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HISTOPATHOLOGICAL INVESTIGATION OF SOME FACIAL COSMETIC PRODUCTS MIXTURE EXPOSED TO FINGERLINGS OF MUD CATFISH (*Clarias gariepinus*)

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ABSTRACT

Cosmetics are one of the major pollutants in the aquatic ecosystem due to the quantum of production to meet the astronomical demand of users that eventually becomes harmful to organisms. Changes in physic-chemical parameters such as potential of hydrogen (pH), Electrical conductivity (EC), Temperature (T °C) Dissolved oxygen (DO) were investigated at the beginning and after 96 hours of cosmetic exposure. This study also investigated the effects of four facial cosmetic products (Powder, foundation, concealer, and primer) histopathologically in fingerlings of *Clarias gariepinus* (gills) exposed to binary, trinary, and quaternary mixtures in laboratory bioassays for 48 hours and 96 hours. The results of pH, T °C, EC and DO ranged from 6.21 - 6.64, 25.40 - 26.70 °C, 0.04 - 0.14 mS/cm, and 10.00 -12.00 mg/l respectively. The joint action toxicity evaluation of the binary, trinary, and quaternary mixtures of the cosmetics prepared based on an equitoxic ratio against the test organism was in concordance with the model of synergism. Results showed the prevalence of severe lamellar necrosis in the gills of *C. gariepinus* which was observed across all combinations except in the combinations of Powder + Primer as well as Powder + Foundation + Concealer + Primer. Moderate lamellar necrosis and hypertrophied epithelium were observed in fish exposed to a mixture of Powder + Foundation + Concealer. The results obtained in this study suggested that the presence of facial cosmetics in the aquatic ecosystem could be dangerous to fish and subsequently human health via biomagnification. Therefore, there is an urgent need for environmental regulators to enforce safety standards for the emission of these selected cosmetics wastes into the waterways to prevent damage to aquatic organisms and public health issues.

Keywords: *Clarias gariepinus*, Cosmetic, Gill, Histological alterations, Joint action.

INTRODUCTION

The environment has been under constant assault from emerging pollutants at an astronomic level due to man's involvement in replicating industries to meet the needs of the growing population. In recent times one of the flourishing industries contributing to environmental pollution is the cosmetic industry (Zulaikha *et al.,* 2015).

Cosmetics otherwise known as beauty or personal care products or makeup are substances (lipstick, mascara, eyeshadow, foundation, blush, highlighter, bronzer, powder, concealer, and several others) introduced on or into the human body (epidermis, hair, nails, lips and external genital organs or mouth/teeth) to enhance beauty, cleanse, perfume, protect against germs, prevent or correct body odour (Singh, 2010) and make one attractive without distorting the body's structure or functions (Alsaffar and Hussein, 2014). The use of cosmetics has no boundaries because it is embraced by all races, genders, and ages which, however, raises significant ecological concerns due to the large amounts used and eventually flushed into the aquatic environment. Cosmetics, especially facial types are mostly applied externally in single or multiple combinations at once on the body to enhance appearance. The composition of these cosmetics is not subjected to transformation metabolically as they chart their course into the environment in an unchanged form after wash-off or during showering, bathing, or rinse-off (Ternes *et al*., 2004). The components of these cosmetics are sometimes environmentally persistent, bioactive, and potentially able to bioaccumulate (Brausch and Rand, 2011). These cosmetics can become potential environmental hazards; to humans and other exposed organisms (Adepoju-Bello *et al.,* 2012; Amasa *et al*., 2012, Popoola *et al.,* 2013; Ramakant *et al.,* 2014, Dhillon

et al., 2015). To evaluate the probable hazard caused by the direct impact of the wholesome rinse–off of some cosmetics facially applied, the histological tool was utilized in this study. Histopathological alteration is an established and widely used biomarker in the evaluation of contaminants, both in the laboratory (Wester and Canton, 1991; Thophon *et al.,* 2003) and in the field. This allows the examination of key organs such as gills, kidneys, and liver, that are responsible for vital functions, such as respiration, excretion, and the accumulation and biotransformation of xenobiotics especially in fish (Gernhofer *et al.,* 2001). In addition, the ease of identifying alterations in these organs makes the histopathological tool useful as an early warning signal of the deteriorating health of the organism (Hinton and Laurén, 1993).

The mud catfish (*Clarias gariepinus*) was used in this study based on its role in the fish web, ease of culturing in the laboratory, sensitivity, availability, abundance, and high consumption rate by the Nigerian populace (Adeogun and Chukwuka, 2012). The study was conducted to assess the histological alterations in *Clarias spp* induced by exposure to multiple mixtures of four facial cosmetic products in a laboratory bioassay.

MATERIALS AND METHODS

Materials

Collection and Acclimatization of Test Animals

Clarias gariepinus (Pisces, Cypriniformes, Clariidae) was the test organism selected based on its availability, abundance, sensitivity, consumption, and ease of maintenance under laboratory conditions. Healthy specimens of hatchery-bred fingerlings of *C. gariepinus* of similar sizes aged four weeks old with a mean weight of 0.96 ± 0.1 g and length of 3.5 ± 0.1 cm were purchased from a fish farm located at Okeke Street, Okota, Lagos, Nigeria. The fishes were taken to the Zoology Department laboratory, University of Lagos, Lagos, Nigeria in an oxygenated polythene bag containing water from the site of collection. In the laboratory, fishes were acclimatized in glass holding tanks (50 x 30 x 30 cm) containing a mixture of water from the site of collection and dechlorinated tap water. The *Clarias gariepinus* samples were left to acclimatize for 7 days. The water in the tanks was continuously aerated using 220 V air pumps (ACO-208-308, WuXi Sunolta, China) and changed once in two days to avoid accumulation of metabolic waste. The fishes were fed twice daily during the period of acclimatization with commercial fish feed pellets at 5% body weight. The fishes were fed every day until 24 hours preceding the bioassay test. Stocking and experimentation were carried out under ambient laboratory conditions (temperature 27 ± 3 °C, relative humidity 79 ± 2%) by guidelines for bioassay techniques.

