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UV Treatment of Fishmeal: A Method To Remove Dioxins?

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This study evaluates the use of UV on contaminated fishmeal and photodegradation of dioxins. Fishmeal samples were placed under UVA or UVB light for 2, 5, and 10 days. Subsequently, analysis of amino acid content, lipid oxidation marker, ethoxyquin content, dioxin, and polychlorinated biphenyl (PCB) profiling was carried out. Exposure of fishmeal for 5 days to UVB light resulted in the degradation of approximately 70% of the dioxin content, while UVA had little effect, only resulting in the degradation of 10% of the dioxin content. UVB did not affect the protein and amino acid content of fishmeal; however, lipid oxidation was triggered. Addition of ethoxyquin prevented oxidation but simultaneously slowed dioxin breakdown. Increasing UVB intensity resulted in a more efficient dioxin degradation of 90%. Exposure of fishmeal to UVB also resulted in an increase in PCBs. UVB light is shown to photodegrade dioxin in fishmeal, indicating the needs to further investigation of methods for application at industrial scale.

KEYWORDS: Fishmeal; UV; dioxins; TCDD; PCBs

INTRODUCTION

Dioxins are ubiquitous polychlorinated organic pollutants mainly introduced into the environment through industrial activity and waste management via, for example, pesticide production and incineration (*1*, *2*). Dioxins generally include two classes of compounds, polychlorinated dibenzo-*p*-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs). Depending on their degree of chlorination and the position of the chlorine atom on the benzene rings, many congeners exist. Only 7 congeners for dioxins and 10 for furans are considered toxic. The most toxic of the dioxin congeners is 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (2,3,7,8 TCDD). Because they are not equally toxic, congeners have been weighted in relation to each other by determining a toxic equivalency factor (TEF). The total dioxin content in fishmeal is obtained by weighting each of the 17 measured concentrations of the toxic congeners with their respective TEF value and summing them to produce a toxic equivalency quantity (TEQ) for dioxins expressed in nanograms of TEQ per kilogram. Dioxins accumulate in the fat of foodstuffs, and the food chain represents more than 90% of the human exposure to dioxins (*3*, *4*). In some populations, seafoods represent the highest human dietary intake of dioxins (*5*, *6*). Due to their adverse health effects (*7*, *8*) and their persistence in the environment (*9*), dioxins have received increasing public attention, especially after the animal feed contamination that hit Belgium in 1999 (*10*). Additionally, bioaccumulation of these lipophilic contaminants in the fat of foods (for review see ref *11*) make it a challenge to lower their intake, not only by reducing their presence in the environment but also by working at the base of the food chain primarily to reduce their concentration in animal feed (*4*, *12*, *13*). Recent EU food safety legislation (commission directive 2003/57/EC of June 17, 2003, amending directive 2002/32/EC) motivates the need for an efficient and cost-effective method to reduce the level of dioxin in feed and food. The present EU legislation sets the limit to 1.25 ng of TEQ/kg for fishmeal and 5 ng of TEQ/kg for fish oil. Depending on the origin, i.e., the pollution of the fishing ground, and thereby the fish material used for fishmeal and fish oil production, these limits cannot always be met representing a huge economic loss for fishmeal companies.

While methods to remove dioxin from oils have been developed (*14*), no cost-effective method or processing alternatives for removing dioxins from fishmeal have been published. Methods based on the lipophilic properties of dioxins have been explored. Reduction of the fishmeal fat content either mechanically or chemically using organic solvents has so far been the most promising alternative, simultaneously removing other liposoluble toxic contaminants such as polychlorinated biphenyls (PCBs). However, this can not only result in fishmeal with altered physicochemical properties and poorer nutritional value but also necessitate modification of operating units for fishmeal producers. A recent laboratory-scale investigation indicates that extraction of the press-cake with vegetable oil could result in a reduction of almost 90% of polychlorinated contaminants, but this has not yet been fully investigated (*15*). In contrast to these methods, which propose physical removal or extraction of contaminants, we investigated the possibilities of a direct decomposition of dioxins in the fishmeal. Dioxins are photodegradable and can to a certain extent be degraded upon exposure to sunlight (*16*). The mechanisms of dioxin photodegradation are in complex matrixes rather controversial. We

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report here for the first time that fishmeal can be decontaminated with UV light treatment, resulting in break down of the dioxin contaminants. Additionally, we propose hypotheses to indicate the mechanism of degradation of dioxins in fishmeal during exposure to UV light.

