Removal of PCDD/Fs and DL-PCBs from fish oil by activated carbon: Compliance with European Legislation

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Introduction

Fish oils are an excellent source of ω-3 fatty acids, mainly eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Several epidemiological studies have shown a correlation between the intake of these compounds and a reduced risk for arterial thrombosis, cardiovascular diseases, multiple sclerosis and autoimmune as well as inflammatory disorders such as psoriasis, asthma and type-1 diabetes[1]. The ω-3 fatty acids are claimed to be essential for normal growth and development and play an important role in the prevention and treatment of hypertension, cancer and several inflammatory and autoimmune disorders[2]. Today, about 2% of the total fats and oils produced in the world are fish oils. During the nineties, they were mainly incorporated into ω-3 enriched margarines but the projected growth from 2000 onwards is in animal feeds and in the production of neutraceuticals (e.g. EPA and DHA enriched products)[3]. However, in Belgium, there is an enormous potential of using purified fish oils in the margarine industry.

Two main issues should be taken into account when considering marine oils as a dietary source of ω-3 fatty acids: (a) their low oxidative stability and (b) their potential contamination with persistent organic pollutants (POPs) such as dioxins and PCBs. The background contamination of crude fish oil with PCDD/Fs is in the range of 2-8 pg WHO-TEQ/g fat while the presence of DL-PCBs in the same sample can vary from 10-100 pg WHO-TEQ/g fat. At present, a maximum PCDD/Fs level for food (human consumption) and feed (animal feeding) purposes of, respectively, 2 and 6 pg WHO-TEQ/g fat in fish oil has been set by the European Commission[4,5]. The upcoming extension of the European Regulations to DL-PCBs will induce more severe specifications and will re-enforce the need of dedicated purification.

One of the main challenges in fish oil refining is to remove these PCDD/Fs and DL-PCBs without losing the valuable minor components and without altering the oxidative status and stability of the oil. Adsorption on apolar adsorbents such as activated carbon is an efficient way to do it.

Materials and methods

The raw material used during the project is a batch of crude cod liver oil (neutralised and winterised) from a Scandinavian fish oil processor.

The adsorbent tested were the following:

· 3 types of bleaching earth (polar adsorbents-variable degree of activation)

· Acid activated silica powder

· Filter aid (Dieatomecious earth)

· 4 types of activated carbon (apolar adsorbents-different suppliers)

Purification of fish oil was performed by mixing the oil with the adsorbent in a rotavapor under different experimental conditions. The oil was subsequently separated from the adsorbent by filtration over a paper filter (Whatman N°1).

Analyses of purified oils were performed by CALUX and GC/HRMS. Only GC/HRMS results are presented here. The comparison between CALUX and GC/HRMS is presented elsewhere[6].

For GC/HRMS analysis of PCDD/Fs and dioxin-like PCBs, samples were cleaned-up using the automated Power-Prep™ system (Fluid Management Systems Inc.). PCDD/Fs and non-*ortho*-PCBs were measured on an Autospec Ultima (Micromass) coupled to an Agilent 6890 Series GC. The column was a 50m VF-5MS (0.20 mm ID x 0.33 µm df) (Varian). Mono-*ortho*-PCBs were measured on an MAT95XL (ThermofinniganMAT) coupled to an Agilent 6890 Series GC. The column was a 25m HT-8 (0.22 mm ID x 0.25 µm df) (SGE).

Results and Discussion

The aims of the project were the understanding of the different purification procedures and their efficiency to remove the contaminants; to preserve nutritional qualities of fish oil after clean-up; to decrease PCDD/Fs and DL-PCBs levels in order to be in compliance with European Legislation. This paper is focused on the third part. As already mentioned, maximum levels for PCDD/Fs have already been set, while discussions of maximum levels for DL-PCBs in food and feed are still ongoing. According to the information available, the trends is to establish a new total TEQ taking into account the PCDD/Fs and the DL-PCBs combined with current PCDD/Fs maximum levels remaining in application for a transitional period[7]. For fish oil dedicated to human consumption, the maximum PCDD/Fs level is 2 pgWHO-TEQ/g fat and the preliminary level under discussion for DL-PCBs would be 6 pgWHO-TEQ/g fat with a total TEQ of 8 pgWHO-TEQ/g fat. In the case of fish oil incorporated in animal feedingstuffs, the maximum PCDD/Fs level is 6 pgWHO-TEQ/g fat and the preliminary level under discussion for DL-PCBs would be 18 pgWHO-TEQ/g fat with a total TEQ of 24 pgWHO-TEQ/g fat.

