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Impact of Diuron Antifouling Paint on Fatty Acid Composition in Asian Sea-Bass "*Lates calcarifer*"

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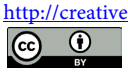

Abstract

In recent decades, the application of antifouling paints to watercrafts has emerged as a significant concern in coastal ecosystems, including coral reefs. Diuron, a booster biocide commonly used as an antifouling agent, is persistent in soil, water, and groundwater. Although it exhibits moderate toxicity to mammals and birds, its primary breakdown product, 3,4-dichloroaniline, is highly toxic and environmentally persistent. The secondary toxicity of Diuron emphasizes its potential to contaminate groundwater. This investigation determined the 96-hour LC50 value for Diuron to be 1.627 ± 0.181 mg/L in *Lates calcarifer*. A 21-day sublethal exposure of *Lates calcarifer* to diuron did not yield statistically significant differences ($P > 0.05$) in fatty acid composition between the fresh and control groups. Both groups demonstrated polyunsaturated fatty acids (PUFAs) as the most abundant fatty acids, followed by saturated fatty acids (SFAs) and monounsaturated fatty acids (MUFAs). However, significant variations ($P < 0.05$) were observed in the fatty acid composition of *Lates calcarifer* exposed to varying diuron concentrations (50%, 30%, and 10% of the LC50 value). The magnitude of these effects increased with increasing exposure levels. The results of this study indicate that diuron, even at concentrations considered safe, can adversely affect marine organisms, including alterations to their fatty acid composition.

Keywords

Diuron, Booster Biocide, Antifouling Paint, Fatty Acid Composition, *Lates calcarifer*

1. Introduction

The International Maritime Organization (IMO) has restricted the utilization of organotin compounds (OTs) in antifouling paints for marine vessels. As a result, biocide compounds have been introduced as alternative antifouling agents (Ali et al., 2021; Hanapiah et al., 2017). Booster biocides, characterized by their rapid degradation and low persistence, are employed to enhance paint efficacy by inhibiting the growth of fouling organisms (Mukhtar et al., 2019; Voulvoulis et al., 2002; Terlizzi et al., 2001; Thomas et al., 2002). In Japan, 18 substances, including Diuron, have been proposed as safe and effective antifouling agents (Voulvoulis et al., 2002; Thomas et al., 2002; Boxall et al., 2000; Yonehara, 2000). While these biocides are hypothesized to be less detrimental to the environment than organotin compounds, their ecological impact remains uncertain (Myers et al., 2006). This lack of understanding is attributed to their recent introduction, limited use, and the perception that they are less toxic than tributyltin (TBT) (Terlizzi et al., 2001; Myers et al., 2006; Evans et al., 2000; Maraldo & Dahllöf, 2004). The majority of pollution research in Malaysia has focused on heavy metals in ports, marinas, and islands. However, studies have detected the presence of Diuron in coastal waters (Ali et al., 2014). Despite this finding, there remains a significant knowledge gap regarding the toxicity of new antifouling chemicals, including diuron, on the health of coral reef organisms and fish globally. This investigation aimed to examine the impact of Diuron on *Lates calcarifer* (Asian sea bass) through short-term and acute exposure experiments, with a focus on alterations in fatty acid composition. *Lates calcarifer* is an economically significant species in the tropical and subtropical regions of Asia and the Pacific Ocean. It is commercially cultivated in various countries and areas, including Malaysia, Thailand, Indonesia, Hong Kong SAR, Taiwan area, and Australia, in brackish water ponds and coastal water cages (Thophon et al., 2003). This species is susceptible to diuron exposure, as the chemical has been identified in the region's coastal waters (Ali et al., 2021; Ali et al., 2014). Fatty acid composition is indicative of specific physiological functions and states in organisms (Sargent et al., 2002). Analysis of lipid content and fatty acid composition in organisms, such as corals, can serve as a potential health diagnostic indicator (Bachok et al., 2006; Ali et al., 2015). In addition to being a primary energy source (Sargent et al., 2002; Sargent et al., 1999; Froyland et al., 2000; Tocher, 2003), fatty acids, particularly polyunsaturated fatty acids (PUFAs), are crucial for normal fish growth, development, and reproduction (Sargent et al., 2002; Sargent et al., 1999; Yanes-Roca et al., 2009).