MATERIALS AND METHODS

Materials. Fishmeal samples with different dioxin contents (2.80 and 3.89 ng of TEQ/kg) were obtained from fishmeal producer FF-of-Denmark, (Skagen, Denmark). Ethoxyquin (1,2 dihydro-2,2,4-trimethylquinolin-6-yl ether) liquid (90−100%) was from SvedaKemi (Frederiksberg, Denmark). UV tubes were from Philips, TL40W/12 (UVB 280−320 nm) and TL 40W/05 (UVA 320−400 nm).

UV Treatment. Fishmeal portions of 200 g were put in rectangular (280 × 180 × 60 mm) stainless steel boxes, and the depth of the fish meal sample was approximately 1 cm. The boxes were placed on a shaking table set at 280 rpm under the UV light for 2, 5, or 10 days. For each experiment a control sample was run in parallel, with a lid on the box preventing exposure of the meal to UV. The UV-light sources consisted of 2 UV tubes, which were placed at 10, 20, or 40 cm directly under the fishmeal samples. The light intensity at 40 cm for UVB tubes was measured over the whole UVA/UVB range to be 300 μW/cm^2 using a research radiometer equipped with a fiber probe (International light, Inc., Newburyport, MA).

Addition of Ethoxyquin to Fishmeal. A fishmeal portion of 100 g was placed in a 250 mL round-bottom flask, which was subsequently mounted onto a rotary evaporator equipped with a temperature-controlled water bath. Ethoxyquin was dissolved in ethanol (5 mL) and added dropwise to the rotating fishmeal portion to a final concentration of 500, 1000, or 3000 ppm. Thereafter, the system was flushed with additional ethanol (5 mL). A control sample was prepared by adding (2 × 5 mL) ethanol to the fishmeal. The fishmeal was allowed to dry for 30 min at 50 °C on the rotary evaporator prior to exposure to UV light.

Lipid Oxidation. Lipid oxidation analyses were performed after UV exposure. Lipids were extracted using chloroform and methanol according to the protocol of Bligh and Dyer, but using a reduced amount of solvent (*17*). Peroxide values were measured on the lipid extract by colorimetric determination using the ferric thiocyanate assay as described by Shantha and Decker (*18*). Volatiles were collected from the fishmeal directly after exposure to UV light by dynamic headspace sampling. The sample was purged with nitrogen at 340 mL/min for 10 min at 37 °C, and the volatiles were trapped on Tenax-GR traps (Varian Chrompack International, Bergen op Zoom, The Netherlands). Volatiles were released from the column by thermal desorption (ATD-400, Perkin-Elmer, MA) and analyzed by GC-MS (Hewlett-Packard 5890 Serie II, Palo Alto, CA) on a 30-m DB 1701 capillary column (J & W Scientific, Folsom, CA,). The temperature program used was 40 °C for 10 min, ramping 3 °C/min to 140 °C, 5 °C/min to 170 °C, and 10 °C/min to 240 °C final temperature. The ionization energy was set to 70 eV in the EI, and the scan ranged from 30 to 250 atomic mass units with repetition rate at 3.4 scans/s. For quantification, aldehyde standards were directly injected to the Tenax-GR traps and analyzed as described above for the samples.

Dioxins and PCBs Analysis. Dioxins and PCB levels of the fishmeal were assessed by Eurofins Gfa, (Münster, Germany), who determined the concentration of the toxic congeners in the fishmeal (in nanograms per kilogram) as well as the toxic equivalent quantity (in nanograms of TEQ per kilogram). Dioxin levels expressed in nanograms of TEQ per kilogram are the sum of weighted congeners relative to 2,3,7,8-PCDD. PCB levels are also expressed in nanograms of TEQ per kilogram as the sum of the 12 PCBs congeners weighted relative to 2,3,7,8-PCDD. Briefly, the method consists first in Soxhlet extraction of the fat with toluene and acetone of a representative sample amount (150 g of fishmeal). After evaporation of the solvent, the fat content was determined gravimetrically. Thereafter the sample was cleaned up by liquid/solid chromatography after addition of 16 $^{13}C_{12}$-labeled internal tetra-through octaCDD/F standards or 18 $^{13}C_{12}$-labeled internal PCB standards according to EU guidelines. Prior to the gas chromatographic analysis, two further ^{13}C-labeled PCDD/F standards or one ^{13}C-labeled PBC congener was added to the sample fraction for the determination of the recovery of internal standard. A capillary gas chromatograph (HRGC Hewlett-Packard 5890, Palo Alto, CA) coupled with a high-resolution mass spectrometer (HRMS, VG-AutoSpec, mass resolution > 8000) was used for the PCDD/F and PCB analysis. The quantitative determination of native tetra-through octaCDD/F or of native PCBs was achieved via the corresponding $^{13}C_{12}$-labeled internal standard (isotope dilution method). On the basis of the concentrations of the PCDD/F and 12 PCB congeners, TEQ values were calculated according to the WHO model.

