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CONTENTS OF THE ISSUE

- i. Copyright Notice
 - ii. Editorial Board Members
 - iii. Chief Author and Dean
 - iv. Contents of the Issue
-
1. Born to Die: Global Healthcare from Ethical Standpoint. **1-2**
 2. Communities of Helminth Parasites in five Carangidae Species from the Coast of Veracruz, Mexico, Southern Gulf of Mexico. **3-13**
 3. Assessment of the Invasive Alien Plant Species *Lantana Camara* in Nile River Millennium Park, Bahir Dar, Ethiopia. **15-22**
 4. Population Estimate, Group Size and Age Structure of the Gelada Baboon (*Theropithecus Gelada*) around Debre-Libanos, Northwest Shewa Zone, Ethiopia. **23-29**
 5. Microorganisms and Aflatoxin Content in Ready-To-Eat Groundnut Paste from Some Markets in Anambra and Edo States, Nigeria. **31-37**
-
- v. Fellows
 - vi. Auxiliary Memberships
 - vii. Process of Submission of Research Paper
 - viii. Preferred Author Guidelines
 - ix. Index



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Born to Die: Global Healthcare from Ethical Standpoint

By Ngoc Bao Huynh

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Abstract- There have been discourses on the issues of health care differences between the developing and developed world and the need to bridge the health care divide or the provision of health care services for all countries. A case study between Haiti and Harvard highlighted the poor and good state of health care system as it relates to causes of death. Though, universal health care is high and unsustainable as a result of several militating factors including cost and politics, cost is not as important as saving a life, recognizing human rights, resources for economic productivity, lowering the cost of health care, which implies that every hand must be on deck including the government, individuals, organizations and other richer countries to help develop a good health care system. Hence, the debate of sustainability health care system or universal health care is based on the matter of social, political and economic sustainability and policymaking.

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Born to Die: Global Healthcare from Ethical Standpoint

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I. INTRODUCTION

In 2015 the London Review published books written by Dr. Paul Farmer titled “Who Lives and Who Dies.” In this work, he describes problems about the iniquities of healthcare funding in Haiti, Rwanda, Lesotho, etc. and the possibility of universal health care. Paul Farmer is an anthropologist and a physician who is well known for his humanitarian work as a founder of Partner in Health, an organization whose mission is to provide suitable health care to help poor developing counties. In the review, Farmer brought up the differences in the healthcare system between his work places in Haiti and Harvard. He saw differences in the cause of death in poor countries because of the poor health system. There is a big difference between the health services in poor and rich countries that gives rise to a debate question; is it because the poor are just unlucky to be born in developing countries instead of rich countries that make their life more at risk of disability and death? Some argue that limited resources where people lack medical insurance and/or treatment access is the issue, while others consider lack knowledge for disease prevention. In these countries, it is not uncommon for people to die of things like malaria, typhoid and childbirth. Resolving these issues could be as simple as educating people water sanitation, hand washing and providing bed nets. There are many questions that arise from Farmer’s works about good health care and access for basic disease treatment; Should there be basic care/proven care in places like Haiti, Rwanda, Lesotho, etc., or is universal health care (healthcare for

all regardless of who you are and where you are) “unsustainable” and therefore not worth doing? If yes, who should pay for truly universal health care?

To address the first question, should there be basic care/proven care in places like Haiti, Rwanda, Lesotho, etc., or it universal health care (healthcare for all regardless of who you are and where you are) “unsustainable” and therefore not worth doing, there are various way to answer this questions. First, we must consider one of the four principles of biomedical ethics-justice. According to Tom Beauchamp and James Childress in the Principles of Biomedical Ethics, healthcare resources need to be distributed fairly since all people have human rights ^[1]. This means all individual have the right to access the same healthcare resources and should not treat unequally no matter in who they are, their age, sex, quality of life, social or economic status, race, etc. Furthermore, based on the second principle of bioethics-beneficence, healthcare providers have obligation to help people in need and healthcare resource should target toward maximizing health gain ^[2]. Therefore, there should be basic care/proven care in places like Haiti, Rwanda, Lesotho, etc., and universal health care is best option.

This brings forth the second part of the first question, is universal health care (healthcare for all regardless of who you are and where you are) “unsustainable” and therefore not worth doing? Some argue that the cost of universal health care is too high. The political systems in many poor countries make universal health care unsustainability and therefore even minimal essential healthcare is not worth offering. Ethically, I disagree, Universal health care is sustainable based on the term rationing which means even though there is a limitation of medical care, and it should be provided to all patients ^[3]. Furthermore, limited resources should be distribution in a fair manner. It is not a smart idea to spend a large amount of money for small essential outcomes in terms of economic decision in business. But this is not true when it comes to healthcare and justice. Consider a good outcome where the cost is not at all important, such as saving a life, recognizing human rights, resources for economic productivity, lowering the cost of health care, and making it more affordable in the long-term. Furthermore, humans possess quality of dignity and claim of life. This means a wealthy society should help the poor society from falling below the baseline of subsistence and dignity.