2. Materials and Methods

Juvenile *Lates calcarifer* specimens, approximately two months old, were obtained from a commercial fish farm in Setiu Province during April and May 2012. These specimens underwent an acclimation period in well-oxygenated 500-liter polyethylene tanks. The tanks contained natural seawater maintained at a salinity of 30 ppt and were exposed to a natural 12-hour light-dark cycle. The water in the tanks

was filtered to remove particles larger than 1 mm, sterilized with UV light, and replenished daily. The specimens were fed a standard commercial diet twice daily. A 24-h fasting period was implemented both before and during the chronic-exposure experiment. A stock solution of diuron, at a concentration of 1000 mg/L, was prepared using acetone. Working solutions were prepared by adding the necessary amounts of this stock solution to seawater.

To assess the 96-hour LC50 of Diuron for *Lates calcarifer*, researchers conducted a static bioassay test following established protocols (Finney, 1971). Juvenile specimens, approximately two months of age, with a mean length of 6.29 ± 0.37 cm and mass of 7.96 ± 0.56 g, were acclimated for two weeks prior to random distribution into plastic aquariums (51 cm × 25 cm × 30 cm) containing 30 liters of water. Each aquarium contained ten specimens, exposed to Diuron concentrations ranging from 1 to 7 mg/L, with three replicates per concentration. The control group was maintained under similar conditions, without Diuron exposure. The daily monitoring of water quality parameters included dissolved oxygen, temperature, and pH. Water samples were collected at the beginning and conclusion of the Diuron analysis. Specimen mortality was evaluated every 24 h, with death defined as the absence of gill movement and unresponsiveness to gentle probing. The deceased specimens were promptly removed. Mortality percentages were documented at 24, 48, 72, and 96 h for each Diuron concentration, and the 96-hour LC50 values were determined using Probit analysis (Finney, 1971). The specimens were not fed during the experiment.

For the sub-chronic exposure phase, specimens were exposed to 10%, 30%, and 50% of the 96-hour LC_{50} values of Diuron, as determined from the acute toxicity tests. The experimental setup was similar to that of the acute exposure, but the duration was extended to 21 days. Each 30-liter aquarium contained natural seawater with 30 ppt salinity and was equipped with a continuous water flow system, filtering water through a biofilter at 4 L/min. Constant aeration was maintained throughout the experiments. Three replicate aquariums were used for each test concentration and the control group. The specimens were fed twice daily during the 21-day exposure. Weekly water changes of 50% were performed to maintain optimal water quality following established guidelines (APHA et al., 1995). Daily monitoring of water quality parameters included oxygen content, temperature, pH level, and water salinity. NH_3-N, NO_2-N, and NO_3-N levels were measured twice weekly, with ammonia and nitrite nitrogen levels strictly maintained below 0.2 mg/L. All chemical parameters were analyzed using analytical-grade reagents and established techniques (APHA et al., 1995). Weekly measurements of the Diuron concentration in the water were measured before and after chemical addition to ensure that the desired nominal level was maintained. **Table 1** summarizes the characteristics of the water quality. Daily observations of mortality and behavioral changes were conducted for each treatment group. At the conclusion of the 21-day exposure, one specimen from each aquarium was sampled for fatty acid analysis in the laboratory.

Table 1. Characteristics of water quality during sub-chronic exposure experiment.

Name	Interval	Average ± SD
Oxygen content (mg/L)	6.2 - 7.4	6.9 ± 0.3
Temperature (˚C)	27.9 - 29.8	28.9 ± 0.66
Salinity (%)	29.3 - 31.2	30.14 ± 0.65
pH level	6.89 - 8.04	7.76 ± 0.29
NH_3-N (mg/L)	0.02 - 0.28	0.19 ± 0.12
NO_2-N (mg/L)	0.03 - 1.25	0.07 ± 0.33
NO_3-N (mg/L)	0.96 - 3.11	2.16 ± 0.44