Table 1. Dioxin Profile of Fishmeal after 5 Days Exposure to UVA or UVB[a]

	dioxin concn (ng/kg)		
PCDDs	control	UVA	UVB
2,3,7,8-tetraCDD	0.18	0.19	0.09
1,2,3,7,8-pentaCDD	0.51	0.46	0.23
1,2,3,4,7,8-hexaCDD	0.09	0.07	0.04
1,2,3,6,7,8-hexaCDD	0.34	0.34	0.14
1,2,3,7,8,9-hexaCDD	0.10	0.08	0.03
1,2,3,4,6,7,8-heptaCDD	0.33	0.28	0.17
octaCDD	0.82	0.89	0.60
ng of TEQ/kg [b]	2.80	2.52	1.02

[a] Distance from light source, 40 cm. [b] Coefficient of variation (CV), 12%.

Amino Acid Analysis. Amino acid analyses were performed by Eurofins (Kolding, Denmark) according to the method of the Association of American Cereal Chemist (*19*).

Determination of Ethoxyquin in Fishmeal. Fishmeal samples (0.5 g) were extracted twice with heptane (5 mL) using ultrasound for 10 min. Thereafter, the sample was centrifuged at 2800 rpm for 10 min, and the heptane fraction was filtered on PTFE membranes (Minisart Filters SRP15, 0.2 μm, Sartorius, Göttingen, Germany). Heptane was evaporated under nitrogen and the residue redissolved in acetonitrile prior to injection of an aliquot (50 μL) onto the HPLC (HP, Paolo Alto, CA) equipped with a Nucleosil 100-5 C18 column (HP, 4 × 125 mm, i.d. = 5 μm) and with a UV detection unit (*20*). The mobile phase consisted of acetonitrile and 1 mM ammonium acetate (80:20, respectively) at a flow rate of 0.8 mL/min. The ethoxyquin content was determined at 362 nm according to the method described by He & Ackman (*20*).

RESULTS

Effect of UV on Dioxins Degradation. Exposure of contaminated fishmeal samples (2.80 ng of TEQ/kg) to UV light for 5 days reduced the dioxin content. The dioxin profiles of fishmeal samples exposed to UV light for 5 days reveal that UVB was more efficient than UVA in decontaminating fishmeal, giving a dioxin content of 1.02 and 2.52 ng of TEQ/kg, respectively (**Table 1**). **Table 1** shows that 5 days exposure to UVB halves the concentration of all PCDDs (except for OCDD), while UVA did not seem to significantly affect any of the toxic congeners. Exposure to UVB for up to 10 days resulted in a dioxin content of 0.8 ng of TEQ/kg (**Table 2**). The value falls below the limit set by the EU legislation for fishmeal (1.25 ng of TEQ/kg); however, samples with higher dioxin contents (3.89 ng of TEQ/kg) were used in the next parts of the experimental work to determine if degradation of highly contaminated samples was also possible under our experimental conditions.

Effect of UV on Proteins and Lipids. Exposure of fishmeal to UV light did not seem to significantly affect the amino acid composition of the fishmeal, inferring preservation of its full nutritional value (**Table 3**). In contrast, UV exposure triggered lipid oxidation of the long-chain n-3 fatty acids present in the fishmeal, which are very susceptible to oxidation. Measurement of peroxide values (**Figure 1**) indicates that both UVA and UVB result in a similar development of lipid oxidation in fishmeal.

Table 2. Dioxin Profile of Fishmeal after 2, 5, and 10 Days Exposure to UVB[a]

PCDDs	dioxin concn (ng/kg) control	2 days	5 days	10 days
2,3,7,8-tetraCDD	0.18	0.20	0.10	0.10
1,2,3,7,8-pentaCDD	0.51	0.40	0.25	0.18
1,2,3,4,7,8-hexaCDD	0.09	0.04	0.06	0.04
1,2,3,6,7,8-hexaCDD	0.34	0.23	0.18	0.14
1,2,3,7,8,9-hexaCDD	0.10	0.06	0.05	0.04
1,2,3,4,6,7,8-heptaCDD	0.33	0.32	0.33	0.31
octaCDD	0.82	0.79	0.83	0.77
ng of TEQ/kg [b]	2.80	1.92	1.12	0.80