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This brings lights to the second question, who should pay for truly universal health care? To answer this question, we must consider many conditions. Though the government has a large responsibility to establish the health system and healthcare decisions, they are not meant to be the only one with the obligation to provide all the essential resources or money to make universal health care sustainable in their countries. In short, everyone including individuals, families, businesses, charities, and churches, share in the responsibility to make it happen and come true. The government cannot force people to help others, but it should authorize, equip, and create a condition for all groups of communities to fulfill a mutual obligation to live the life of justice.

The answers to these questions are not easy to make, but the debate of sustainability health care system or universal health care is based on the matter of social, political and economic sustainability and policymaking. In my view, health policymaking is an important force to make universal health care sustainable and suggests a definitive solution to solve the problem. Without good political systems and right policies, the issue of healthcare will remains a controversial topic for debate.

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COMMUNITIES OF HELMINTH PARASITES IN FIVE CARANGIDAE SPECIES FROM THE COAST OF VERACRUZ MEXICO SOUTHERN GULF OF MEXICO

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Communities of Helminth Parasites in five Carangidae Species from the Coast of Veracruz, Mexico, Southern Gulf of Mexico

Jesús Montoya-Mendoza ^α, Guillermo Salgado-Maldonado ^ο, Mario E. Favila-Castillo ^ρ, Gabriela Vázquez-Hurtado ^ω & María del Refugio Castañeda-Chávez [¥]

Abstract- In 140 specimens of five carangid species were captured in Playa Las Barrancas and El Cabezo Reef, Veracruz Reef System National Park, Veracruz State, Southern Gulf of Mexico: *Caranx crysos* (n=51), *Caranx hippos* (n=18), *Chloroscombrus chrysurus* (n=28), *Oligoplites saurus* (n=24) and *Trachinotus carolinus* (n=19), a total of 44 helminth species were recovered, distributed as follows: 18 digeneans (17 adults, and 1 metacercaria), 12 monogeneans, 9 nematodes (6 adults, and 3 larvae), 4 cestodes (all larvae), and 1 acanthocephalan (juvenile). Parasite of helminths species with the highest prevalence in five communities were *Pseudobicotylophora atlantica* and *Amphipolycotyle chloroscombrus*, while species with mean intensity were *Hurleytrema catarinensis*, and the nematode *Hysterothylacium* sp., was registered in all five communities. The component community with highest richness and diversity was for *C. crysos* ($S=21$, Shannon index $H'=2.19$), at infracommunity level highest richness was for *T. carolinus* ($S=4.5 \pm 2.1$) and *C. hippos* ($S=4.1 \pm 2.8$, while the highest diversity was for *C. chrysurus* (Brillouin index $H=1.03 \pm 0.32$) and *C. crysos* (Brillouin index $H=1.01 \pm 0.44$). The highest Similarity Index of was between the communities of *C. crysos* and *C. hippos* (Jaccard index=60%). Results suggest that compositions, richness and diversity are similar to other founded marine fish from tropical and temperate latitudes.

Keywords: communities, helminth parasites, *c. crysos*, *c. hippos*, *c. chrysurus*, *o. saurus*, *t. carolinus*, *mexico*.

I. INTRODUCTION

Taxonomic studies of parasitic helminths in marine fish families, including Carangidae species, are numerous in Mexico and different areas (Lamothe-Argumedo et al. 1997; Konh et al. 2006; García-Prieto et al. 2006; Pérez-Ponce de León et al. 2007; Overstreet et al. 2009; Jensen 2009). However, These studies were concern to species with commercial interest, such as *Trachinotus carolinus* (Sánchez-Ramírez & Vidal-Martínez

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2002), *Eugerresplumieri*, *Hexanematichthys assimilis*, *Oligoplites saurus* and *Scomberomorus maculatus* (Aguirre-Macedo et al. 2007), *Symphurus plagiosa* (Rodríguez-González & Vidal-Martínez 2008), *Centropomus nigrescens* (Violante-González et al. 2010), *Lutjanus campechanus* and *L. synagris* (Montoya-Mendoza et al. 2014; 2016). In respect to carangid fish species, in Mexico, have records of parasite communities of *T. carolinus* (Sánchez-Ramírez & Vidal-Martínez 2002) and *O. saurus* (Aguirre-Macedo et al. 2007), only, unlike other areas from Brazil, e.g. *O. palometa*, *O. saurus*, and *O. saliens* (Takemoto et al. 1995), *Caranx hippos* and *C. latus* (Luque & Alves 2001), *C. hippos* (Boada et al. 2012), *Selene setapinnis* (Cordeiro & Luque 2004), and *T. goodei* (Luque & Cezar 2004).

These authors have indicated that communities of helminth parasites of marine fish are rich, abundant and diverse, particularly in tropical latitudes (Rohde & Heap 1998; Alves & Luque 2006; Luque & Poulin 2007), in this paper, its assumed that the community parasites obtained from five carangid fish species are similar in richness species and diversity compared to the parasite communities reported in marine fish species from other temperate and tropical latitudes. Moreover, the helminth communities are described in terms of species composition, species richness, diversity and similitude.