Measurement of the Physico-Chemical Parameters of the Test Media

Physico-chemical parameters of the test media were measured at the commencement and end of the experiment. The potential of hydrogen (pH) was estimated using a Testr-2 pH meter. Electrical conductivity (EC) was measured using a portable combined Electrical conductivity Total Dissolved Solid (TDS) / Temperature (T°C) meter (HM Digital 716160 COM-100, USA). Dissolved oxygen (DO) was determined using a portable Orion 3 DO meter while temperature was measured using a HANNA instrument (H9835, USA).

Test Compounds

Two brands of Powder, Foundation, Concealer, and primer (sample size = eight) were used as test compounds against the fish fingerling. The cosmetics products were purchased from renowned stores within Lagos, a metropolis, in Nigeria though imported. The brand name, shade, Batch no, manufacturing date and expiry dates were also recorded (Table 1). The choice of these facial cosmetics was based on availability, affordability and most commonly used by teens and adults in Nigeria.

Table 1: Brand of Cosmetics Products

S/N	Cosmetic product	Shade	Batch No
1	Milani Compact Powder (MP)	110 DEEP	7320A1
2	Zikel Compact Powder (ZP)	CAPPUCCINO	N/A
3	Maybelline Fit Me Foundation (MF)	360 MOCHA	20R609
4	Glam Gals Matte Foundation (GF)	3GOLDEN TOFFEE	86841564,
5	LA Girl PRO. Concealer (LC)	GC983 FAWN	23664
6	Zaron Liquid Concealer (ZC)	AZ15 DEEP	LOT: C98
7	Ponds Primer (PP)	N/A	449SMK
8	Milk of Magnesia as Primer (MP)	N/A	WMR020

N/A= Not Available

Preparation of Stock Solution

Two (2) brands of Powder (P), Foundation (F), and Concealer (C) were prepared by dissolving 5 g of each in 100 mL of methanol after which serial dilutions of different concentrations were made. The Pond's primer ((P) solution was prepared by dissolving 5 g in 1000 mL of distilled water from which serial dilutions of different concentrations were made.

Response (Mortality) Assessment

Test animals were assumed to be dead when all body parts stopped moving even when probed with a glass rod. Mortality was assessed once every 24 hours for 96 hours for all the experimental setups.

Relative Acute Toxicity of Cosmetic Products Acting against *C. gariepinus* Fingerlings

Active *Clarias gariepinus* (n = 10) of similar sizes were randomly assigned to the test media in separate triplicate bioassay containers using a sieve. A total of 30 fish fingerlings were exposed per treatment including untreated control (dechlorinated tap water). The range finding of concentration was extrapolated to obtain definitive concentrations for the acute toxicity test over 96 hours' static bioassay setup.

Preparation of Test Media

The stock solutions were serially diluted to achieve solutions for exposure to the fishes. To prepare a test media, an amount of the stock solution was taken out with a pipette and poured into the bioassay container where it was made up to 1000 mL using the appropriate amount of dechlorinated water as diluents to obtain test concentrations (Table 2).

Table 2: Acute Toxicity Test Concentration

S/N	Brand of Cosmetics	Concentrations (mg/L)				
1	Milani Powder (MP)	25.0	75.0	125.0	175.0	225.0
2	Zikel Powder (ZP)	50.0	100.0	150.0	200.0	250.0
3	Maybelline Foundation (MF)	375.0	500.0	625.0	750.0	
4	Glam Gals Foundation (GGF)	250.0	375.0	500.0	625.0	750.0
5	La Girl Concealer (LGC)	125.0	250.0	375.0	500.0	625.0
6	Zaron Concealer (ZC)	100.0	200.0	300.0	400.0	500.0
7	Ponds Primer (PP)	125.0	187.5	250.0	312.5	375.0
8	Milk of magnesia (MM)	1.0	2.0	3.0	4.0	5.0

Relative Joint Toxicity of Cosmetics Mixtures against *C. gariepinus* for Analysis

A series of bioassays similar to those described for single action tests were carried out but at different concentrations of equitoxic ratios (1:1, 1:1:1 and 1: 1: 1:1). Each concentration of a mixture tested, the proportion of each constituent toxicant was dictated by the predetermined ratio of the mixture was computed, measured out into a conical flask and was made up to the required test media volume by adding dechlorinated tap water as diluent. The fish fingerlings were then exposed to varying concentrations of the mixtures. At the 96 hour' expiration, fishes were taken from each concentration randomly for histopathological analysis (Table 3).

Table 3: Joint Action Toxicity Test Concentration

S/N	Brand of Cosmetics	Ratio	Concentration (mg/L)
1	Powder (P) + Concealer (C)	1:1	400.0
2	Powder (P) + Foundation (F)	1:1	420.0
3	Powder (P) + Primer (Pr)	1:1	232.5
4	Powder (P) + Foundation (P) + Primer (Pr)	1:1:1	343.0
5	Powder (P) + Concealer (C) + Primer (Pr)	1:1:1	462.0
6	Powder (P) + Foundation (F) and Concealer (C)	1:1:1	351.0
7	Powder (P) + Foundation(F) + Concealer (C) + Primer (Pr)	1:1:1:1	404.3

Histological Analysis of the Gills of *C. gariepinus*

Histopathological analysis was performed after 48 hours and 96 hours of exposure to the cosmetic mixture. Fish were randomly selected from experimental and control mediums. Fish were euthanized and the gills were harvested and fixed in Bouin's solution. Then tissues were processed in a routine histological pattern and embedded in paraffin. Sections of 5 μm thick were taken which were stained in haematoxylin and eosin. Stained sections were produced as permanent slides and examined under a light microscope (Nikon TE 3000, Japan) at a magnification of X40 (high objective lenses) and a photomicrograph was taken with a digital camera (Nikon 9000, China).