3. Analytical Method

For fatty acid composition analysis, liver samples were obtained. A combined extraction and esterification technique in a single vessel was utilized (Ali et al., 2015; Abdulkadir & Tsuchiya, 2008). Three duplicate liver tissue samples (200 - 300 mg) were combined with 4 mL of hexane and 1 mL of an internal standard solution in a 50 mL centrifuge tube. Subsequently, 2 mL of 14% boron trifluoride in methanol and a magnetic stir bar were introduced. Nitrogen gas was employed to flush the tube's headspace before sealing it tightly with a Teflon-lined cap. The sealed container was heated to 100˚C for 120 min while being continuously stirred. After cooling to ambient temperature, 1 mL hexane and 2 mL distilled water were added. The tube underwent vigorous agitation for one minute and centrifugation at 2500 rpm (650 × g) for three minutes. Using a Pasteur pipette, the upper hexane layer containing fatty acid methyl esters (FAMEs) was carefully transferred to a clean sample vial for subsequent gas chromatography-flame ionization detection (GC-FID) analysis. Fatty acid concentrations (CFA, mg/g dry weight) were determined by comparing each fatty acid's peak area in the sample to the internal standard's peak area using the following equation (Ali et al., 2015; Abdulkadir & Tsuchiya, 2008):

$$C_{FA} = A_S / A_{IS} \times C_{IS} / W_S$$

where;

A_S = peak area of fatty acid in the sample in chromatogram

A_{IS} = peak area of internal standard in chromatogram

C_{IS} = concentration of internal standard (mg)

W_S = weight of sample (g)

The percentage composition of individual fatty acids was determined by comparing each fatty acid's peak area to the total peak area of all fatty acids in the sample as explained by (Ali et al., 2015; Abdulkadir & Tsuchiya, 2008).

4. Gas Chromatography

A Shimadzu GC-14B gas chromatograph with a flame ionization detector utilized

to separate and quantify fatty acid methyl esters (FAMEs). The separation was conducted using a 30-meter FFAP-polar capillary column (0.32 mm internal diameter, 0.25 μm film thickness). Hydrogen served as the carrier gas. The oven temperature program commenced at 60˚C, increased at 40˚C/min to 150˚C, then at 3˚C /min to 230˚C, and was maintained at 230˚C for 30 min. The flame ionization detector maintained at 240˚C. FAME peaks were identified by retention time comparison with authentic standards from Supelco, Inc. Fatty acids were denoted using the n:pωx system, where "n" represents the carbon atom count in the aliphatic chain, "p" represents the double bond count, and "x" indicates the position of the first double bond from the terminal methyl group. The analytical precision for total amount and major components of FAMEs in samples was typically below 5%.

5. Results

The Probit analysis method (Finney, 1971) was employed to calculate 96-hour LC50 values. To determine the lowest concentration of a substance that causes observable effects on organisms, the mortality data from acute toxicity tests were analyzed. Moreover, fatty acid levels were measured and average values and variability were calculated. To compare the effects of different treatments, the statistical tests like one-way ANOVA and two-way ANOVA were used, followed by post-hoc tests like LSD and Duncan's multiple range test ($P < 0.05$). These tests helped to identify significant differences between treatment groups and understand how different factors, such as chemical type, concentration, and species, interact to influence the observed effects.

6. Visual Observation

Acute Exposure: Control fish exhibited normal swimming behavior throughout the 96-hour test period without any signs of abnormality. In contrast, treated fish displayed abnormal swimming patterns, including erratic movements and excessive mucus secretion in the opercula. Some fish exhibited hyperventilation after 48 h of exposure, which intensified within the 96-hour exposure period, ultimately resulting in mortality. Table 2 presents the percentage mortality rates for diuron-exposed fish at 24, 48, 72, and 96 h at various concentrations. Table 3 presents the calculated 96-hour LC50 values (with 95% confidence intervals) for Diuron, determined using different methods. No mortality observed in the control or acetone-treated (check) tanks. As indicated in Table 3, the 96-hour LC_{50} value for diuron, using *Lates calcarifer* as the test organism, was determined to be 1.627 ± 0.031 mg/L.