II. MATERIAL AND METHODS

a) Sampling Procedures

A total of 140 organisms from five species of the Carangidae Family were examined between August, 2004 and February, 2007. Specimens were caught in Playa Las Barrancas (18°59'31"N, 95°57'83"W), of Alvarado Municipality, Veracruz, Mexico. With a beach seine net (500m long x 4–5 m high; ¼–1 in mesh). Larger fish were caught by hook-and-line at El Cabezo Reef (19°03'07"N, 95°52'05"W), 11.7 km east of Playa Las Barrancas. All fish collected were transported alive to the laboratory and placed in 1,000 L tanks, while dead organisms were kept in plastic containers with ice, and transported to the lab for examination within 24 hours post-capture. Taxonomic designations of fish were done according to Froese & Pauly (2016).

Tissues and organs were reviewed using a stereomicroscope. The external examination included skin, scales, fins, gills, eyes, nostrils, mouth, and anus. Gills were removed and analyzed separately in Petri dishes with seawater. Internal examination included mesenteries, liver, kidney, and gonads, and the whole digestive system was placed in Petri dishes with 0.75% saline for examination. Helminths were fixed with hot 4% formalin and preserved in 70% ethyl alcohol.

For taxa identification, monogeneans, digeneans, cestodes, and acanthocephalans were stained using either Mayer's paracarmine, Gomori's triple stain or Erlich's hematoxylin, and then dehydrated in a graded alcohol series, cleared with clove oil, and mounted whole in Canada balsam. Nematodes were studied on temporary slides and cleared in glycerin, and then preserved in 70% alcohol. In order to study sclerotized structures, some specimens of monogeneans were fixed with ammonium picrate (Vidal-Martínez et al. 2001). Voucher specimens were deposited at the National Helminths Collection (*Colección Nacional de Helminthos*) (CNHE), Institute of Biology of the National Autonomous University, Mexico City.

b) Sample Size

Helminth communities in the five species of carangids were analyzed at the component community (all helminths in all individuals of species examined), and infra-community (helminths in each fish examined) levels (Holmes & Price 1986). Helminth species richness observed was one measure of the adopted community structure. Sampling adequacy for the component community was evaluated with procedure similar to the one used for helminth parasites communities of *L. campechanus* (Montoya-Mendoza et al. 2014), and *L. synagris* (Montoya-Mendoza et al. 2016), using a randomized (100x) sample-based species accumulation curve computed in Estimate S (version 8.5 RK Colwell, <http://viceroy.eeb.unconn.edu/estimates>) (Moreno & Halffter 2001). For the component community, we examined the asymptotic richness based on the Clench's model equation (Soberon & Llorente, 1993), besides the final slope of the randomized species accumulation curve (Jiménez-Valverde & Hortal 2003). Clench's model is described by the following function: $V2 = (a \times V1) / [1 + (b \times V1)]$, where $V2$ is the observed richness, $V1$ is the number of hosts examined, and a and b are curve parameters; a equals the new species adding rate, and b is a parameter related to the curve shape. These values were calculated using the Estimate S and Statistica software (Stat Soft, Inc., Tulsa, Oklahoma) as in Jiménez-Valverde & Hortal (2003). The slope of the cumulative species curve was calculated as $a / (1 + b \times n)^2$, where a and b are parameters cited above and n is the number of hosts examined for a given component. The Clench's model equation allows

estimating the total number of species in a component as a/b . To calculate the number of rare species missing at the component community level, the nonparametric species-richness estimator bootstrap was calculated from data observed, as recommended by Poulin (1998).

c) Data analysis

The prevalence (percentage of infected hosts) and mean intensity (mean number of parasites per infected fish) were calculated following Bush et al. (1997); as well as the correlation between the total number of species with the total number of helminths, and compared with the host size and weight. We analyzed the distribution of helminth species abundance for community components by rank-abundance curves and data were adjusted to predictive distribution models ($\chi^2, p = 0.05$) recording the dominant species in each community. These values were calculated using the PAST version 3.14 (Hammer et al. 2001). The Shannon index of diversity (H'), was calculated for the component community as in Magurran (2004). Infra-community descriptors included the mean number of helminth species per fish, the mean number of helminth individuals per fish, and the mean value of the Brillouin's diversity index per fish (H). Similarity among all five parasite communities was estimated with a cluster analysis using the Jaccard similarity index (Magurran 2004).

III. RESULTS

A total of 140 specimens from five carangids species were collected: 51 blue runners, *Caranx crysos*; 23 Crevalle jacks, *Caranx hippos*; 28 Atlantic bumpers, *Chloroscombrus chrysurus*; 24 Leather jackets, *Oligoplites saurus*; and 19 Florida pompanos, *Trachinotus carolinus*. Size and weight are displayed in Table 1.

a) Parasite parameters

The 44 species of parasites collected, belonged to 18 trematodes species (17 adults and 1 metacercaria); 12 monogeneans; 9 nematodes (6 adults and 3 larva); 4 cestodes (all larvae); and 1 acanthocephalan (juvenile) (see Table 2). Hosts fish species with the highest proportion of trematodes were *T. carolinus* (9 species, 50%) and *C. crysos* (8 species, 38%), and the highest proportion of monogeneans was found in *C. hippos* (6 species, 33%).

Hysterothylacium sp., nematode larvae were recorded in all five host species, while the cestode larva of *Scolex polymorphus*, the trematode *Gonocerca* sp., and the nematode *Hysterothylacium fortalezae* were recorded in four host species. The other parasites were recorded in one, two or three host species. According to inventory of parasites of this study, are reported 12 new locality records and 17 new host records (Table 2).