Statistical Analysis

Physiochemical parameters were analysed by calculating the mean and standard deviation. Toxicological dose-response data involving quantal response (mortality) were analysed by Probit after Finney (1971) using SPSS (Statistical Package for Social Sciences) model 20.0. Toxicity indices derived from these analyses were LC_{50}. (lethal concentration that will bring about 50% mortality of the exposed population.

RESULTS

The result of the physico-chemical parameters of test media measured showed marked variation throughout the duration of the static tests. The pH, T °C, EC and DO ranged from 6.21 - 6.64, 25.40 - 26.70 °C, 0.04 - 0.14 mS/cm, and 10.00 - 12.00 mg/L respectively (Table 4). These parameters increased from the beginning of treatment to After 96 hours treatment except for dissolved oxygen. The pH increased highest by 0.24 in LGC and MM from the Beginning of facial product exposure to after (96 hours) Treatment compared to the Control (0.02). Temperature increased highest by 1.20 °C in LGC, ZC and MM from the Beginning of facial product exposure to after (96 hours) Treatment compared to the Control (0.30°C).

Electrical conductivity increased highest by 0.07 mS/cm in MM from the Beginning of facial product exposure to after (96 hours) Treatment compared to the Control (0.01 mS/cm).

Dissolved Oxygen decreased most by 3.00 mg/l in LA from the Beginning of facial product exposure to after (96 hours) treatment compared to the Control (2.00 mg/L).

Table 4: Physicochemical parameters of Test Media at the Beginning and After Exposure (96 hours)

Toxicant		Potential of Hydrogen (pH)	Temperature (T °C)	Electrical conductivity (mS/cm)	Dissolved oxygen (mg/L)
Control	Beginning	6.38 ± 0.01	25.80 ± 0.01	0.13 ± 0.01	12.00 ± 0.01
	After	6.40 ± 0.01	26.10 ± 0.01	0.14 ± 0.01	10.00 ± 0.01
Milani	Beginning	6.40 ± 0.01	25.50 ± 0.02	0.08 ± 0.01	10.00 ± 0.01
Powder	After	6.60 ± 0.01	26.50 ± 0.01	0.10 ± 0.01	8.00 ± 0.01
Zikel	Beginning	6.48 ± 0.01	25.40 ± 0.01	0.09 ± 0.01	11.00 ± 0.01
Powder	After	6.58 ± 0.01	26.50 ± 0.01	0.12 ± 0.01	9.00 ± 0.01
Maybelline	Beginning	6.50 ± 0.01	25.60 ± 0.01	0.07 ± 0.01	9.00 ± 0.02
Foundation	After	6.64 ± 0.01	26.40 ± 0.02	0.14 ± 0.01	8.00 ± 0.01
Glam gals	Beginning	6.58 ± 0.01	25.40 ± 0.02	0.08 ± 0.01	11.00 ± 0.01
Foundation	After	6.64 ± 0.01	26.50 ± 0.01	0.12 ± 0.01	10.00 ± 0.02
L.A Girl	Beginning	6.21 ± 0.01	25.50 ± 0.02	0.04 ± 0.01	10.00 ± 0.01
concealer	After	6.45 ± 0.01	26.70 ± 0.01	0.06 ± 0.01	7.00 ± 0.01
Zaron	Beginning	6.32 ± 0.01	25.40 ± 0.01	0.05 ± 0.01	10.00 ± 0.01
Concealer	After	6.50 ± 0.01	26.60 ± 0.01	0.08 ± 0.01	9.00 ± 0.02
Ponds	Beginning	6.37 ± 0.01	25.50 ± 0.01	0.04 ± 0.01	11.00 ± 0.01
Primer	After	6.57 ± 0.01	26.50 ± 0.01	0.07 ± 0.01	9.00 ± 0.01
Milk of	Beginning	6.21 ± 0.01	25.50 ± 0.01	0.04 ± 0.01	10.00 ± 0.02
Magnesia	After	6.45 ± 0.01	26.70 ± 0.01	0.06 ± 0.01	7.00 ± 0.01

'Beginning' means at the instance of exposure; 'After' means 96 hours of exposure

Single action toxicity results based on 96h LC_{50} showed that Milani Powder (184.14 mg/L) was the most toxic followed by Ponds Primer (230.95 mg/L), Zikel powder (233.58 mg/L), Zaron concealer (384.47 mg/L), Glam gals foundation (421.83 mg/L), Maybelline foundation (565.32 mg/L), L.A Girl concealer (587.75 mg/L) and Milk of Magnesia (2218.00 mg/L) in descending order of toxicity. The Toxicity Factor (TF) showed that Milani Powder was 12. 04 times more toxic than Milk of Magnesia (Table 5).

