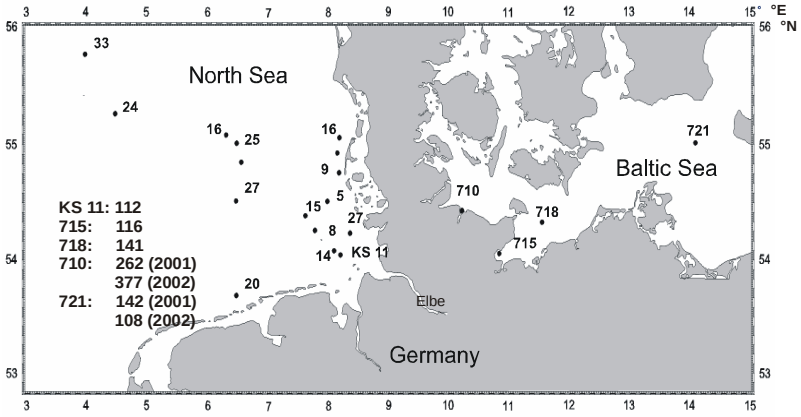

Sampline Aug/Sept 2001/2002 and May 2003

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Chain length profiles

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- ⇒ More mCP (>C_{13}) than sCP
- ⇒ C_{14} dominiates
- ⇒ sCP profiles North and Baltic sea similar, but different for suspended particulate matter from rivers
- ⇒ mCP profiles all very similar
- ⇒ s+mCP Concentrations in Nord and Baltic Sea comparable relative to TOC, but absolutely higher in Baltic Sea.

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Origin of sCP?

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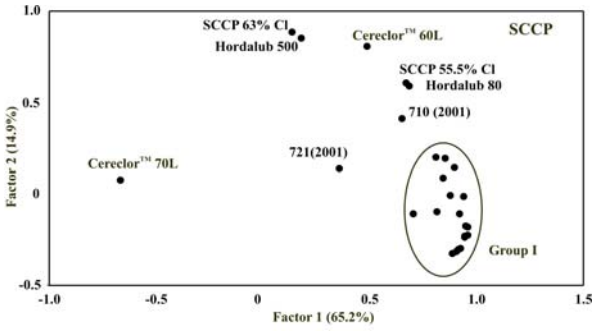

- ⇒ Congener pattern in sediments rather similar to some technical sCP.
- ⇒ Same valid for mCP.

Group 1: SCCP 51.5% Cl, Hordalub 17, 710 (2002 and 2004), 715 (2001), 718 (2001 and 2004), 721 (2002, 2004), ODER, RUDEN, ECKFBU, KS 8 (2003, 2004), KS 11 (2002, 2003 and 2004)

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CP content of fish livers from North and Baltic Sea

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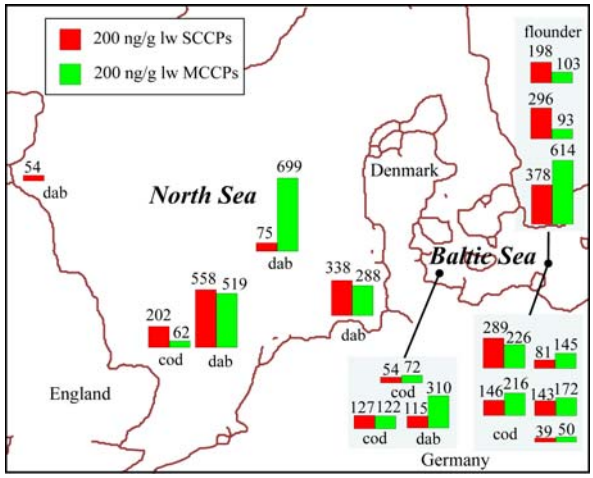

⇒ Typically 5 livers pooled

⇒ Levels comparable to PCBs

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CP concentrations in fish livers from the North Atlantic

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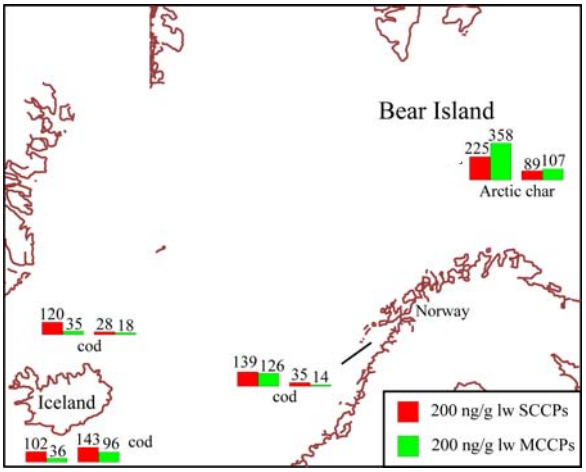

