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Effects of oil from Bonny Light Crude on Adult Winglength, Fluctuating Asymmetry, Body size and Nutritional reserve in *Anopheles gambiae* (s.l.)

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ABSTRACT: The study was carried out to determine the influence of crude-oil contaminated mosquito larval habitat on the wing length, fluctuating asymmetry, adult body size and nutritional reserve of *Anopheles gambiae* (s.l.) The oil was obtained by distillation of bonny light crude obtained from Port-Harcourt, Nigeria using appropriate standard methods. Results obtained showed that the left and right wing lengths varied significantly across concentrations in the male population ranging from 3.25±0.42 (0.5 mlL^{-1}) to 3.83±0.41mm (0.1 mlL^{-1}) and 3.33±0.26mm (1.00 mlL^{-1}) to 3.75±0.27mm (0.1 mlL^{-1} and 0.25 mlL^{-1}), respectively. The left and right wing lengths of the female mosquitoes, however, did not vary significantly; ranging from 3.53±0.26mm (0.5 mlL^{-1}) to 3.83±0.26 mm (0.1 mlL^{-1}) and 3.58±0.20mm (0.75 mlL^{-1}) to 3.67±0.41mm (1.00 mlL^{-1}), respectively. The female population showed varying fluctuating asymmetry across concentrations; these were statistically different ranging from 0.16±0.25mm (in Control) to 0.30±0.26mm (in 0.50 mlL^{-1}). The mosquitoes varied significantly in body sizes across concentrations. Male mosquito body size ranged from 37.58±8.20mm (in 1.00mlL^{-1}) to 55.08±10.41 mm (in 0.1mlL^{-1}), while female body size ranged from 37.72±4.19 (in 0.5mlL^{-1}) to 54.85±8.05 mm (in 0.1mlL^{-1}). The teneral reserve showed a consistent trend of decrease along the concentration as it increases. The glycogen reserve was significantly low (5.44±0.41 µg/mL) in the 1.0mlL^{-1} population and highest (8.91±0.23 µg/mL) in the control population, though the glycogen in the control was not significantly different from those of 0.1mlL^{-1}, 0.25mlL^{-1} and 0.5mlL^{-1} but was significantly different from 0.75mlL^{-1} (7.10±0.24 µg/mL). The result of this study gives a better understanding of the interaction between crude oil contamination and vectorial fitness attributes of *Anopheles gambiae* to serve as prelude for effective vector control in such area with crude oil spillage.

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Mosquitoes are slender, fragile, flying insects of about 3 – 6 mm in length belonging to the family culicidae (Okwa and Savage, 2018). Mosquitoes are the most important insect affecting human health. There are presently 3550 species of mosquitoes recognized worldwide that are contained in two subfamilies and 112 genera (MTI; Mosquito Taxonomic Inventory, 2016). Mosquitoes are responsible for the spread of malaria, the most common deadly disease ranking next to HIV/AIDS (Rowton, 2005). Vector borne diseases accounts for a significant number of the global infectious diseases burden, including morbidity and mortality. Mosquito borne diseases in particular have the greatest burden in number of cases, mortality and disability adjusted life years (WHO; World Health Organization, 2018). Insects are good bio indicators for environmental impact assessment due to their high species diversity and importance in the functioning of natural ecosystem. Exposure of insect to hydrocarbon is generally lethal, hence the use of some aromatic

hydrocarbon aerosol as insecticides (Asimea and Zakka, 2011). Petroleum hydrocarbon pollution of the environment may result from oil wells drilling, production operations, transportation and storage in the upstream industry (Onwurah *et al*, 2007; Arimoro and Adamu, 2008). Petroleum hydrocarbons or carbon containing compounds are converted into free radicals or activated metabolites during their oxidation in the cells. The activated metabolites react with some cellular components such as membrane lipids and produce peroxidation products which may lead to membrane damage (Odo *et al.*, 2012). Mosquito larvae are found in habitats possessing a wide range of physico-chemical factors. The larvae of mosquitoes are known to thrive significantly in clear water of suitable pH, temperature and nutrient condition (Devi *et al*., 2014). Scientific research has shown that many species of mosquitoes prefer habitats without oxygen tension while some breed in open, sunlit pools (Okogun, 2005). Water of a near neutral pH value of 6.8 - 7.2 is preferable for breeding of many species of mosquitoes generally (CDC; Centre for Disease Control and Prevention, 2004). High prevalence of malaria in Nigeria has been reported severally because the country is home to the most efficient vector of the disease (i.e., *Anopheles gambiae*). This mosquito thrives greatly in Nigeria because of the favourable tropical climatic conditions and proliferations of suitable larval breeding habitats especially during raining seasons. The Niger delta area in Nigeria is characterized by extensive wetlands suitable for mosquito breeding and play host to oil exploration activities in the country, to the extent that Nigeria is rated as one of the major oil producing nations in the world. Due to reported reckless oil exploration activities in the Niger Delta of Nigeria encouraged by poor monitoring activities and application of sanctions by the regulatory bodies, rampant oil spillage and hence, contamination of water bodies including potential mosquito breeding habitats is the order of the day in this region. Though, mosquito densities are high even in the Niger delta in Nigeria as earlier reported by different authors despite the fact that the level of pollution of crude oil in the area should ordinarily discourage mosquito breeding, this therefore suggest a complex, interaction between crude oil or its components and physiological activities of mosquitoes breeding in such habitats.