Extended Exposure Study: For duration of 21 days, fish were exposed to sublethal concentrations of Diuron at 50% (0.81 mg/L), 30% (0.49 mg/L), and 10% (0.16 mg/L) of the 96-hour LC_{50} value (1.627 ± 0.031 mg/L) determined from acute toxicity experiments. Throughout this extended exposure period, the control fish exhibited normal swimming behavior without any observable anomalies.

Table 2. The cumulative percentage mortality data of *Lates calcarifer* (n = 10) in acute exposure experiment.

Concentration (ppm)	1st replicate (% Mortality)				2nd replicate (% Mortality)				3rd replicate (% Mortality)			
	24 h	48 h	72 h	96 h	24 h	48 h	72 h	96 h	24 h	48 h	72 h	96 h
0 (control)	0	0	0	0	0	0	0	0	0	0	0	0
0 (check/acetone)	0	0	0	0	0	0	0	0	0	0	0	0
1	0	0	0	30	0	0	10	40	0	0	0	30
2	0	0	40	50	0	0	20	60	0	0	40	60
3	0	20	60	80	0	20	40	60	0	10	50	70
5	0	30	80	90	10	30	50	80	10	40	60	90
7	10	70	100	100	10	70	100	100	20	80	100	100

Table 3. The calculated LC_{50} (95% C.I) value of Diuron from different method.

Method	1st replicate LC_{50}	2nd replicate LC_{50}	3rd replicate LC_{50}	Mean	SD
BINOMIAL	2.000	1.414	1.592	1.669	0.300
MAA	1.754	1.414	1.626	1.598	0.172
PROBIT	1.676	1.563	1.647	1.629	0.059
SPEARMAN	1.799	1.414	1.622	1.612	0.193
MEAN AVERAGE				1.627	0.031

Table 4. Fatty Acid Profile (mg/g dry weight, liver sample) of *Lates calcarifer* (Siakap) after 3-week exposure.

FATTY ACIDS	FRESH	CONTROL	10% LC_{50} DIURON	30% LC_{50} DIURON	50% LC_{50} DIURON
SAFA					
C14:0	4.66ab ± 0.36	4.96a ± 1.46	4.18ab ± 1.35	4.17ab ± 1.91	1.78cd ± 1.55
C16:0	35.9a ± 11.6	30.9ab ± 7.31	27.0ab ± 9.84	27.1ab ± 12.2	18.8bc ± 4.30
C18:0	12.8a ± 1.56	13.3a ± 3.22	11.0ab ± 4.35	10.3abc ± 4.67	9.34abc ± 0.95
C20:0	0.00	1.19a ± 0.60	0.00	0.00	0.00
MUFA					
C16:1	3.85bc ± 0.82	6.68a ± 2.33	5.58ab ± 1.91	4.80abc ± 2.07	2.20c ± 1.92
C17:1	1.72a ± 0.38	0.00	0.00	0.00	0.00
C18:1ω9c	34.6ab ± 0.82	37.1a ± 11.5	31.2abc ± 11.8	27.4abc ± 13.3	17.4c ± 9.26
C18:1ω9t	8.13b ± 0.17	0.00	0.00	0.00	0.00
C20:1	1.32bc ± 1.15	4.34a ± 1.94	1.59abc ± 0.15	2.80abc ± 2.70	0.00
PUFA					
C18:3ω6	28.7a ± 0.80	24.9ab ± 7.03	19.3bc ± 7.54	17.8bc ± 8.08	11.9cd ± 6.36
C18:3ω3	9.25a ± 1.34	8.44a ± 6.96	9.95a ± 3.13	8.83a ± 3.38	10.1a ± 3.05
C20:3ω3	6.48a ± 2.50	5.37a ± 1.93	1.06b ± 0.93	1.11b ± 0.98	1.69b ± 0.35
C20:5ω3	7.73a ± 0.02	6.33a ± 0.20	3.84cd ± 1.42	3.65cd ± 1.64	2.61d ± 1.44
C22:6ω3	6.40ab ± 1.70	8.56a ± 1.97	6.61ab ± 2.88	5.56ab ± 0.46	8.07a ± 1.36