[a] Distance from the light source, 40 cm. [b] Coefficient of variation (CV), 12%

Table 3. Amino Acid Content of Fishmeal after 5 Days Exposure to UVB[a]

	control	UVB	CV (%)
crude protein (%)	74.4	74.6	0.3
amino acids (%)			
Cys + Cystine	0.6	0.6	4.5
Met	2.2	2.1	3.8
Asp	6.8	6.7	2.6
Thr	3.1	3.1	2.7
Ser	3.1	3.0	2.9
Glu	9.9	9.5	2.3
Pro	3.0	2.9	4.2
Gly	4.6	4.5	3.2
Ala	4.7	4.6	2.6
Val	4.0	3.9	3.2
Ile	3.3	3.2	4.2
Leu	5.4	5.3	3.7
Phe	2.9	2.8	2.8
His	2.2	2.0	6.9
Lys	6.0	5.8	2.9
Arg	4.4	4.3	3.0

[a] Distance from the light source, 40 cm. CV, Coefficient of variation.

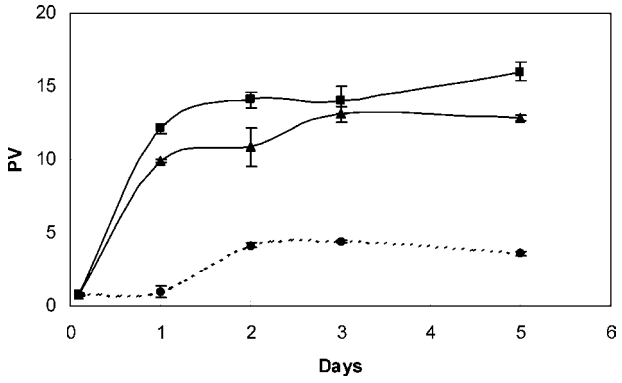


Figure 1. Development of lipid oxidation in fishmeal measured as peroxide value (PV) expressed in milliequivalent per kilogram of oil, during exposure to UVA (– ■ –) or UVB (– ▲ –). Control, no UV exposure (••• ● •••). Fish meal 2.80 ng of TEQ/kg. Distance from light source, 40 cm. Results are presented as the mean of triplicate measurements ± standard deviation.

This was further confirmed by measurement of volatiles using GC-MS (not shown). A slight increase in PV is observed in the control samples, which is probably due to exposure of the fishmeal surface to air in combination with the temperature at which the experiment is performed.

To prevent lipid oxidation and to assess whether lipid oxidation and dioxin degradation are coupled, ethoxyquin was added to fishmeal prior to UVB light exposure. Fishmeal

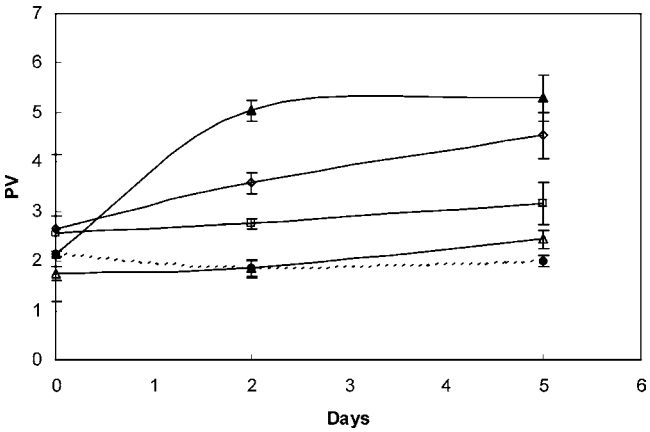


Figure 2. Development of lipid oxidation in fishmeal measured as peroxide value (PV) expressed in milliequivalent per kilogram of oil, during exposure to UVB and after addition of ethoxyquin (– ◇ –) 500 ppm, (– □ –) 1000 ppm, or (– △ –) 3000 ppm ethoxyquin or (– ▲ –) no addition of ethoxyquin. Control, no UV exposure (••• ● •••). Fish meal, 3.89 ng of TEQ/kg. Distance from light source, 40 cm. Results are presented as the mean of triplicate measurements ± standard deviation.