Table 1: Total Length (Lt, cm) and Weight (W, g) of the five Carangid Species from Alvarado, Veracruz, Mexico.

Host	n	Lt(± ES)	Range	W(± ES)	Range
<i>C. crysos</i>	51	24.5 ± 11.6	13.4-43.5	262.1 ± 290.1	25.3-885
<i>C. hippos</i>	18	30.1 ± 33.2	11.6-116	1077.1 ± 2670	25-8250
<i>C. chrysurus</i>	28	15.9 ± 1.6	10.5-19.5	41.12 ± 8.2	30-63.1
<i>O. saurus</i>	24	20.6 ± 4.1	13.6-27.3	70.7 ± 36.6	17.3-155
<i>T. carolinus</i>	19	32.8 ± 9.3	13.6-45.9	491.1 ± 280.7	40-1072

Parasites with the highest prevalence were the monogenean *Pseudobicotylophoraatlantica* (89.5%) and the trematode *Huerleytremaatarinensis* (73.7%) for the host *T. carolinus*; the monogenean *Amphipolycotylechloroscombrus* (82.1%) for the host *C. chrysurus*; and the monogenean *Probursataveraecrucis* (66.6 %) for the host *Amphipolycotyle chloroscombrus* (82.1 %). Trematodes with the highest abundance were *H. catarinensis* (7033, $P_i = 0.98$) for the host *T. carolinus*, and *Manteriabrachydera* (198, $P_i = 0.51$) for *C. hippos*. Most abundant monogeneans were *Cemocotylec-arangis* (414, $P_i = 0.367$) for *C. hippos* and *Amphipolycotyle chloroscombrus* (112, $P_i = 0.25$) for the host *C. chrysurus* (see Table 2).

b) Sample Size

The cumulative species curves developed with the Clench model, showed that our species inventories are almost complete, considering that the slope value of the last point of the curve was less or close to 0.1 ($b_{xi} \leq 0.1$), and that we collected between 80% and 95% of the species that make up each community. Also, the Clench model showed that there are some species needing to be collected, based on the a/b value (S_e , richness expected), and corroborated by the Bootstrap richness estimator (Table 3).

Table 3: Richness of component communities of helminthes parasites of five species of Carangids from Alvarado, Veracruz, Mexico. Data include: n, number of hosts examined; #th, total number of helminths; So, number of observed helminth species; Se, number of helminth species estimated with the Clench model; R^2 , correlation coefficient between date and Clench model; b_{xi} , date of the condition species curve as calculated from Clench model; % sp Cle, proportion of species by the Clench model; S_{Boot} , richness estimated by Bootstrap.

Host	n	#th	So	Se	R^2	b_{xi}	% sp Cle	S_{Boot}
<i>C. crysos</i>	51	1126	21	23	0.9981	0.03	89	22
<i>C. hippos</i>	23	1620	18	22	0.9992	0.13	80	19
<i>C. chrysurus</i>	28	455	12	14	0.9996	0.04	89	14
<i>O. saurus</i>	24	388	7	8	0.9739	0.02	95	8
<i>T. carolinus</i>	19	10184	18	22	0.9991	0.18	80	20

c) Correlations of richness and abundance

Significant correlation ($\alpha = 0.05$) was found between the total number of species (S) or the total number of helminths (N), when compared to the host size was to *C. crysos* (total host length vs. S, $r = 0.76$; vs. N, $r = 0.56$), and *C. hippos* (total host length vs. S, $r = 0.82$; vs. N, $r = 0.86$), but no significant correlation to *C. chrysurus* (total host length vs. S, $r = 0.34$; vs. N, $r = 0.01$), *O. saurus* (total host length vs. S, $r = 0.41$; vs. N, $r = 0.39$), and *T. carolinus* (total host length vs. S, $r = 0.37$; vs. N, $r = 0.21$).



Table 2: Helminth parasites of 5 carangid species from Alvarado, Veracruz, Mexico. Data include: nhp, number of hosts parasitized; tnhl, total number of helminthes; mnt, mean intensity; % prev, prevalence; in, intestine; ic, intestinal ceca; s, spleen; m, mesentery; bv, biliary vesicle; g, gill; h, head; un, under skin; l, larve; mt, metacercaria, *new locality record, **new host record.