Anopheles could lay eggs in oily breeding sites and both the eggs and larvae emerging from the eggs will undergo series of selection pressure which ends with the emergence of the oily breeding sites of adult *Anopheles gambiae* which are capable of withstanding lethal doses of pyrethroids (Djouaka *et al*., 2007a). Hence, the objective of this paper is to evaluate the effect of crude-oil contamination on wing length and nutritional reserve of *Anopheles gambiae* using bonny light crude obtained from Port-Harcourt, Nigeria.

MATERIALS AND METHODS

Source of crude-oil and preparation of water soluble fraction of crude oil: Bonny light crude oil was obtained from the Nigerian National Petroleum Corporation (NNPC), Port-Harcourt, Nigeria and water soluble fraction (WSF) was made with distilled water following standard procedures (Nwabueze and Agbogidi, 2010; Olaifa, 2012; Rishika *et al*., 2022) with slight moderations in the ratio of 1:1 in which 500ml of crude oil was mixed with 500ml of distilled water in a 2000ml capacity screwed conical flask, placed on a table top Gallenkamp magnetic stirrer and stirred for an accumulated 24 hours at ambient temperature. The mixture was then left to stand for 3 hours undisturbed to obtain a clear interphase between oil and water and clear water soluble fraction (WSF) obtained at the lower part of the funnel was siphoned and used for the experiment.

Bio-Assay of Crude Oil against Anopheles gambiae Immature Stages: The study was carried out in eight replicates each of the concentrations (1.0ml L^{-1}, 0.75ml L^{-1}, 0.5ml L^{-1}, 0.25ml L^{-1}, and 0.1ml L^{-1}), respectively after pilot studies. Each replicate contained 25, larvae of *Anopheles gambiae* species per plastic container. The larvae were reared following standard techniques (Olayemi *et al*., 2012; Abubakar *et al.,* 2018), in plastic containers of 350ml capacity which were labelled appropriately containing 100 mls of borehole water. The experiment was monitored daily at 0700 and 1900 hours for larval mortality, metamorphosis (duration of development) pupation and adult eclosion. The whole experiment was repeated within 2 weeks of the termination of the first for enhanced replication. A control experiment similar to the treatment but devoid of crude oil were also set up.

Determination of Adult Body Size (Wing length), Volume and Fluctuating Asymmetry: After 24-hours of emergence, the wings were carefully detached (i.e right and left) with the aid of a dissecting microscope fitted with an ocular micrometer gauge, measurement of the wing from the apical margin to the alular notch will be taken and the length will be measured (Ukubuiwe *et al*. 2016). Volume of adult mosquitoes were expressed as cube values of wing lengths while fluctuating asymmetry is difference between lengths of the right and left wings.

Evaluation of Teneral/Nutrient Reserve of Anopheles gambiae: To evaluate the accumulation of

teneral/nutrient reserve, 10 randomly selected adults of each treatment were analysed for teneral reserves i.e., lipid, glycogen, sugar and protein content were measured by methods described by Bradford (1976), Van Handel (1985a,b), Ukubuiwe *et al*. (2018).

Standard preparation for lipids: Lipid standard were prepared using 100mg of soybean oil mixed with 100ml of chloroform as the solvent and placed in a clean test tube in triplicate concentrations of 50, 100, 200 and 400µl.the solution was heated at 100^0C until the solvent evaporates then allowed to cool to room temperature, 0.2ml of acid was added and heated for 10 mins at 100^0C. Vanillin – phosphoric acid reagent [(vanillin 600mg) + hot distilled water (100ml) + 400ml of 85% phosphoric acid] was added to make it up to 5ml and allowed to react for 10 mins until a reddish colour was obtained before being plated in a 96 well micro plate reader.