Table 4. Effect of Addition of Ethoxyquin (3000 ppm) on Lipid Oxidation (PV) and on the Dioxin Content of Fishmeal Exposed to UVB for 5 Days[a]

	no UVB, no ethoxyquin	+ UVB, no ethoxyquin	+ UVB, + ethoxyquin
dioxin (ng of TEQ/kg [b])	3.89	1.15	1.72
PV (milliequiv/kg)	2.14	4.37	2.19

[a] Distance from light source, 40 cm. PV: Peroxide Value. [b] Coefficient of variation (CV), 12%.

samples do initially contain 150 ppm ethoxyquin from addition during manufacturing; however, extra ethoxyquin was added at levels of 500, 1000, and 3000 ppm prior to UVB exposure. Results presented in **Figure 2** show that addition of ethoxyquin hindered lipid oxidation significantly in a concentration-dependent manner. Addition of 3000 or 1000 ppm ethoxyquin to a contaminated fishmeal sample was able to completely prevent the development of lipid hydroperoxide throughout UVB exposure, as seen in **Figure 2**. For fishmeal with 3000 ppm ethoxyquin added, the dioxin content was higher (1.72 ng of TEQ/kg) than in the sample without any ethoxyquin (1.15 ng of TEQ/kg) (**Table 4**). This indicates that dioxin degradation and lipid oxidation might be coupled processes.

Effect of Light Intensity. Light intensity correlates with the distance of the sample according to the inverse square law: $I = k/D^2$ with I = intensity, k= constant, and D = distance to the light source. The intensity at 40 cm was estimated to be 300 μW/cm^2, and the intensities at 20 and 10 cm were calculated to be 1200 and 4800 μW/cm^2, respectively. Fishmeal samples were placed 40, 20, or 10 cm under the UVB light source for 5 days. Development of volatiles was measured using GC-MS, and measurement of 1-penten-3-ol and hexanal as markers for oxidation of n-3 and n-6 fatty acids, respectively, indicate that increasing light intensity enhanced lipid oxidation significantly (**Figure 3**). Peroxide values also confirm increasing lipid oxidation with increasing light intensity (not shown). However, this was not due to an increase in temperature of the sample as the temperature was the same for each experiment and reached 19 °C (±1). Moreover, ethoxyquin degradation profile, presented in **Figure 4**, shows that ethoxyquin was degraded following the same pattern irrespective of the distance of the sample from

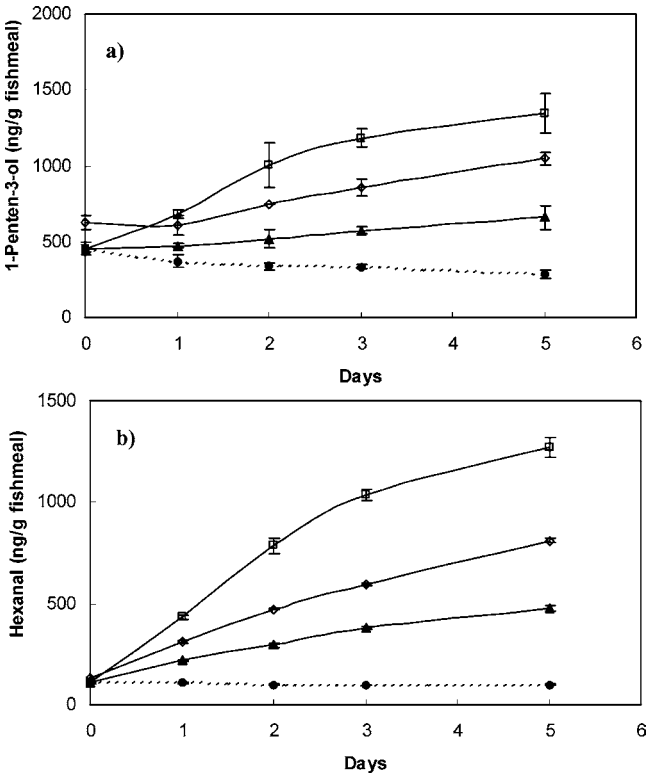


Figure 3. Development of volatiles (**a**) 1-penten-3-ol and (**b**) hexanal in fishmeal, during exposure to UVB at a distance of 40 (– ▲ –), 20 (– ◇ –), or 10 cm (– □ –) from the light source. Control, no UV exposure (••• ● •••). Fish meal, 3.89 ng of TEQ/kg. Results are presented as the mean of triplicate measurements ± standard deviation.