The first part of the project consisted in the selection of the best adsorbent for PCDD/Fs and DL-PCBs removal with pre-defined operating conditions (0.1 and 0.5% w/w of adsorbent, 70°C and 30 min contact time) in a rotary evaporator (50 mbar). Figure 1 summarizes the results obtained from the same batch of contaminated cod liver oil. Results are presented in pg WHO-TEQ/g fat for the sum of PCDD/Fs, the sum of N-O PCBs and the sum of M-O PCBs. It was decided to separate the total TEQ into these 3 sub-families in order to evaluate the adsorbent efficiency. The contaminated cod liver oil contains 7.1 pgTEQ/g fat of PCDD/Fs, 28.1 pgTEQ/g fat of N-O PCBs and 8.0 pg pgTEQ/g fat of M-O PCBs for a total sum of 43.2 pgTEQ/g fat. Figure 1 shows that only activated carbon used during this study can remove significantly the PCDD/Fs and D-L PCBs. It also indicates that 0.5% (w/w) of activated carbon is more efficient than 0.1% (w/w) in the same experimental conditions. In addition, all activated carbons are very efficient for PCDD/Fs removal (more than 99%). However, the removal of DL-PCBs is lesser and the efficiency varies with the different carbon used. The most efficient carbon is activated carbon (3) (see Figure 1) which can efficiently adsorb the N-O PCBs and up to 50% of M-O PCBs with 0.5% w/w in the oil. In these operating conditions, two samples (treated with 0.5% of activated carbon (1) and (3)) are compliant with the preliminary European Legislation on total TEQ (i.e. 8 pgWHO-TEQ/g oil for human consumption).

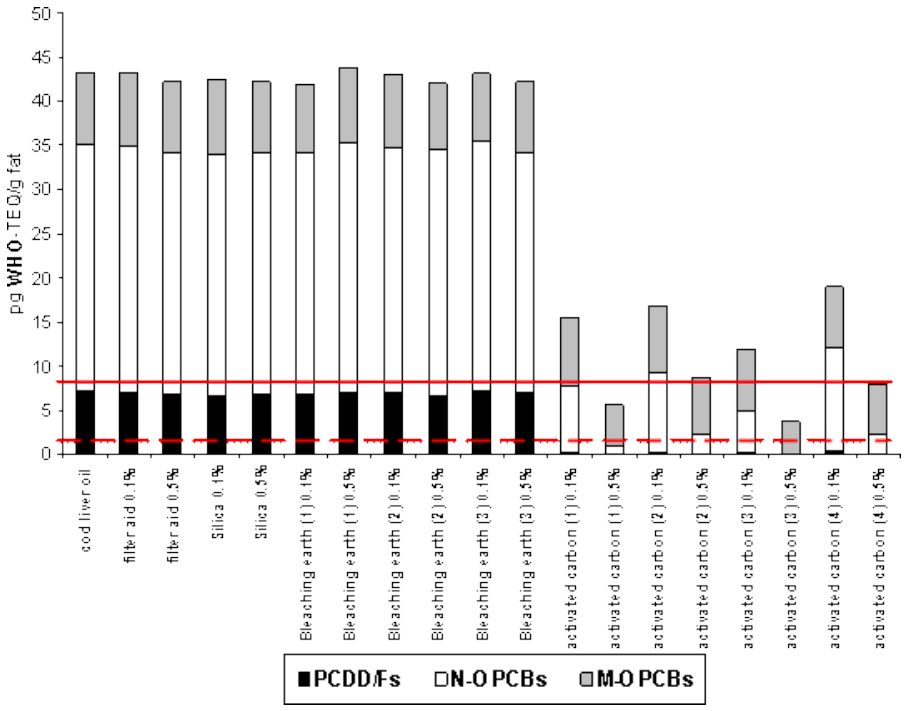

Figure 1: Purification of cod liver oil with different adsorbents.