The optical density (OD) was determined at 625nm and µg lipid was plotted against OD. (Van Handel, 1985b; Ukubuiwe *et al*., 2018).

Standard preparation for glycogen and glucose: 100ml distilled water was used to dissolve 100mg of anhydrous glucose in triplicate concentrations of 25, 50, 100, 150 and 200µl and placed in a glass test tubes. Anthrone reagent [95-98% sulphuric acid (385ml) + distilled water (150ml) + anthrone (750mg)] was added to make it up to 5ml level and mixed. The solution was heated t 100^0C for 17mins and allowed to cool. The OD was read at 625nm and plotted against µg glycogen. (Van Handel, 1985b; Ukubuiwe *et al*., 2018).

Standard preparation for Protein: This was prepared using procedures of Bradford (1976). 50µl of bovine serum albumin in serial concentration of 10, 20, 40, 80 100µg was pipetted into test tubes. The volume in each test tubes were made up to 1ml with phosphate buffer (0.1M, pH 6.6). 5 ml of protein reagent [Coomassie Brilliant Blue G-250 (100mg) + 95% ethanol (50ml)] was added to the test tube and mixed. The OD was read at 595nm. Ice cold saline buffer (0.9% Nacl) was added to the mosquitoes in a centrifuge tube and homogenized, the homogenate was centrifuged at 4000 rpm for 20 minutes. Supernatant was stored at -20^0C until used. 100ml of phosphoric acid 85% was added to the protein reagent (coomassie brilliant blue G-250 (100mg) + 95% ethanol (50ml)) and the resulting solution was made up to a 1000ml by diluting with distilled water. 50µl of the solution was pipetted into test tubes and made up to 1 ml with phosphate buffer (0.1M, pH 6.6) OD was measured at 595nm after 2 minutes and before1 hour against blank prepared from 1ml of phosphate buffer and 5ml protein reagent (Ukubuiwe *et al*., 2018).

Extraction of lipid, glycogen, glucose and protein from mosquitoes: 0.2ml of 2% sodium sulphate solution was added to the mosquito sample in a centrifuge tube and homogenized with a glass rod until no identifiable part was left, the glass rod was washed clean with 0.8ml of 1:1 volume of chloroform/methanol solution into a clean centrifuge tube and centrifuged at 3000rpm for 1 minute. The supernatant was transferred to a clean centrifuge tube, while the pellets was kept for glycogen analysis. 0.6 ml of distilled water was added to the supernatant, mixed and centrifuged at 3000rpm for I min. the top fraction (water and methanol) was used for sugar analysis and the bottom portion was used for lipid analysis (Ukubuiwe *et al*., 2018).

Lipid analysis: The portion for Lipid analysis was placed in a test tube already marked at 5ml level and heated at 100^0C until the solvent evaporates then allowed to cool to room temperature, 0.2ml of sulphuric acid was added and heated for 10 minutes at 100^0C. Vanillin reagent [(vanillin 600mg) + hot distilled water (100ml) + 400ml of 85% phosphoric acid] was added to make it up to 5ml and allowed to cool and react for 30 minutes for a reddish colour to be developed. The optical density (OD) was determined at 625nm. (Van Handel, 1985b; Ukubuiwe *et al*., 2018).

For Glycogen and glucose: Anthrone reagent [95-98% sulphuric acid (385ml) + distilled water (150ml) + anthrone (750mg)] was added to the pellet to the 5ml level mark and mixed. The solution was heated at 100^0C for 17mins and allowed to cool. The OD was read at 625nm. (Van Handel, 1985b; Ukubuiwe *et al*., 2018).

For glucose: The fraction for glucose analysis was placed in a tube already marked at 5ml level and heated at 100^oC to evaporate the solvent down to 0.15ml. anthrone reagent was added to make it up to 5ml and mixed, the solution was heated at 100^oC for 17 minutes and then left to cool. The OD was determined at 625nm (Ukubuiwe *et al*., 2018).

For Protein: This was prepared using procedures of Bradford (1976). 50µl of bovine serum albumin in serial concentration of 10, 20, 40, 80 100µg was pipetted into test tubes. The volume in each test tubes were made up to 1ml with phosphate buffer (0.1M, pH 6.6). 5 ml of protein reagent [Coomassie Brilliant Blue G-250 (100mg) + 95% ethanol (50ml)] was added to the test tube and mixed. The OD was read at 595nm.