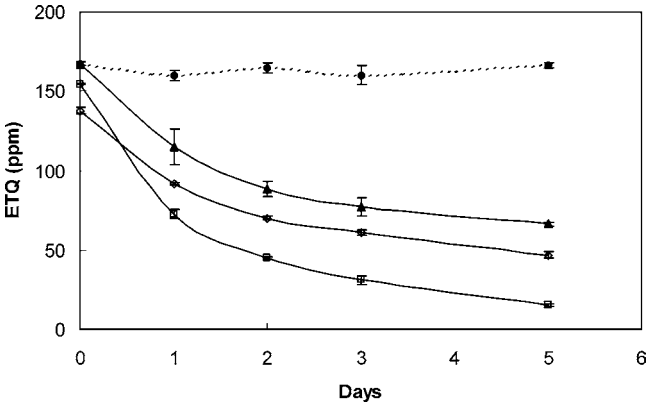


Figure 4. Degradation profile of ethoxyquin in fish meal during exposure to UVB at a distance of 40 (– ▲ –), 20 (– ◇ –), or 10 cm (– □ –) from the light source. Control, no UV exposure (••• ● •••). Fish meal, 2.80 TEQ. Results are presented as the mean of triplicate measurements ± standard deviation.

the light source, but that decay was more pronounced at 10 cm when compared to 40 cm. Dioxin profiles of the different samples are presented in **Table 5** and reveal that an increase in light intensity by decreasing the distance of the sample to the light source successfully reduced the dioxin content of the fishmeal sample considerably. TCDD was reduced by a factor of approximately 6.7 corresponding to a degradation of 85% of the TCDD congener, when the fishmeal was placed at 10 cm from the light source (corresponding to a light intensity of 4800 μW/cm^2). The dioxin value of 0.42 TEQ obtained for the sample placed at 10 cm from the light source also indicates that it is possible to go much below 1 ng of TEQ/kg using UV treatment.

Table 5. Dioxin Content of Fishmeal after 5 Days Exposure to UVB at a Distance of 40, 20, or 10 cm from the Light Source

	dioxin concn (ng/kg)			
PCDDs	control	40 cm	20 cm	10 cm
2,3,7,8-tetraCDD	0.27	0.10	0.08	0.04
1,2,3,7,8-pentaCDD	0.67	0.27	0.20	0.09
1,2,3,4,7,8-hexaCDD	0.11	0.06	<0.05	<0.05
1,2,3,6,7,8-hexaCDD	0.46	0.21	0.12	0.06
1,2,3,7,8,9-hexaCDD	0.18	0.08	<0.05	<0.05
1,2,3,4,6,7,8-heptaCDD	<0.46	<0.31	<0.33	<0.33
octaCDD	<1.16	<0.83	<0.83	<0.83
ng of TEQ/kg [a]	3.89	1.15	0.80	0.42

[a] Coefficient of variation (CV), 12%.

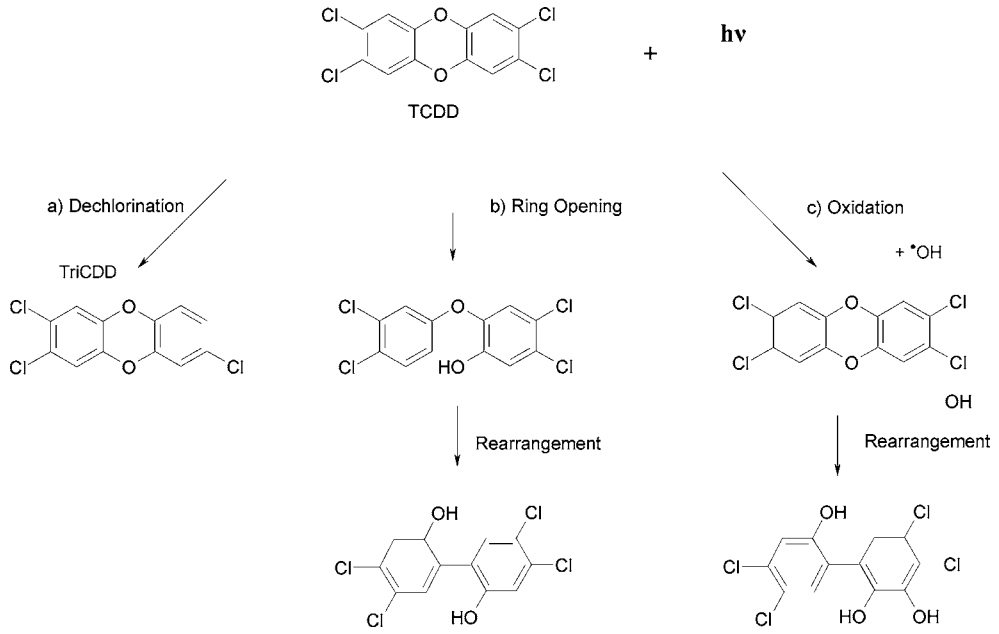
Table 6. Furans and PCBs Content Profile of Fishmeal after 5 Days Exposure to UVB[a]