Table 5: Relative Acute Toxicity of Cosmetic Products tested Singly against *Clarias gariepinus* Fingerlings based on 96 hours

Toxicants (mg/L)	LC_{50} (mg/L)	LC_{95} (mg/L)	Probit line equation	Slope ± S.E	TF	DF
Milani	184.140	2776.663	Y= (-3.2)+1.4X	1.396 ± 0.628	12.04	3
Zikel	233.558	1401.700	Y= (-5.0)+2.1X	2.114 ± 0.884	9.50	3
Maybelline	565.322	660.878	Y= (-66.7)+24.3X	24.251 ± 7.12	3.92	3
Glam Gals	421.931	1043.531	Y=(-11.0) +4.2X	4.183 ± 1.227	5.26	3
LA Girl	587.750	4870.811	Y= (-5.0) +2.0X	1.791 ± 0.846	3.77	3
Zaron	384.466	1910.482	Y= (-6.1) + 2.4X	2.362 ± 0.881	5.77	3
Pond's	230.947	536.505	Y= (-10.6) + 4.5X	4.493 ± 1.252	9.60	3
Milk of Magnesia	2218	6360	Y= (-1.2) + 3.6X	3.595 ± 0.919	1.00	3

S.E = Standard Error: Toxicity Factor (TF) = 96h LC_{50} value of test compound/ LC_{50} value of most toxic compound and DF= Degree of Freedom

Histopathological examination of gills analysed at 48 hours of exposure of *Clarias gariepinus* showed that the gills in control displayed normal architecture while alteration such as shortening of the lamellar (Plate 1b), mild aneurism in secondary lamella (Plate 3a), severe lamellar necrosis (Plate 3b) of both primary and secondary lamellae (Plates 5b and 6b) was more pronounced but was mild in treatment combination of P + F + C + Pr (Plate 7b).

Histopathological alterations observed under 96 hours showed hypertrophied epithelium (Plate 9b), moderate lamellar necrosis (Plates 10b and 14b), shortening and blunting of primary and secondary lamellar (Plates 11b and 12b) while exposure to P + C + F showed hypertrophied epithelium and shortening of the secondary lamellar (Plate 13b).

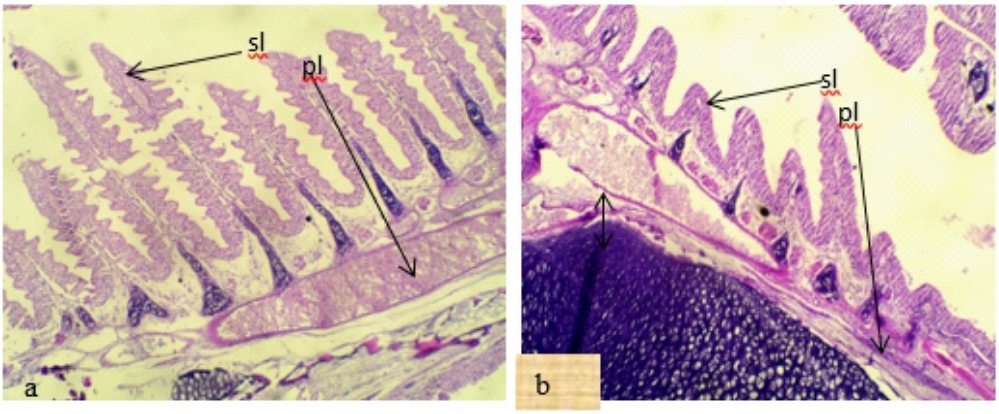

Plate 1: (a) Control with normal gills architecture with primary (pl) and secondary lamellae (sl). (b) Gill showed a shortened blunt lamellar tip and severe necrosis to the powder + Foundation mixture (double arrow).

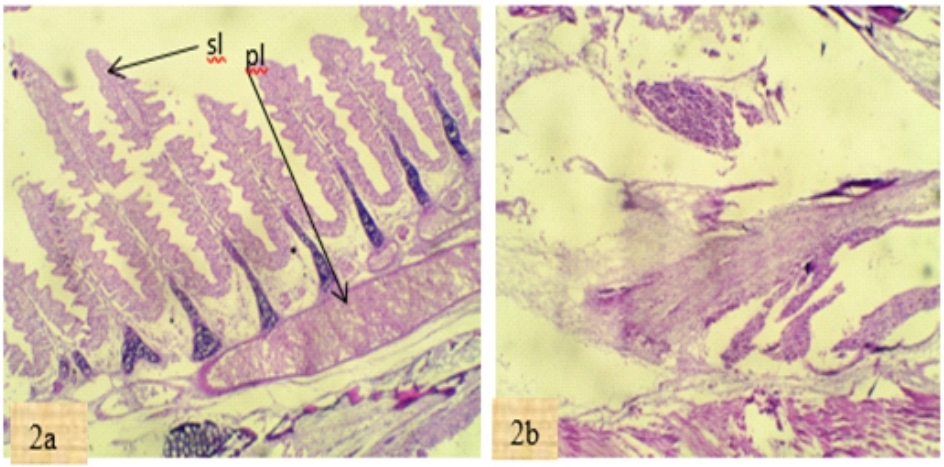

Plate 2 (a) Control with normal gills architecture with presence the of primary (pl) and secondary lamellae (sl). (b) No observed damage to exposure to Powder + Concealer.

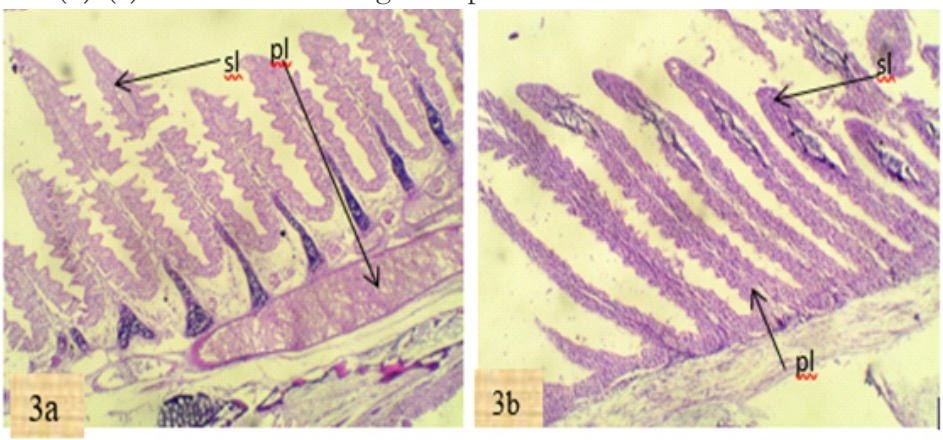

Plate 3 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl) (b) Exposure to Powder + Primer showed mild necrosis in secondary lamellar (sl).