Ice cold saline buffer (0.9% Nacl) was added to the mosquitoes in a centrifuge tube and homogenized, the homogenate was centrifuged at 4000 rpm for 20 minutes. Supernatant was stored at -20^{0}C until used. 100ml of phosphoric acid 85% was added to the protein reagent (coomassie brilliant blue G-250 (100mg) + 95% ethanol (50ml)) and the resulting solution was made up to a 1000ml by diluting with distilled water. 50µl of the solution was pipetted into test tubes and made up to 1 ml with phosphate buffer (0.1M, pH 6.6) OD was measured at 595 nm after 2 minutes and before1 hour against blank prepared from 1ml of phosphate buffer and 5ml protein reagent (Ukubuiwe *et al*., 2018).

RESULTS AND DISCUSSION

Effect of water soluble fraction of crude oil on wing length of Anopheles gambiae: Wing length (mm) of *Anopheles gambiae* s.l reared in water soluble fraction of crude oil medium is presented in Table 1. The left and right wing length varied significantly (p=0.05) across concentrations in the male population ranging from (3.25±0.42 (0.5 mlL^{-1}) to 3.83±0.41mm (0.1 mlL^{-1}) and 3.33±0.26 (1.00 mlL^{-1}) to 3.75±0.27mm (0.1 mlL^{-1} and 0.25 mlL^{-1}) while that of the females did not vary significantly ranging from 3.53±0.26 (0.5 mlL^{-1}) to 3.83±0.26 mm (0.1 mlL^{-1}) and 3.58±0.20 (0.75 mlL^{-1}) to 3.67±0.41mm (1.00 mlL^{-1}), respectively. The mean wing length were significantly different (p=0.05) among the concentrations (range=3.33±0.2mm in 1.00 mlL^{-1} to 3.79±0.25mm in 0.1mlL^{-1}) and (3.35±0.12 mm in 0.5 0mlL^{-1} to 3.79±0.19 mm in 0.1mlL^{-1}) in the male and female population respectively. The aggregate population had significantly varying wing length across concentration (Range=3.39±0.36 in 0.50mlL^{-1} to 3.83±0.33 in 0.1mlL^{-1} and 3.50±0.37 in 1.00mlL^{-1} to 3.75±0.26 in 0.1mlL^{-1}) in the left and right wings respectively.

Effect of water soluble fraction of crude oil on the fluctuating Asymmetry of Anopheles gambiae: The fluctuating asymmetry FA (mm) of *Anopheles gambiae* s.l reared in water soluble fraction of crude oil medium is shown in Table 2. In the male population, 1.00mlL^{-1} produced mosquitoes with no fluctuating asymmetry (0.00 ±0.00) while 0.1mlL^{-1} produced mosquitoes with the highest fluctuating asymmetry (0.25±0.41). The female population showed varying fluctuating asymmetry across concentrations, ranges though not statistically significant (p=0.05) (range=0.16±0.25mm to 0.30±0.26mm in control and 0.50 mlL^{-1}, respectively). The fluctuating asymmetry did not vary significantly (p=0.05) (range=0.13±0.21mm in control to 0.27±0.26 in 0.50mlL^{-1}) in the aggregate.

Effect of water soluble fraction of crude oil on the Body size of Anopheles gambiae: Body size (mm^3) of *Anopheles gambiae* s.l reared in WSF of crude oil medium is presented in Table 3. The mosquitoes showed significantly (p=0.05) varying body sizes across concentrations, (range=37.58±8.20mm^3 in 1.00mlL^{-1} to 55.08±10.41 mm in 0.1mlL^{-1}) and 37.72±4.19mm^3 in 0.5mlL^{-1} to 54.85±8.05mm^3 in 0.1mlL^{-1}) in the male and female population respectively. In the aggregate population, 0.1mlL^{-1} produced mosquitoes with the largest body size (54.96±8.88mm^3) while 0.5mlL^{-1} produced the least body size (39.42±10.08mm^3).