	furan or PCB concn (ng/kg)	
PCDFs	control	5 days UVB
2,3,7,8-tetraCDF	4.69	2.69
1,2,3,7,8-pentaCDF	1.13	0.62
2,3,4,7,8-pentaCDF	4.38	1.87
1,2,3,4,7,8-hexaCDF	0.44	0.12
1,2,3,6,7,8-hexaCDF	0.47	0.22
1,2,3,7,8,9-hexaCDF	<0.01	<0.01
2,3,4,6,7,8-hexaCDF	0.45	0.22
1,2,3,4,6,7,8-heptaCDF	0.16	<0.09
1,2,3,4,7,8,9-heptaCDF	<0.1	<0.09
octa-CDF	<0.25	<0.24
ng of TEQ/kg [b]	3.89	1.87
non-ortho PCBs		
PCB 77	96.9	689
PCB 81	<1.6	<7.02
PCB 126	26.5	98.3
PCB 169	5.14	6.59
mono-ortho PCBs		
PCB 105	1460	1310
PCB 114	52.9	53.7
PCB 118	4170	3670
PCB 123	45.2	44
PCB 158	518	437
PCB 157	118	109
PCB 167	325	324
PCB 189	41.4	40.6
ng of TEQ/kg [b]	3.63	10.8

[a] Distance from the light source, 40 cm. [b] Coefficient of variation (CV), 12%.

Effect of UV on PCBs. To investigate if UV treatment could simultaneously degrade furans and PCBs present in fishmeal, both PCDF and PCBs were measured before and after exposure of the fishmeal sample to UVB for 5 days and at a distance of 40 cm from the light source. Results presented in **Table 6** show that PCDF are also degraded by UV light exposure to the same extent as PCDD. Surprisingly, the total TEQ for PCBs increases after exposure of the fishmeal to UVB and was approximately multiplied by a factor of 3; however, some congeners are degraded while others seemed to accumulate (**Table 6**). The congeners that increased significantly are PCB 77 and PCB 126; therefore further studies are needed to validate the mechanism by which these congeners are formed under the present conditions. To date, there are no European legislations for PCBs in fishmeal and fish oil, however, these are imminent.

Scheme 1. Proposed Degradation Pathways for TCDD in Fishmeal Exposed to UVB Light with (A) Reductive Dechlorination, (B) Ring Opening via C–O Cleavage, and (C) Oxidation via, for Example, Hydroxyl Radical Attack



DISCUSSION

Dioxins, Protein, and Lipids. The most important mechanism for dioxin degradation in the environment is believed to be degradation by the UV component of sunlight (*16*). Many studies report photolytic decomposition of dioxins in water (*21*, *22*), in organic solutions (*23*, *24*), in gases (*25*), and also in soils and sediments (*26*, *27*). Here we report that UVB light can be used to selectively remove dioxin from fishmeal. The exposure time in our study was 5 days, but it is clear that for any industrial relevance a much shorter exposure time should be adopted. The UVB exposure did not seem to affect the amino acid composition and the protein quality of the fishmeal. However, fishmeal contains a high level of unsaturated fat, which under UV light exposure oxidized and resulted in high peroxide values. This is not by itself necessarily a problem because to date there are no limits to lipid oxidation levels in fishmeal. This lack of a maximum value for oxidation in fishmeal is due to the difficulty of setting up universal methods but also due to large variation between different fishmeal samples (*28*).

Dioxin degradation and lipid oxidation are not necessarily linked, but it is obvious that both dioxins and lipids are photodegradable under UV light. Photooxidation of lipids under UV light by activation of oxygen and generation of singlet oxygen, which can then directly react with lipid, is very likely to occur. The results indicate that the two reactions, i.e., dioxin degradation and lipid oxidation, might be linked as prevention of oxidation by ethoxyquin also slowed dioxin degradation. Indeed, reactive oxygen species such as $^1O_2$, $HO^\bullet$, and H_2O_2 generated during exposure of fishmeal to UV might also contribute to the degradation of dioxins as suggested by others (*29*). It is possible that reactive oxygen species formed during UV exposure interact with the fishmeal inducing oxidation. Fenton's reagent has been used with success to degrade TCDD to biodegradable intermediate, indicating that oxidative reactions can also result in detoxification of dioxins (*30*). If oxygen is a limiting factor, oxidation of lipid is likely to be hindered. Using nitrogen atmosphere optimal dioxin degradation might be achieved avoiding lipid oxidation.