Helminth	<i>Caranx cyosus</i> (n = 51)				<i>Caranx hippos</i> (n = 23)				<i>Chloroscombrus cyosurus</i> (n = 28)				<i>Oligoplites saurus</i> (n = 24)				<i>Trachinotus carolinus</i> (n = 19)			
	site	nhp (% prev)	tnhl (Pi)	mnt (range)	nhp (% prev)	tnhl (Pi)	mnt (range)	nhp (% prev)	tnhl (Pi)	mnt (range)	nhp (% prev)	tnhl (Pi)	mnt (range)	nhp (% prev)	tnhl (Pi)	mnt (range)	nhp (% prev)	tnhl (Pi)	mnt (range)	
TREMATODA																				
<i>Lobatosomakeim</i>	i																			
<i>ostomaMacCallum</i>																				
<i>&MacCallum, 1913</i>																				
<i>Lobatosomaringe</i>	i																			
<i>ns(Linton, 1907)</i>																				
<i>Manteribrachyder</i>	i																			
<i>a</i>																				
<i>(Manter, 1940)</i>																				
<i>Stephanostomum d</i>	i	12	64	5.3 ± 5.25	2	12	6 ± 5.7													
<i>itrematis(Yamaguti,</i>		(23.5)	(0.057)	(1-17)	(8.7)	(0.007)	(2-10)													
<i>1939)*</i>																				
<i>Stephanostomum g</i>																				
<i>hanensisFischthal</i>																				
<i>& Thomas, 1968</i>																				
<i>Stephanostomum h</i>	i	6	28	4.7 ± 4.8	3	17	5.6 ± 8.1													
<i>megacephalumMa</i>		(11.8)	(0.025)	(1-13)	(13.4)	(0.01)	(1-15)													
<i>nter, 1940*</i>																				
<i>Ectenurosyamaguti</i>	i	3	10	3.3 ± 3.2																
<i>Nahas& Powell,</i>		(5.8)	(0.009)	(1-7)																
<i>1971*</i>																				
<i>Bucephalus</i>																				
<i>margaritaeOzaki &</i>	i, ic	14	72	5.1 ± 4.5	14	709	50.6 ± 97													
<i>Ishibashi, 1934*</i>		(27.5)	(0.064)	(1-14)	(61)	(0.44)	(1-282)	5	13	2.6 ± 2										
<i>Tergestiapectinata</i>	i	5	10	2 ± 1.2	3	9	3 ± 3.5													
<i>(Linton, 1905)*</i>		(9.8)	(0.009)	(1-4)	(13.04)	(0.006)	(1-7)	3	4	1.3 ± 0.6										
<i>Huerleytremacatari</i>																				
<i>nensisAmato,</i>	i, ic																			
<i>1982*</i>																				
<i>Opechonachlorosc</i>																				
<i>ombriNahas&</i>	i																			
<i>Cable, 1964*</i>																				
<i>Huerleytremasp.*</i>	i																			
<i>Neolepidapedons</i>	i																			
<i>P.</i>																				
<i>Stephanostomums</i>	i	3	11	3.7 ± 0.6																
<i>P.</i>		(5.9)	(0.01)	(3-4)																
<i>Gonocerellasp.</i>	i																			
<i>Gonocercasp.*</i>	g	2**	2	1 ± 0	3**	4	1.3 ± 0.6	1**	1	1 ± -	1**	1	1 ± -	1**	1	1 ± -	1**	1	1 ± -	
		(3.9)	(0.002)	(1-1)	(13.04)	(0.002)	(1-2)	(3.6)	(0.002)	(1)	(4.1)	(0.003)	(1)	(4.1)	(0.003)	(1)	(4.1)	(0.003)	(1)	
<i>Macrorchytremasp</i>	i																			
<i>*</i>																				
<i>Didymozoidae</i> ^{(ml)*}	s	16**	83	5.21 ± 0																
		(31.4)	(0.074)	(1-1)																

MONOGENEA		9	3	95	31.7 ± 13 (29-46)	27	414	15 ± 13.2 (1-45)	9**	58	6.4 ± 8.9 (1-28)	23	112	4.8 ± 2.3 (2-10)	14	70	5 ± 4.8 (1-15)	
<i>Hargicolaoligoplite</i> S (Hargis, 1957)	g					(52.9)	(0.367)		(39.1)	(0.036)		(82.1)	(0.246)		(58.3)	(0.18)		
<i>Allopyragraphorus hippos</i> (Hargis, 1956)	g	3	(13.04)			(5.9)	(0.01)	3.7 ± 1.5 (2-5)	3	(0.002)	1 ± 0 (1-1)							
<i>Amphipolycotyle chloroscombrus</i> Hargis, 1957	g								1	(0.01)	2 ± - (2)							
Table 2 (Cont.)																		
<i>Cernocylocaranga</i> (MacCallum, 1913)	g	27	(52.9)				414 (0.367)	15 ± 13.2 (1-45)	9**	(39.1)	58 (0.036)		23	112	4.8 ± 2.3 (2-10)	14	70	5 ± 4.8 (1-15)
<i>Cernocytenovob oracensis</i> (MacCall um, 1919)	g	3	(5.9)			(5.9)	(0.01)	3.7 ± 1.5 (2-5)	3	(0.002)	1 ± 0 (1-1)							
<i>Cernocytlelaelon gata</i> (Meserve, 1938)	g								1	(0.01)	2 ± - (2)							
<i>Proburstataveraer ucis</i>	g																	
Bravo-Hollis, 1983																		
<i>Pseudobicotyloph ora atlantica</i>	g																	
Arnato, 1994																		
<i>Protomicrocotyle mirabilis</i>	g	15	(29.4)			(29.4)	(0.04)	3 ± 4.1 (1-15)	6	(0.015)	31.2 ± 40 (1-108)							
(MacCallum, 1918)																		
<i>Pseudomazocraes selene</i>	g	6	(11.8)			(11.8)	(0.009)	1.7 ± 0.8 (1-3)	10	(0.229)	33.7 ± 89 (1-301)							
Hargis, 1957																		
<i>Pyragraphoruspyra graphorus</i> (MacCall um&MacCallum, 1913)	g																	
<i>Engraulicola</i> sp.	g																	
CESTODA																		
<i>Dasyrhynchusgiga nteus</i> Diesing, 1850**	h																	
<i>Callitetrarhynchuss p.</i> **	i, m	18	(35.3)			(35.3)	(0.035)	2.2 ± 1.7 (1-6)	6	(0.014)	3.7 ± 3.6 (1-10)							
<i>Nybelinia</i> sp. **	i	7**	(13.7)			(13.7)	(0.01)	1.6 ± 0.5 (1-2)	3	(0.019)	10 ± 9.6 (3-21)							
<i>Scolexpolymorphu s</i> **	i	5	(9.8)			(9.8)	(0.177)	40 ± 86.6 (1-195)	6	(0.014)	5.5 ± 4.8 (1-11)							
Muller, 1788																		
NEMATODA																		
<i>Arisakia</i> sp. **	i	10	(19.6)			(19.6)	(0.045)	5.1 ± 6.3 (1-20)	6	(0.014)	24							
<i>Contracaecum</i> sp. **	i, ic																	