Continued

ΣSAFA	53.4a ± 13.4	50.4ab ± 13.2	42.2ab ± 11.9	41.6ab ± 11.9	30.0bc ± 8.62
ΣMUFA	49.6a ± 14.0	48.1a ± 15.6	38.4ab ± 13.4	35.0ab ± 11.6	19.6bc ± 7.62
ΣPUFA	58.5a ± 6.76	53.6ab ± 9.23	40.8bc ± 7.05	37.0bcd ± 6.58	34.4bcd ± 4.93
ΣFA	161a ± 11.8	152ab ± 12.0	121ab ± 10.2	114bc ± 9.48	83.9bcd ± 6.88

Note: The average value and variability of the data are shown, calculated from samples (n = 3). SAFA, MUFA, and PUFA refer to saturated, monounsaturated, and polyunsaturated fatty acids, respectively. The average values within each row marked with the same superscript (s) are not statistically different (P > 0.05).

Fish exposed to diuron maintained regular swimming patterns and displayed no indications of atypical behavior. Neither the control group nor the treated groups experienced mortality during the 21-day study period.

Lipid Analysis: Subsequent to the extended exposure test on *Lates calcarifer*, a single specimen from each replicate tank underwent fatty acid composition analysis in the laboratory. Table 4 presents the results of this analysis.

7. Discussion

7.1. Toxicity Testing

This investigation established a 96-hour LC_{50} of 1.627 ± 0.031 mg/L for Diuron in *Lates calcarifer*. Studies on the toxicity of diuron in other species are limited. For instance, Bao et al. (2011) reported a 96-hour LC_{50} of 110 μg/L for Diuron in Synechococcus species, which is substantially higher than the 72-hour EC_{50} of 0.55 μg/L reported by Devilla et al. (2005). The findings from this study exceed the maximum permissible concentration (MPC) of 430 ng/L for diuron, as determined by Giacomazzi & Cochet (2004). Diuron, known to inhibit photosystem II (Jones & Kerswell, 2003) is particularly toxic to autotrophic aquatic organisms including cyanobacteria, algae, macrophytes, and symbiotic dinoflagellates in corals. These organisms exhibited acute EC_{50} values, ranging from ng/L to μg/L. In contrast, Diuron toxicity is lower for crustaceans and fish, with acute LC_{50} values typically in the mg/L range (Zhang et al., 2008; Hall Jr. et al., 1999). The results of the current study are consistent with these observations. Diuron can disrupt photosynthetic processes in coral symbiotic dinoflagellates at concentrations below 1 μg/L (Jones & Kerswell, 2003). Consequently, at detectable levels above 1 μg/L, Diuron can significantly affect microalgal and coral growth in marine ecosystems. These findings corroborate the conclusions of (Jones & Kerswell, 2003; Chesworth et al., 2004), indicating the general toxicity of Diuron to various test organisms.

7.2. Fatty Acid Composition

Table 4 presents the mean concentrations and standard deviations of saturated fatty acids (SAFAs), monounsaturated fatty acids (MUFAs), and polyunsaturated fatty acids (PUFAs) in liver samples from Lates calcarifer. PUFAs were the predominant fatty acid group in both fresh and control fish, followed by SAFAs and MUFAs. Specifically, total PUFA content was 58.5 ± 6.76 mg/g dry weight (36.2%)

and 53.6 ± 9.23 mg/g dry weight (35.2%) in fresh and control fish, respectively. SAFA levels were 53.4 ± 13.4 mg/g dry weight (33.0%) and 50.4 ± 13.2 mg/g dry weight (33.1%), while MUFA levels were 49.6 ± 14.0 mg/g dry weight (30.7%) and 48.1 ± 15.6 mg/g dry weight (31.6%) in fresh and control fish, respectively. Among the three exposure groups (10%, 30%, and 50% of the 96-hour LC_{50}), only the 50% group demonstrated a similar pattern to the control group. The 30% and 10% groups exhibited different trends, with SAFAs being the most abundant, followed by PUFAs and MUFAs. Notably, in all exposed groups of *Lates calcarifer* (**Figure 1**), MUFAs contributed the least to the overall fatty acid composition.