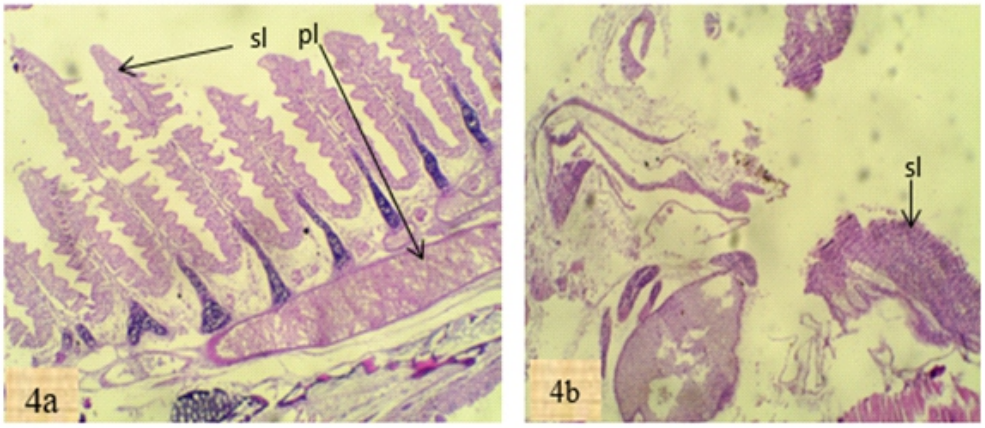

Plate 4 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) Exposure to Powder + Foundation + Primer mixture showed Severe lamellar necrosis (sl).

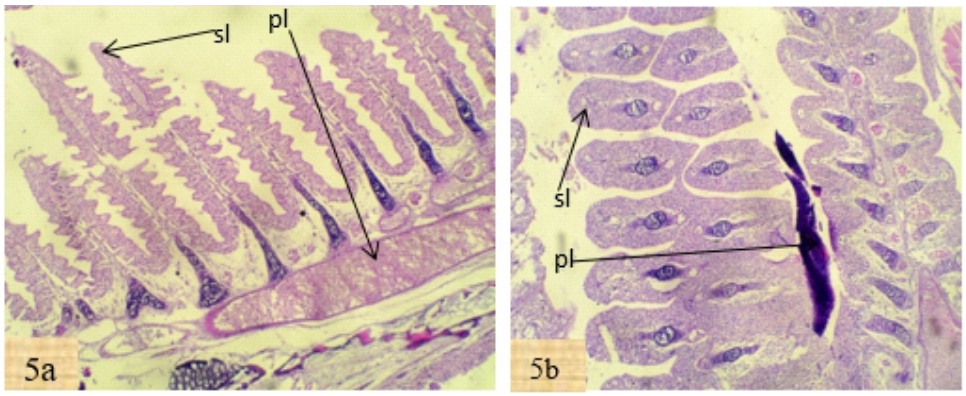

Plate 5 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) Exposure to Powder + Concealer + Primer showed shortening of secondary and severe necrosis of both primary (pl) and secondary lamellae (sl).

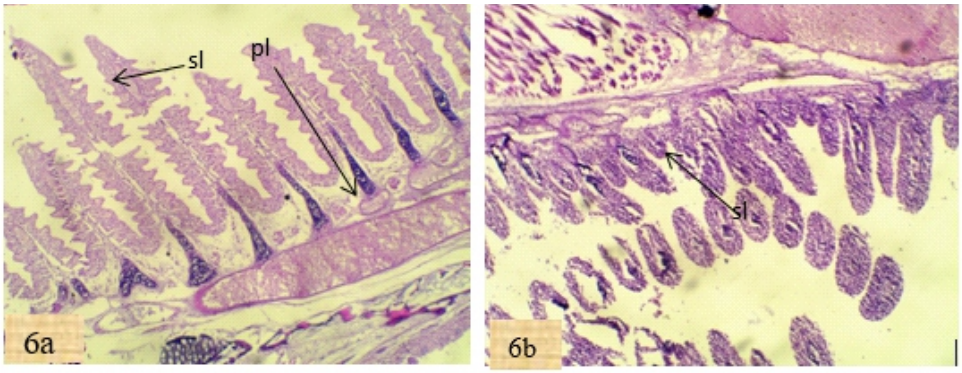

Plate 6 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) Exposure to Powder + Foundation + Concealer showed aneurism of secondary and primary lamellar, severe lamella necrosis (sl).