After selection of the most promising adsorbent, experimental conditions were refined by experimental plan design. Three different variables were tested: time, temperature and amount of activated carbon (3) were investigated by means of a Central Composite Design (CCD). 20 experiments were required to obtain the mathematical model; the range of contact time varies from 10 to 50 min, the range of temperature from 30 to 110°C and the percentage of activated carbon from 0 to 1%. It includes 6 axial points, 8 design points and 6 centre points. An overview of the applied conditions is summarized in table 1.

It can be concluded from the CCD that, firstly, reaction time within the range (10 to 50 min) has virtually no influence on the adsorption of contaminants. Secondly, there is an optimum temperature to maximize the adsorption of PCDD/Fs and DL-PCBs at 74°C. Thirdly, the percentage of activated carbon is the most influential variable. The higher the % (w/w) of active carbon used the higher the adsorption of contaminants.

In these optimum conditions with a minimum of 0.5% (w/w) of activated carbon, Figure 2 shows that PCDD/Fs and DL-PCBs levels can be efficiently decreased well below the preliminary proposed total TEQ values. After purification, the remaining PCBs that contribute to the total TEQ are the M-O PCBs and especially PCBs 118 and 105.

Table 1: Overview of the applied process conditions

CCD	AC (%)	Temp (°C)	Time (min)	Point
Sample 0	-	-	-	crude oil
Sample 1	0	70	30	Axial
Sample 2	1	70	30	Axial
Sample 3	0,5	30	30	Axial
Sample 4	0,5	110	30	Axial
Sample 5	0,5	70	10	Axial
Sample 6	0,5	70	50	Axial
Sample 7	0,25	50	20	Design
Sample 8	0,25	50	40	Design
Sample 9	0,75	50	20	Design
Sample 10	0,75	50	40	Design
Sample 11	0,25	90	20	Design
Sample 12	0,25	90	40	Design
Sample 13	0,75	90	20	Design
Sample 14	0,75	90	40	Design
Sample 15	0,5	70	30	Center
Sample 16	0,5	70	30	Center
Sample 17	0,5	70	30	Center
Sample 18	0,5	70	30	Center
Sample 19	0,5	70	30	Center
Sample 20	0,5	70	30	Center

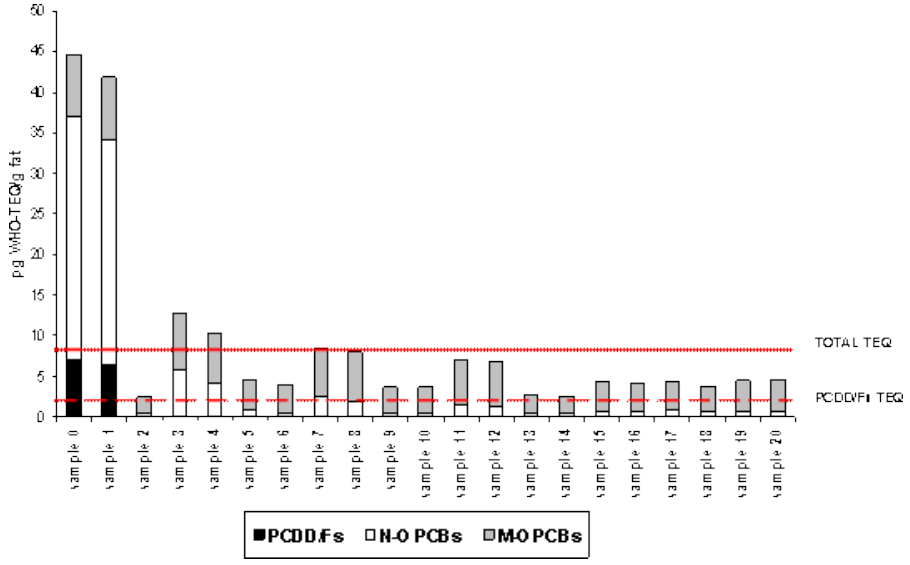

Figure 2 : Removal of PCDD/Fs and DL-PCBs for the applied CCD.

Acknowledgements

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