7.3. Saturated Fatty Acids (SAFA)

Lates calcarifer exhibited elevated levels of saturated fatty acids (SAFAs), with no statistically significant difference ($P > 0.05$) between the fresh and control groups. However, significant variations ($P < 0.05$) were observed among the other exposure groups and individual SAFAs in *Lates calcarifer* (**Table 4**). The predominant SAFA in both fresh (35.9 ± 11.6 mg/g, 67.2%) and control (30.9 ± 7.31 mg/g, 61.3%) liver samples of *Lates calcarifer* was palmitic acid (C16:0) (**Table 4**). Analogous results have been reported in freshwater fish species, including channel catfish (*Ictalurus punctatus*) and rainbow trout (*Oncorhynchus mykiss*), where palmitic acid was also the principal SAFA, constituting 19.2% and 21.3% of the total SAFAs, respectively (Sathivel et al., 2002; İbrahim Haliloğlu et al., 2004). Previous research has also documented the prevalence of SAFAs in freshwater species (Jabeen & Chaudhry, 2011).

7.4. Monounsaturated Fatty Acids (MUFA)

As illustrated in **Table 4**, the total monounsaturated fatty acid (MUFA) content did not differ significantly between the fresh (49.6 ± 14.0, 30.7%) and control (48.1 ± 15.6, 31.6%) Lates calcarifer groups ($P > 0.05$). Oleic acid (C18:1ω-9c) was the most abundant MUFA and was significantly higher ($P < 0.001$) across all tested groups in this study (**Table 4**). Previous studies have indicated that marine fish contain lower levels of oleic acid than freshwater fish do. For instance, Ho & Paul, (2009) reported that Asian sea bass contains only approximately 8% oleic acid in its total fatty acid composition. In contrast, research has demonstrated that freshwater tilapia exhibits notably higher levels of oleic acid compared to marine fish, such as flat sardine and sea bream (Steiner-Asiedu et al., 1991). Furthermore, American freshwater channel catfish were found to have a high oleic acid content of approximately 50% in their flesh, which is substantially higher than the less than 1% found in sardine and sea mullet (Ackman, 1994; Nettleton et al., 1990).

7.5. Polyunsaturated Fatty Acids (PUFA)

The overall PUFA content in *Lates calcarifer* showed no significant variation among the fresh, control, and acetone-treated groups ($P > 0.05$). Nevertheless, PUFAs were the most prevalent fatty acids in most experimental groups (**Figure 1**). In fresh

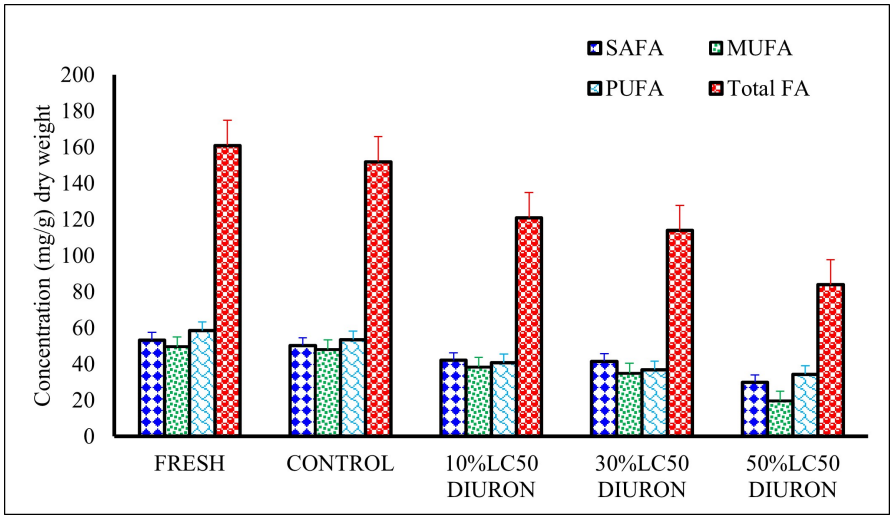

Figure 1. Levels of fatty acids and total fat (mg/g of dry liver tissue) in *Lates calcarifer* fish exposed to different amounts of diuron. Data is the average ± standard deviation, with three replicates per treatment.