<i>Hysterothylacium</i>																				
<i>frataezae</i> Deardorff & Overstreet, 1980*	i	5** (9.8)	6 (0.005)	1.2 ± 0.4 (1-2)	2** (8.7)	9 (0.006)	4.5 ± 2.1 (2-6)	11** (39.3)	56 (0.126)	5 ± 7.3 (1-7)	1 (4.1)	3 (0.008)	3 ± - (3)							
<i>Hysterothylacium</i>	i																			
<i>liquens</i> Norris & Overstreet, 1975	i							9** (32.1)	89 (0.196)	9.8 ± 9.8 (1-26)										
<i>Hysterothylacium</i> sp. [¶]	i, ic, m	11** (21.6)	27 (0.024)	2.5 ± 1.9 (1-7)	6** (26.1)	12 (0.007)	2 ± 1.3 (1-4)	8** (28.6)	23 (0.051)	2.8 ± 1.9 (1-7)	12** (50)	63 (0.162)	5.2 ± 9.6 (1-35)	4 (0.003)	30 (0.003)	7.5 ± 6.9 (1-14)				
<i>Cucullanus carangis</i> (MacCallum, 1921)*	i	11** (21.6)	22 (0.02)	2 ± 1.2 (1-5)	3** (13.04)	4 (0.002)	1.3 ± 0.6 (1-2)													
<i>Cucullanus trachinoti</i> (Pette & Sey, 1997)*	i																			
<i>Caranginemaameri</i> canum Moravec, Montoya-Mendoza & Salgado-Maldonado, 2008	us				3 (13.04)	54 (0.033)	18 ± 8.2 (11-27)													
Capillariidae	bv	5** (9.8)	9 (0.008)	1.8 ± 1.1 (1-3)																
ACANTOCEPHALA																				
<i>Gorgorhynchoides</i> sp. [¶]	m	1 (1.9)	1 (0.001)	1 ± - (1)																
														2 (10.6)	4 (0.0004)	2 ± 1.4 (1-3)				

d) *Distribution of abundance*

Distribution of abundances of helminth parasites in component communities was analyzed with rank-abundance curves and plotted graphs of the share of individuals of each species of parasite on the total of helminths collected for every community (see Figure 1), and data were adjusted to predictive distribution log-normal for *C. hippos* and *T. carolinus*, and broken-stick to *C. crysos*, *C. chrysurus* and *O. saurus*.

In all communities, some parasites were more abundantly recorded, but these species were dominant only in a particular host community. The three most

abundant species were *H. catarinensis* ($\rho_i = 0.69$) for *T. carolinus*, *M. brachydera* ($\rho_i = 0.51$) for *O. saurus* and *B. margaritae* ($\rho_i = 0.44$) for *C. hippos* (Table 2, Fig. 1). Most abundant species did not exceeded the others, therefore, dominance of these species had no effect on the type of distribution of abundance, because the different communities adapted to the log-normal and broken-stick model, showing that in all community components they have a high proportion of species of median and low abundance, conditions that have effects on the richness of community components.

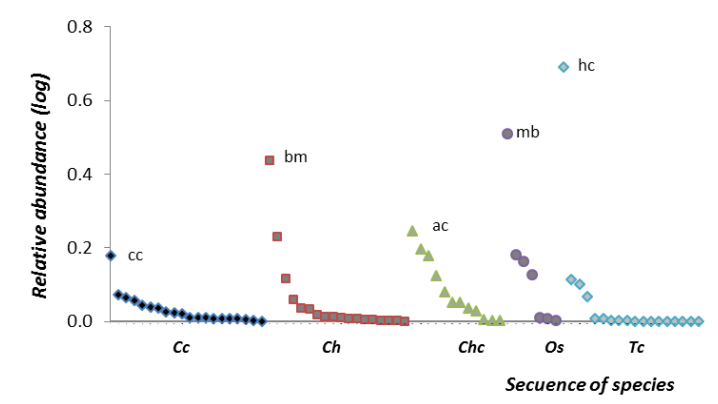


Fig. 1: Dominance-species plot of component communities of five species of Carangids from Alvarado, Veracruz, Mexico. Host: Cc, *C. crysos*; Ch, *C. hippos*. Chc, *C. hrysurus*; Os, *O. saurus*; Tc, *T. carolinus*; Parasites: cc, *C. carangis*; bm, *B. margaritae*; ac, *A. chloroscombrus*; mb, *M. brachydera*; hc, *H. catarinensis*

e) *Component communities and infracommunities*

In the component communities, 388 to 10,184 individual helminths and 7 to 23 parasite species (*S*) were collected. The Shannon diversity index (*H'*) was of 0.07-2.19, and the Berger-Parker dominance index (I_{B-P}) was of 0.24-0.69 (see Table 4).