Table 1: Wing length (mm) of *Anopheles gambiae* as influenced by water soluble fraction of crude oil

Concentrations (mlL^{-1})	Wing length (mm)								
	Male			Female			Aggregate (Male and Female)		
	Left	Right	Mean	Left	Right	Mean	Left	Right	Mean
Control	3.67± 0.26^{ab*}	3.57± 0.22^{ab}	3.62± 0.22^{abc}	3.67± 0.26^{a}	3.67± 0.25^{a}	3.67± 0.21^{b}	3.67± 0.25^{ab}	3.62± 0.23^{a}	3.64± 0.20^{bc}
0.10	3.83± 0.41^{b}	3.75± 0.27^{b}	3.79± 0.25^{c}	3.83± 0.26^{a}	3.75± 0.27^{a}	3.79± 0.19^{b}	3.83± 0.33^{b}	3.75± 0.26^{a}	3.79± 0.21^{c}
0.25	3.67± 0.41^{ab}	3.75± 0.27^{b}	3.71± 0.29^{bc}	3.58± 0.20^{a}	3.58± 0.38^{a}	3.58± 0.26^{ab}	3.63± 0.33^{ab}	3.67± 0.33^{a}	3.65± 0.27^{bc}
0.50	3.25± 0.42^{a}	3.58± 0.38^{ab}	3.42± 0.38^{ab}	3.53± 0.26^{a}	3.67± 0.26^{a}	3.35± 0.12^{a}	3.39± 0.36^{a}	3.63± 0.31^{a}	3.38± 0.30^{ab}
0.75	3.58± 0.37^{ab}	3.50± 0.00^{ab}	3.54± 0.19^{abc}	3.75± 0.27^{a}	3.58± 0.20^{a}	3.67± 0.20^{b}	3.67± 0.31^{ab}	3.54± 0.14^{a}	3.60± 0.20^{bc}
1.00	3.33± 0.26^{a}	3.33± 0.26^{a}	3.33± 0.26^{a}	3.58± 0.38^{a}	3.67± 0.41^{a}	3.63± 0.34^{ab}	3.46± 0.32^{a}	3.50± 0.37^{a}	3.48± 0.33^{a}

*Valuesfollowedbysamesubscriptalphabetsinacolumnarenotsignificantly different using Duncan MRT at P=0.05
Values are expressed as Mean ± SD

Effect of water soluble fraction of crude oil on the Nutritional reserve of Anopheles gambiae: The teneral/nutritional reserves of *Anopheles gambiae* reared in water soluble fraction of crude oil is presented in table 4. The teneral reserve showed a consistent trend of decrease along the concentration as

it increases in protein and sugar. The glycogen reserve was significantly low (5.44±0.41µg/mL) in the 1.0mL^{-1} population and highest (8.91±0.23 µg/mL) in the control population, though the glycogen in the control was not significantly different from those of 0.1mL^{-1}, 0.25mL^{-1} and 0.5mL^{-1} but was significantly different from 0.75mL^{-1} (7.10±0.24 µg/mL). 0.75mL^{-1} population had the lowest lipid reserve (5.34±0.56µg/mL), it was significantly different (p=0.05) from 1.00mL^{-1} (5.87±0.21µg/mL) and 0.5mL^{-1} (5.67±0.29 µg/mL) but differed significantly from 0.1mL^{-1} (6.04±0.19µg/mL) and 0.25mL^{-1} (6.21±0.33 µg/mL) while the control (0.00mL^{-1}) accumulated the highest (7.24±0.36 µg/mL) lipid differed significantly from others. Protein reserve was least (1.88±0.11µg/mL) protein when compared to the control (6.11±0.21µg/mL) and it differed significantly (p = 0.05) from other population except for 0.75mL^{-1} (2.14±0.18µg/mL). The glucose reserve was lowest in the 1.0mL^{-1} (1.90±0.18µg/mL) as compared to the control population which had the highest (5.00±0.14µg/mL) sugar reserve. The population showed significantly varying glucose reserve except for 0.25mL^{-1} (3.25±0.56µg/mL) and 0.1mL^{-1} (3.27±0.26µg/mL).

Table 2: Fluctuating asymmetry (mm) of *Anopheles gambiae* wings as influenced by water soluble fraction of crude oil.

Concentrations (mL^{-1})	Male	Female	Aggregate
Control (0.00)	0.10±0.20^{a*}	0.16±0.25^{a}	0.13±0.21^{a}
0.10	0.25±0.41^{a}	0.25±0.27^{a}	0.25±0.34^{a}
0.25	0.25±0.27^{a}	0.17±0.26^{a}	0.21±0.26^{a}
0.50	0.25±0.27^{a}	0.30±0.24^{a}	0.27±0.25^{a}
0.75	0.25±0.27^{a}	0.17±0.26^{a}	0.21±0.26^{a}
1.00	0.00±0.00^{a}	0.25±0.27^{a}	0.13±0.23^{a}

*Valuesfollowedbysamesubscriptalphabetsinacolumnarenotsignificantly different using Duncan MRT at P=0.05
Values are expressed as Mean±SD

Table 3**:** Body size (mm^3) of *Anopheles gambiae* reared water soluble fraction of crude oil.