⇒ Concentrations comparable to Baltic Sea

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CP chain length distributionin cod livers: Comparison „Arctic“ versus Baltic Sea

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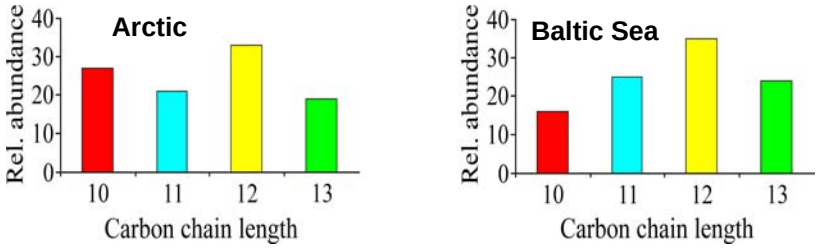

⇒ Fraction of sCP higher in Arctic samples:
⇒ Fraktionation by atmospheric long range transport?

⇒ C_{10}/C_{12}-ratio:
Arctic 0.76 ± 0.36 Baltic Sea: 0.52 ± 0.36 (n=6).

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CP congener patterns in fish liver: Species differences

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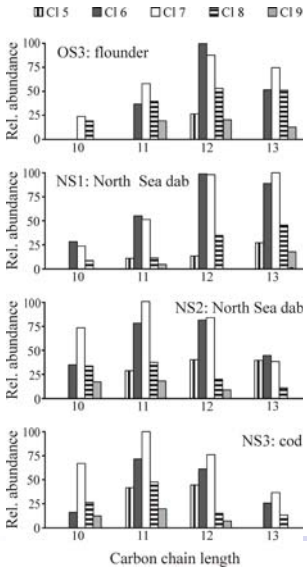

⇒ Considerable differences between species

⇒ Also differences between individuals from the same species and same sampling site.
Reasons?

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A view into the future of CP analysis?

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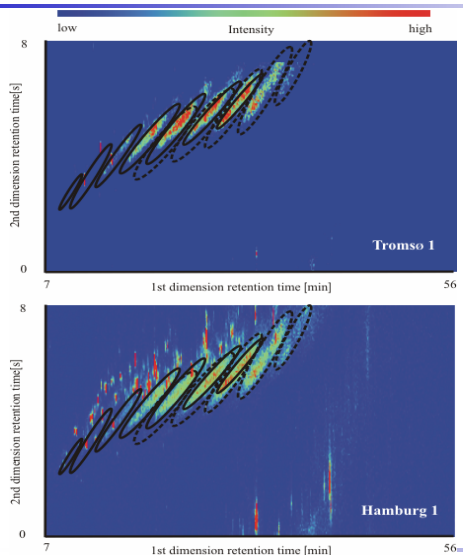

⇒ Comprehensive two-dimensional, HRGC (GC x GC) allows the separation of CPs into chain length groups (here C_{10}-C_{15}) and according to degree of chlorination.

⇒ Examples show clearly that also other compounds are present in the CP fraction (impurities technical mixtures)

Harbour sediments

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Conclusions: Fish livers

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- s- and mCP concentrations in fish liver from North and Baltic Sea show no **species specific differences**.
- s**+mCP concentration ranges of North Sea** (54-3880 ng/g lipid, mean 985 ng/g lipid) and **Baltic Sea comparable** (90-3170 ng/g lipid, mean 615 ng/g lipid).
- Highest **s+mCP contents are >1 ppm**, comparable to **PCB burden**.
- **s+mCP concentrations** in **cod liver from background areas** (Lofot Islands/Iceland) are **lower** (46-265 ng/g lipid, mean 149 ng/g lipid) than in cod from North/Baltic Sea (range 62-3170 ng/g lipid, mean 622 ng/g lipid).
- Cp congener patterns can be different in individuals from the same species.
- Arctic char (200-2500 ng/g lipid, mean1005 ng/g lipid) from the **Bear Island** has comparable CP content **as cod from North/Baltic Sea**.

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Conclusions: Sediments

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- **CP present in all sediments by EI-MS/MS ("screening")** (5-377 ng/g dry weight (dw)).
- **Different analytical methods** give same results within **10-15 %**.
- **CP concentrations in sediments from the Baltic Sea** (45-377 ng/g dw) generally higher than in **North Sea** (5-355 ng/g dw, 10 of 16 samples <50 ng/g dw). However comparable **on TOC basis** (North Sea 2.3-33.1 ng/g TOC, Baltic sea 2.1-9.4 ng/g TOC) .
- **TOC content good indicator for CP content**.
- **Highest CP concentrations** at sites **KS8 and KS11** (Waste and sewage sludge dump sites.
- **mCP concentrations** (C_{14-16}, 42-303 ng/g dw) **always higher than sCP levels** (C_{10-13}, 18-128 ng/g dw). Ratio mCP/sCP 1,7-3,2.

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Acknowledgement

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