The infra-communities richness ranged from 1 to 12 species of helminths per fish. Out of all hosts, only seven had no parasites, and all others had from one to 12 species but most often one, two or three species of parasites were found (Table 5). The most frequent co-occurrences between two species of parasites were, *Cucullanus carangis* and *Anisakis* sp., in *C. crysos* (7/51 hosts); *Cemocotyle carangis* and *Pseudomazocraesaelene* in *C. hippos* (6/23 hosts); *Amphipolycotyle chloroscombrus* and *Engraulicolasp.*, in *C. chrysurus* (13/28 hosts); *Hargicola oligoplites* and *Probursatavera crucis* in *O. saurus* (11/24 hosts); *Huerleytremashorti* and *Lobatostomaringens* in *T. carolinus* (8/19 hosts). The average number of parasites species per individual host was 2.7 ± 1.5 to 4.5 ± 2.1 , while the average number of helminth individuals per host was 16.2 ± 19.4 to 536 ± 1106 . The value of the Brillouin's index for each infracommunity ranged from 0.1-1.54 to 0.54-1.73 with average values of 0.66 ± 0.44 to 1.03 ± 0.32 , for indexes such as evenness and dominance see Table 6.

Table 4: Descriptive parameters of component communities of five carangid species from Alvarado, Veracruz, Mexico. #th, no total helminth; *S*, richness; *H'*, Shannon diversity index; *J'*, Equitativity index; I_{B-P} , Berger-Parker dominance index; *spd*, specie dominante. cc, *C. carangis*; bm, *B. margaritae*; ac, *A. chloroscombrus*; hc, *H. catarinensis*.

Host	<i>n</i>	#th	<i>S</i>	<i>H'</i>	<i>J'</i>	I_{B-P}	<i>spd</i>
<i>C. crysos</i>	51	1126	21	2.19	0.71	0.36	cc
<i>C. hippos</i>	23	1620	18	1.76	0.61	0.43	bm
<i>C. chrysurus</i>	28	455	12	2.01	0.8	0.24	ac
<i>O. saurus</i>	24	388	7	1.31	0.67	0.5	mb
<i>T. carolinus</i>	19	10184	18	1.07	0.37	0.69	hc



Table 5: Frequency of parasites species per host in five species of carangids from Alvarado, Veracruz, Mexico.

Host	Number of parasite species per host												
	0	1	2	3	4	5	6	7	8	9	10	11	12
<i>C. crysos</i>	3	10	10	9	2	5	3	1	4	2	2		
<i>C. hippos</i>	2	2	5	5	2	3	1		1	1			1
<i>C. chrysurus</i>	2	2	5	5	2	3	1		1	1			
<i>O. saurus</i>		8	4	2	7	3							
<i>T. carolinus</i>		2	1	3	3	5	1	3		1			

Table 6: Infracommunities of five species of carangids from Alvarado, Veracruz, Mexico. Data include: S, Richness helminth species; $\bar{x}S$, average helminth species; $\bar{x}n$, average number of helminth; $\bar{x}H$, average Brillouin index; $\bar{x}J'$, average evenness index; $\bar{x}J_{B-P}$, average Berger-Parker index.

	<i>C. crysos</i>	<i>C. hippos</i>	<i>C. chrysurus</i>	<i>O. saurus</i>	<i>T. carolinus</i>
S	21	18	12	7	19
$\bar{x}S \pm ES$	3.8 ± 2.7	4.1 ± 2.8	3.6 ± 1.5	2.7 ± 1.5	4.5 ± 2.1
Range	1-10	1-12	1-7	1-5	1-9
n	1127	1620	455	388	10184
$\bar{x}n \pm ES$	23.5 ± 35.5	77.1 ± 158	17 ± 13	16.2 ± 19.4	536 ± 1106
Range	1-224	1-640	2-54	1-84	10-4883
$\bar{x}H \pm ES$	1.01 ± 0.44	0.85 ± 0.4	1.03 ± 0.32	0.86 ± 0.32	0.66 ± 0.44
Range	0.27-2.1	0.1-1.54	0.54-1.73	0.28-1	0.14-1.41
$\bar{x}J' \pm ES$	0.75 ± 0.2	0.67 ± 0.25	0.83 ± 0.1	0.7 ± 0.2	0.43 ± 0.27
Range	0.27-1	0.1-1	0.63-1	0.29-1	0.11-0.88
$\bar{x}J_{B-P} \pm ES$	0.7 ± 0.2	0.67 ± 0.2	0.6 ± 0.2	0.8 ± 0.2	0.7 ± 0.2
Range	0.2-1	0.3-1	0.2-1	0.3-1	0.3-1

f) Similarity among component communities

Among component communities, the highest similarity record was for communities of *C. crysos* and *C. hippos*, a rate near 60% ($I_j = 0.56$). Among community components, the highest similarity was observed for communities of *C. crysos* and *C. hippos*,

with an index near to 60% ($I_j = 0.56$), because they have 14 species of parasites in common, but for other communities this value was below 25% (see Figure 2), where they only have some larvae of parasites roundworms and tapeworms in common (see Table 2).