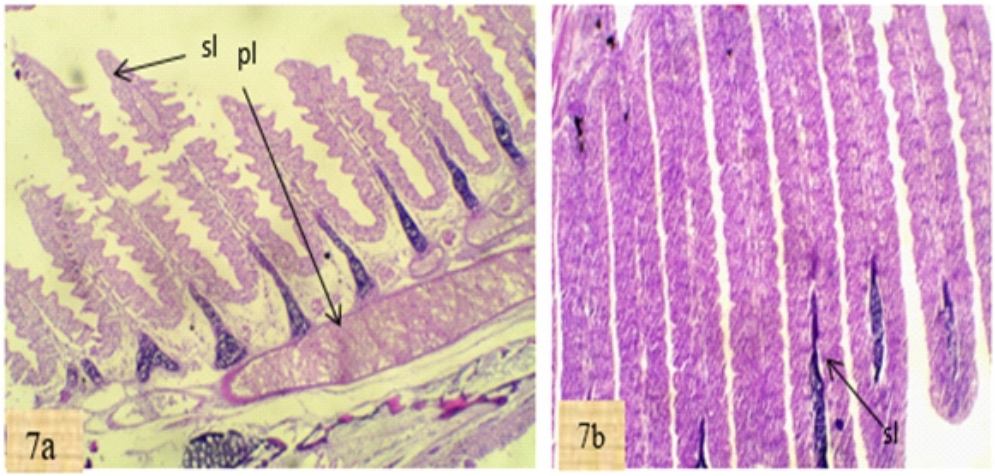

Plate 7 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) A combination of Powder + Foundation + Concealer + Primer showed mild necrosis on the secondary lamellar (sl).

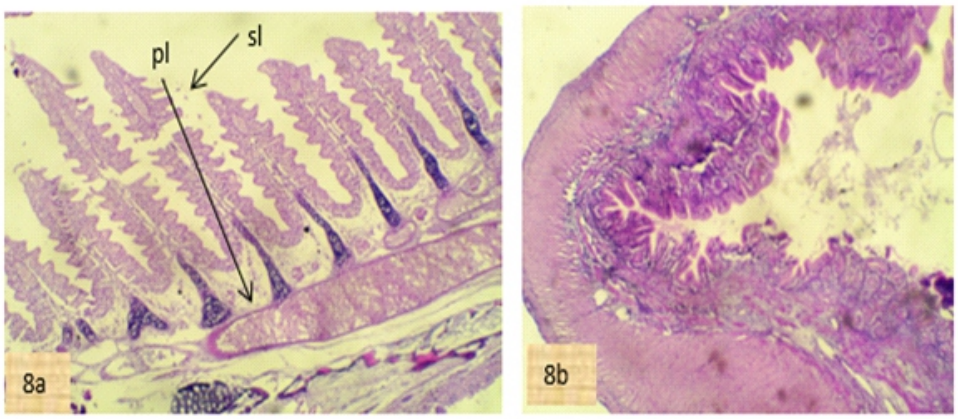

Plate 8 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) No observed damage to exposure to a Binary mixture of Powder + Foundation.

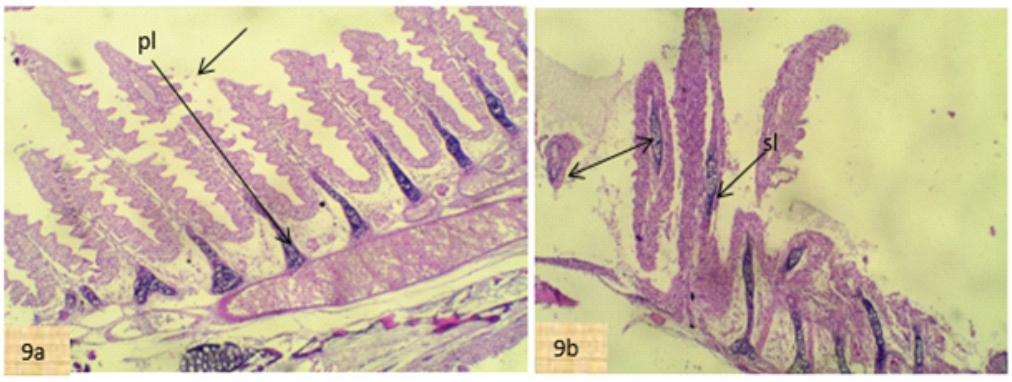

Plate 9 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) Exposure to a Binary mixture of Powder + Concealer showed lamellar tip fusion (double arrow) with hypertrophied epithelium.

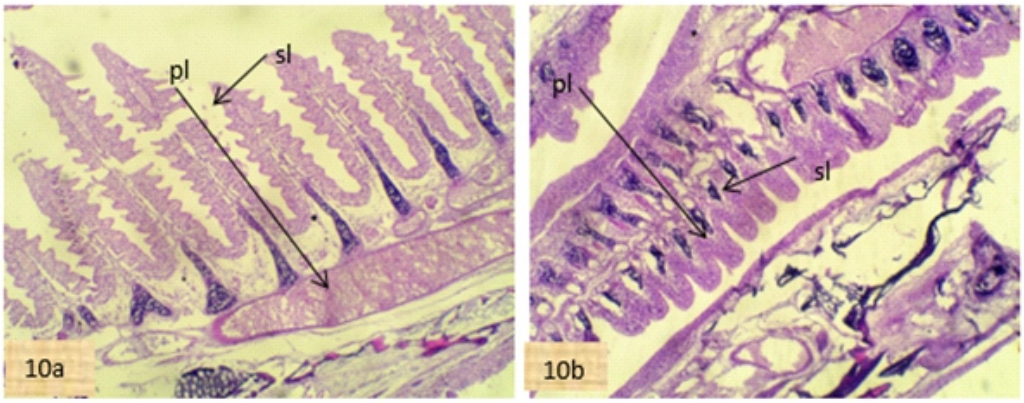

Plate 10 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) Exposure to a Binary mixture of Powder + Primer showed moderate lamella necrosis, shortening, and blunting of primary (pl) and secondary lamellar (sl).