To protect fishmeal from oxidation, the manufacturer usually introduces an antioxidant, ethoxyquin, during the production. Legislation in Denmark only allows 150 ppm ethoxyquin, while in South American meals it can reach 1000 ppm. This high level of ethoxyquin present is to avoid the possible problem of spontaneous ignition of fishmeal during transport caused by the high level of unsaturated fat. This high level of ethoxyquin is enough to protect fishmeal from oxidative reaction upon exposure to UVB without compromising the decomposition of dioxins (**Table 4**). Ethoxyquin is also consumed during UVB exposure, and in the sample without addition of ethoxyquin only approximately 20% ethoxyquin (40 ppm) is left after 5 days UVB exposure, while in the sample with 3000 ppm added approximately 80% remained after exposure (2400 ppm). Ethoxyquin oxidation products also possess some antioxidative activity, and ethoxyquin is an effective and low-cost antioxidant (*31*). Nevertheless, reports indicate that ethoxyquin is toxic (*32*); therefore other types and concentrations of antioxidant should be optimized for UV exposure of fishmeal. Addition of other types of antioxidants, for example capable of scavenging singlet oxygen such as carotenoids, should be investigated, and synergistic studies should be performed.

Light Intensity. The light intensity had a significant impact on the degradation of dioxin in fishmeal. Therefore, a higher intensity lamp would shorten the exposure time, and this would be a much more realistic setting for industries that deal with large-scale production of fishmeal, and where time is a cost factor. However, oxidation of unsaturated fatty acids still remains the main drawback as increasing light intensity also resulted in more pronounced development of oxidation of unsaturated fatty acids.

Degradation Pathways. Scheme 1 illustrates possible dioxin decontamination pathways in fishmeal. The mechanism of photodegradation of PCDD has been elucidated in water and organic solvent but not fully in more complex matrixes such as soil and food. Accumulation of dechlorinated products with accumulation of the toxic TCDD has been reported as direct evidence for dehalogenation in soil (*26*). In the present experiments no accumulation of TCDD was observed; in contrast all

low chlorinated congeners decayed by 40−50% after 5 days, while OCDD and HeptaCDD were not degraded (**Table 2**). A recent study indicates that photoproducts of 2,3,7,8-TCDD in solution include dechlorinated congeners and chlorinated dihydroxybiphenyls, which are also polychlorinated biphenyls (*33*). Here, we suggest that the presence of lipids, protein, and photosensitizer (i.e. chlorophyls, cytochrome residues) in fishmeal also enhance the degradation of dioxin in fishmeal exposed to UV light. Reactive oxygen species generated during exposure to UV light are likely to degrade dioxins as for example reported for hydroxyl radicals (*27, 34*). In natural water, proteins were found to have a strong sensitizing effect on the photolysis of organochlorine compounds (*35*). Dung and O'Keefe (*36*) have reported that in soils humic acids could be excited by UV light and transfer energy to other molecules generating reactive intermediates, which may react rapidly with dioxins. In addition, a study performed on soils indicates that substances favoring the transport of PCDD at the soil surface increase the photodegradation (*37*). This idea has been recently explored, and olive oil was used to extract PCB and PCDD from soil prior to UV degradation, to overcome the shielding effect of soil particles (*38*). It is likely that in fishmeal the fish oil present is playing this role by transporting the PCDD to the irradiated surface, therefore increasing the efficacy of UV light treatment.

PCBs are photodegradable and are therefore also decomposed by UV light (**Table 6**). An increase in congeners PCB 77 and PCB 126 was observed resulting in a total increase in toxic equivalency quantity. It is clear that further studies are needed to confirm the formation of these congeners via for example dechlorination of higher chlorinated congeners.

The dominant degradation pathway for dioxins and PCBs undergoing photolysis in fishmeal remains unresolved due to the complexity of the matrixes but also probably due to the fact that (1) the degradation mechanism involves more than one reaction pathway, (2) the degradation pathways are different for different congeners and strongly influenced by the environment, and (3) products generated during photodegradation are unstable, rearrange, polymerize, or further oxidized or photodegraded as also proposed by others (*36*).

Conclusions. It is likely that in a complex matrix such as fishmeal, more than one degradation pathway is effectively contributing to the degradation of dioxins. In our study no photoproducts were isolated or identified and the mechanisms put forward need further investigation. Additionally, to be applied at an industrial scale, the light intensity should be increased facilitating shorter exposure times. The question of oxidation of long-chain fatty acids should be tackled by, for example, addition of selected antioxidants or use of modified atmosphere.

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