samples, γ-linolenic acid (C18:3ω6) was the dominant PUFA (49.1%), with α-linolenic acid (C18:3ω3), followed by 15.8% in *Lates calcarifer* (**Figure 2**). Other fatty acids, including cis-11,14,17-eicosatrienoic acid (C20:3ω3), cis-5,8,11,14,17-eicosapentaenoic acid (C20:5ω3), and docosahexaenoic acid (DHA, C22:6ω3), also contributed substantially to the total PUFA content of *Lates calcarifer* (**Table 4**). High omega-3 and omega-6 fatty acid levels in this species are expected because marine organisms typically acquire these fatty acids from oceanic plankton (Steffens, 1997) or fishmeal diets containing them (Yanes-Roca et al., 2009; Henderson, 1996). The impact of diuron on the organisms was evident in the PUFA profiles of the exposed groups (**Figure 2**). A clear inverse relationship was observed between the Diuron dose and the activity levels of the exposed organisms. Higher Diuron concentrations corresponded to decreased activity levels in the exposed organisms.

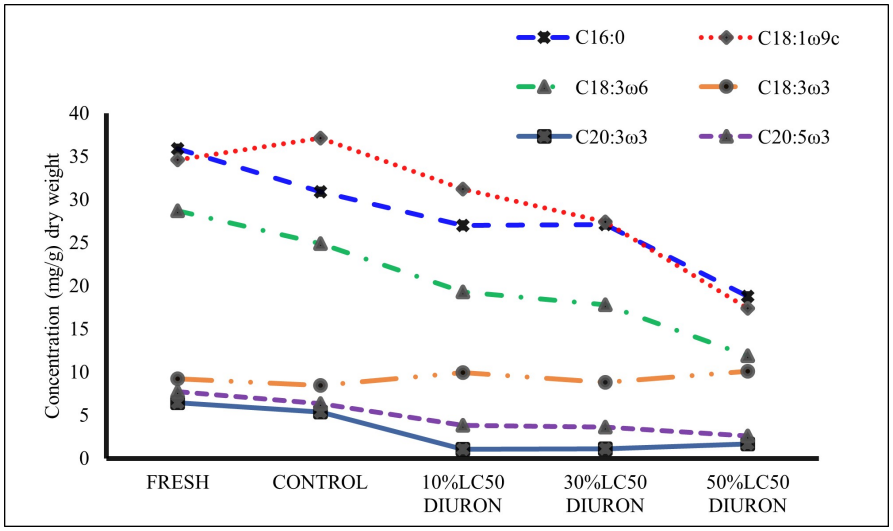

Figure 2. *Lates calcarifer* fatty acid concentrations following exposure to various diuron doses.

8. Conclusion

Laboratory experiments were conducted to examine the toxicological impact of Diuron on *Lates calcarifer*. The investigation yielded several significant findings. During acute toxicity of *Lates calcarifer*, the 96-hour LC_{50} of Diuron was determined to be 1.627 ± 0.181 mg/L. Moreover, upon exposure of *Lates calcarifer* to sub-lethal Diuron concentrations for 21 days, notable alterations were observed in the fish's fatty acid profile. On top of that, Polyunsaturated fatty acids (PUFAs) were found to be predominant in fresh and control *Lates calcarifer* samples, followed by saturated fatty acids (SAFAs) and monounsaturated fatty acids (MUFAs). In addition to that, all tested Diuron concentrations adversely affected the overall health and behavior of the exposed fish, as evidenced by their movement patterns. These effects intensified with increasing exposure concentrations. Concisely the investigation revealed that Diuron can exert toxic effects on *Lates calcarifer* even at low exposure levels (10% and 30% of the 96-hour LC50), which are typically considered non-hazardous. Based on the current findings, the following are recommendations:

1. Researchers should increase investigations on short and long-term exposure for the antifouling biocide Diuron using different species. Emphasis should be on monitoring, chronic exposure, risk assessment, organism's responses and pollution models to better sympathetic of the mechanisms and sustainable marine ecosystem health.
2. Government agencies, private sectors, researchers and other stakeholders should use the data for future planning, formulation of technical strategies to control antifouling contamination, risk assessment and develop new alternative antifouling chemicals in coastal regions, which is very important for conserving the ecological marine biodiversity.

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Conflicts of Interest

The author declares no conflicts of interest regarding the publication of this paper.

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