Concentrations (mL^{-1})	Body size		
	Male	Female	Aggregate
Control (0.00)	47.73±8.99^{abc*}	48.82±8.58^{b}	48.78±8.46^{bc}
0.10	55.08±10.41^{c}	54.85±8.05^{b}	54.96±8.88^{c}
0.25	51.78±11.70^{bc}	46.61±10.32^{ab}	49.20±10.86^{bc}
0.50	41.12±14.11^{b}	37.72±4.19^{a}	39.42±10.08^{a}
0.75	44.73±7.02^{b}	49.68±8.52^{b}	47.21±7.88^{abc}
1.00	37.58±8.20^{a}	48.68±12.55^{ab}	43.13±11.65^{ab}

*Values followed by same superscript alphabets in a column are not significantly different using Duncan MRT at P=0.05
Values are expressed as Mean±SD

Table 4**:** Nutritional composition of *Anopheles gambiae* S.L (Adult) reared in Water Soluble Fraction of Crude oil

Concentrations (mL^{-1})	Nutrients(µg/mL)			
	Glycogen	Lipid	Protein	Glucose
Control (0.00)	**8.91±0.23^{c*}**	**7.24±0.36^{c}**	**6.11±0.21^{e}**	**5.00±0.14^{d}**
0.1	8.90±0.24^{c}	6.04±0.19^{b}	4.11±0.18^{d}	3.27±0.26^{c}
0.25	8.34±0.38^{c}	6.21±0.33^{b}	3.40±0.26^{c}	3.25±0.56^{c}
0.50	8.51±0.38^{c}	5.67±0.29^{ab}	2.74±0.39^{b}	2.67±0.19^{b}
0.75	7.10±0.24^{b}	**5.34±0.56^{a}**	2.14±0.18^{a}	2.36±0.27^{ab}
1.00	**5.44±0.41^{a}**	5.87±0.24^{ab}	**1.88±0.11^{a}**	**1.90±0.18^{a}**

*Values followed by same superscript alphabets in a column are not significantly different using Duncan MRT at P=0.05
Values are expressed as Mean±SD

The aggregate adult wing length were reduced in the 0.5mL^{-1} concentration and this could be as a result of the damage detection and control in mosquitoes. This could imply that mosquitoes emerging from this concentration will be smaller in body size as compared to other concentrations. Female mosquitoes from the 0.1 mL^{-1} were the largest in body size and by implication would be more efficient in malaria transmission as they will need more host to bite for blood meal. The fluctuating asymmetry was increased in the 0.5mL^{-1} in the female population and the aggregate population. Mosquitoes with increased fluctuating asymmetry will have difficulty in flying to a host for blood meal. FA results from internal and external stresses during immature development and its used as indicator of suitable breeding sites for mosquito development and adult fitness for disease transmission. In this study, the quantities of Lipid and glycogen across all concentrations were higher than protein and glucose, this could be as a result of the physiological importance of the lipid and glycogen in mosquitoes. These findings correspond with the

reports of Ukubuiwe *et al.* (2018) that says the lipid and glycogen reserve were significantly higher than the protein and glucose reserve in *Culex quinquefasciatus*. Among the concentrations of WSF of crude oil, the highest concentration ($1.00mlL^{-1}$) caused significant reduction in the teneral reserve. This suggests that breeding and development in $1.00mlL^{-1}$ WSF of crude oil may discourage greater accumulation of teneral reserves. Earlier studies have shown relationship between greater quantities of reserve components with higher body weights (Lanciani and Anderson, 1993), it could be suggested that individual from this $1.00mlL^{-1}$ have lesser body weight and could not be better fit as vectors of diseases.

Conclusion: Crude-oil contamination of mosquito larval habitat affects the wing length, body size and nutritional reserve of *Anopheles gambiae*. The least concentration tested produced mosquitoes with the largest body size. Nutritional reserve were affected negatively, as the reserve decreases with increasing concentrations of WSF of crude oil which is indicative that though the concentration may not be lethal but it is capable of giving a long term effect on the mosquitoes.

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