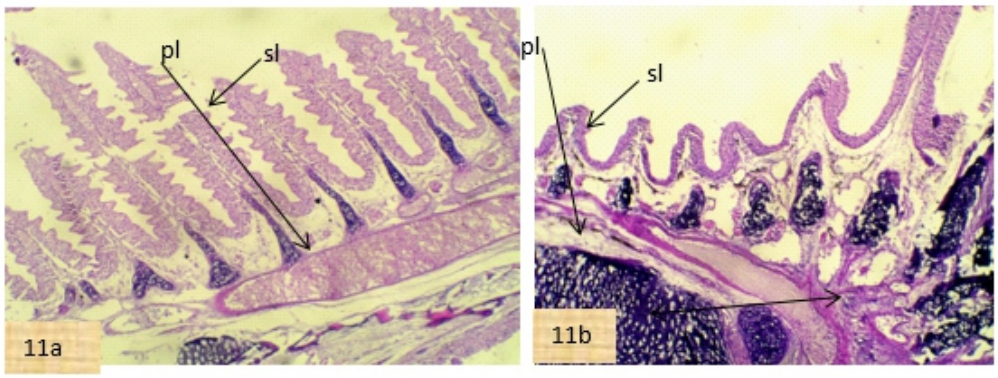

Plate 11 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) Exposure to a Binary mixture of Powder + Foundation+ Primer showed severe lamellar necrosis (double arrow) and shortening of primary (pl) and secondary lamellar (sl).

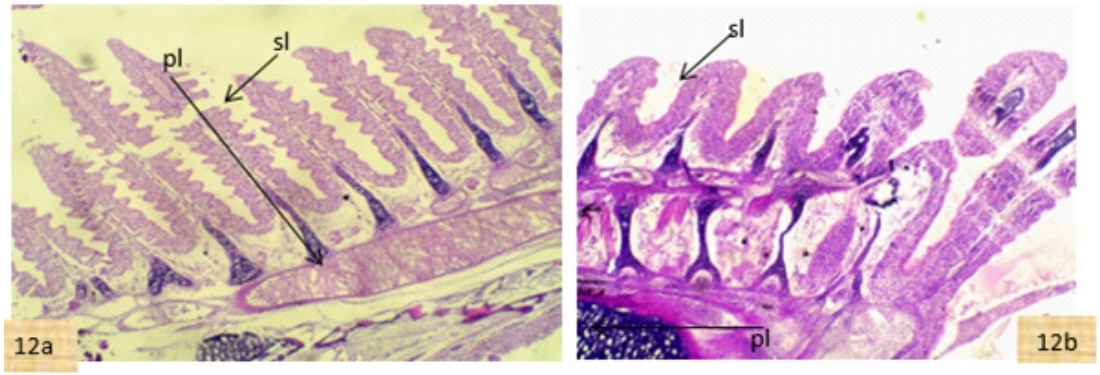

Plate 12 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) Exposure to a Triple mixture of Powder + Concealer + Primer exposure showed shortening of primary and secondary lamella (sl) and severe necrosis of primary lamellar (pl).

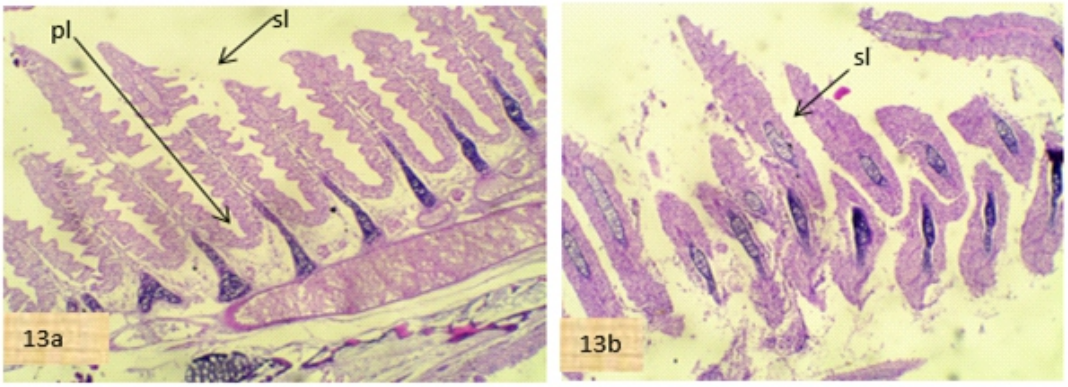

Plate 13 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) Exposure to a Triple mixture of Powder + Foundation + Concealer showed hypertrophy of epithelia and shortening of secondary lamellar (sl).

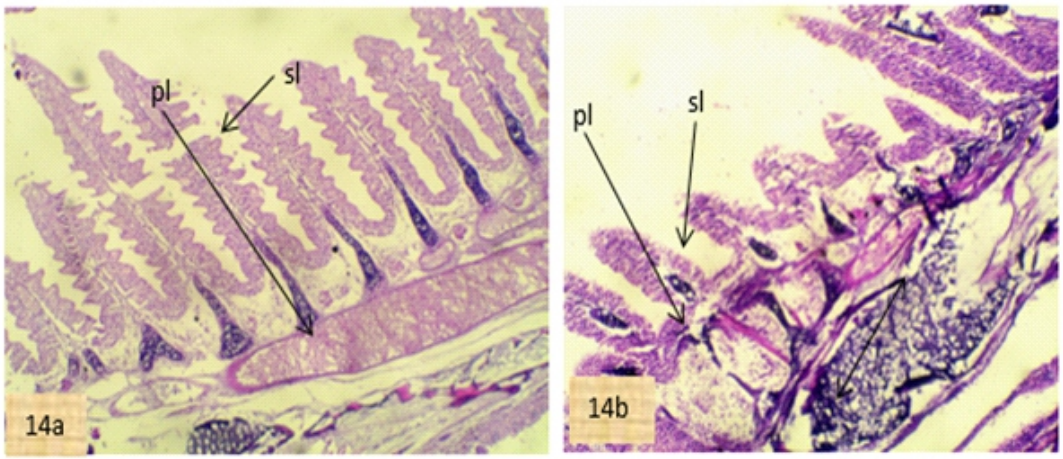

Plate 14 (a) Control with normal gills architecture with the presence of primary (pl) and secondary lamellae (sl). (b) Exposure to a quaternary mixture of Powder + Foundation+ Concealer + Primer showed moderate lamellar necrosis (double arrow).

DISCUSSION

All over the world especially in developing countries like Nigeria, the aquatic system and its inhabitants have been inadvertently exposed to assaults from numerous pollutants such as cosmetics that stand as one of the major contributors of pollutants in the aquatic ecosystem. This is based on the release of large amounts hinged on increased usage resulting in environmental degradation.

This study investigated the Physico-chemical parameters and histopathology of *Claris gariepinus* (gills) exposed to eight commonly used facial cosmetic products (in the form of powder, foundation, concealer, and primer) in mixtures under laboratory bioassays for 48 hours and 96 hours.

The physic-chemical parameters of the test media recorded showed slight variation in the level of the parameters measured both at the beginning and the end of observation. Decreased dissolved oxygen at the end of the test period when compared to its values at the beginning could be attributed to the presence of facial cosmetics. Ezemonye and Ogbomida, (2010) had earlier reported that the introduction of a toxicant into an aquatic system might decrease dissolved oxygen concentration, which will impair respiration leading to asphyxiation. This probably explains why the tested facial cosmetic products could be responsible for decline in oxygen levels in the treatment bioassay containers.

In the case of dissolved oxygen, the treatments did not only show a dose-dependent decline in concentration, but also rapid depletion of dissolved oxygen with time.

Milani Powder which was the most toxic based on acute toxicity test could be based on the fact that the fishes were most sensitive to the characteristic component of the toxicant compared to the other facial cosmetic products. These characteristics determine the ability of the toxicant to penetrate living organisms at the metabolism site hence exerting its toxic effect (Sogbanmu and Otitoloju, 2014). However, the use of acute toxicity assay to rank the selected cosmetics products used in this study does not suffice because it does not measure harmful effects on exposed test animals thus a complementary method of histopathology was inculcated into this study.

Histopathological assay falls in line because it measures direct toxic effects on target organs (Schwaiger *et al*., 1997). This could be attributed to the fact that histopathology shows changes caused by pollutants determined by time of exposure, dose/concentration and adaptability of the tissue exposed (Ferreira *et al*., 2004). Structural alterations in exposed tissues of animals are detected through histological examination (a simple and sensitive tool). In fish, gills are the first organ to which the pollutant primarily contacts and also functions as the site of respiration, osmoregulation and excretion (Mallatt, 1985). Thus, gills are often predisposed to damage.

Gills are generally good indicators of water quality (Rankin *et al*., 1982), being models for the studies of environmental impact (Bonga and Locke, 1992) as the primary route for the entry of toxicants. Gills are the major respiratory organs and all metabolic pathways depend upon the efficacy of the gills for their energy supply, and damage to these vital organs causes a chain of destructive events, which ultimately lead to respiratory distress. Secretion of mucus over gills curtails the diffusion of oxygen (David *et al.,* 2002) which ultimately reduces oxygen uptake by the fish. Histopathological results indicated that the gill was the primary target tissue affected by the cosmetic products.

The histopathological examination of the gills of *Clarias gariepinus*, exposed to the facial cosmetic mixture for 48 hours, revealed shortened lamellae, necrosis, and mild aneurysm of the lamellae. This is in contrast to the normal architecture in the control group, suggesting that the cosmetic components had a direct effect on the gill structure. This corroborates the findings of (Temmink *et al*., 1983 and Garcia-Santos *et al*., 2007). This could also be a defense mechanism for the fish, an early warning signal. Alteration of the gill could hamper the respiratory ability of the fish which probably leads to damage of the pillar cells, weakness of their support function, and overall disorder of the lamellae. This finding is also in agreement with that of Abdel-Moneim, *et al.* (2008).

Histopathological alterations observed under 96 hours indicated hypertrophy of epithelial cells of the gills that is also a defense mechanism. This could result in the increased distance between the external environment and the blood thus serving as a barrier to the entrance of contaminants (Fernandes and Mazon, 2003) in this case, of the facial cosmetic mixture. A higher concentration of cosmetic products and prolonged hours of exposure could be responsible for pathological changes such as lamellar fusion due to simple apposition as well as hypertrophy of epithelial cells.

It must be emphasized that histopathology can evaluate the early effects and the responses to acute exposure to chemical stressors.

CONCLUSION

As a consequent action of anthropogenic activities, emerging contaminants (cosmetic formulation inclusive) flushed into the environment are growing astronomically daily. The study showed that facial cosmetic mixtures in the aquatic environment could deplete dissolved oxygen, and result in histological alteration in the gills that could cause mortality of aquatic fauna. Histological alterations observed in the gills as indicated in this study rout under the toxicity of multiple mixtures of cosmetics can be utilized as a sensitive tool to monitor aquatic pollution. Therefore, further studies on the acute and chronic toxicity of the varied types of cosmetic

products in singular and multiple mixtures should be assessed to ascertain the actual ecological and health risks. In addition, manufacturers should be compelled to display the environmental impacts (adverse effects) of cosmetics on their packaging to furnish consumers with information that will censor the use of these products. Regulators should ensure strict prohibition of those products with adverse effects on the environment into our market space.

AUTHORS' CONTRIBUTIONS

O.F.I.: Conceptualization and Methodology; A.O.A.: Data curation; Writing- Original draft preparation; I.M.N.: Visualization; O.O.P.: Investigation; O.F.I.: Supervision; H.O.F.: Writing, Reviewing and Editing.

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CONFLICT OF INTEREST

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