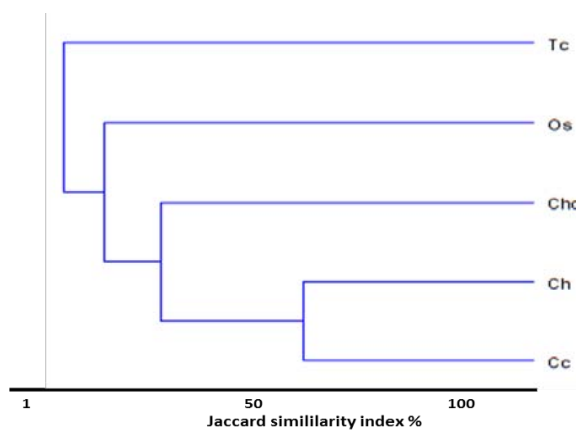


Fig. 2: Cluster of similarity of taxonomic composition of component communities of five species of carangids from Veracruz. Data include: Host: Cc, *C. crysos*; Ch, *C. hippos*. Chc, *C. hrysurus*; Os, *O. saurus*; Tc, *T. carolinus*

IV. DISCUSSION AND CONCLUSIONS

Parasite helminths of Carangidae fish have been widely studied in the Gulf of Mexico and the Caribbean Sea (Nahhas & Powell 1971; Hendrix 1994; Pérez-Ponce de León et al. 2007; Overstreet et al. 2009), with registers for all five helminth groups. However, this

study adds new host and location records (Table 2). Groups with the highest number of species were trematodes and nematodes in four host species, excepting *C. hippos* that included monogeneans. As related to the range of species per helminth group, these results are similar to records in other carangids in Mexico (Sánchez-Ramírez & Vidal-Martínez 2002;

Aguirre-Macedo et al. 2007) and Brazil (Takemoto et al. 1995; 1996; Luque & Alves 2001). It has been stated, generalizing, that trematodes and nematodes are the most numerous species in marine fish (Rohde & Heap 1998; Zander et al. 1999), including those from tropical (Moravec et al. 1997; Sabas & Luque, 2003; Luque & Poulin, 2007), and temperate latitudes (Campos & Carbonell 1994; Zander et al. 1999; Madhavi & Sai Ram, 2000). This condition was also observed in hosts included in this study.

On the other hand, records of cestodes, nematodes and some digeneans larvae suggest the importance of carangids as intermediate hosts of these parasites, highlighting their relevance in the food chain of studied hosts that can be infected by parasites, including ichthyophagous fish, birds or mammals, which participate as final hosts (Chaves & Luque 1999; Luque & Alves 2001; Sánchez-Ramírez & Vidal-Martínez 2002), i. e., sharks, completing life-cycles e.g., *Callitetrarhynchus* sp., *Contraecaecum* sp., and *Dasyrhynchus* sp., strengthening the relevance of carangids in the transmission mechanisms of these helminths (Overstreet, 1978; Deardoff & Overstreet 1981; Sánchez-Ramírez and Vidal-Martínez 2002; Aguirre-Macedo et al. 2007).

As related to the correlation among the parasites size, richness and abundance, it is generally considered that larger hosts have higher richness and abundance (Kennedy et al. 1986; Holmes 1990; Bush et al. 1990), as we found in larger hosts, such as *C. crysos* and *C. hippos*, associated to a higher possibility of infection, higher vagility and contact with intermediate infected hosts (Poulin et al. 2003; Poulin & Mouillot 2003), enhancing the parasites' life-cycle (Luque & Poulin, 2008; Muñoz et al. 2006).

As related to the abundance distribution types, log-normal and broken-stick models were adjusted, frequent distribution types for helminth communities parasitizing marine fish (Poulin & Justice, 2008), with parasite species with relative abundances from low to medium, without significant dominance. It was observed, in both distribution types, that parasites with the highest abundance never exceeded one half of the total abundance (relative abundance < 0.5), excepting *H. catarinensis*, with prevalence of 70% and abundance of 7033 worms in the *T. carolinus* community. The similitude analysis revealed that carangids have and share typical helminth fauna, as the highest similitude was observed among sympatric species living in this area, such as *C. crysos* and *C. hippos*, as described for lutjanids in the same zone (Montoya-Mendoza et al. 2014; 2016).

Finally, it has to be noted that parasitic relations of hosts in wild populations, with biological, commercial and food relevance, and high farming potential, as those of carangids (Hutson et al. 2007), pose no zootic risk, excepting *Anisakis* larvae, and that richness and

diversity found for parasite helminth community components and infracommunities in hosts studied, are similar to those reported for carangids on the West Atlantic coast, such as *T. carolinus* ($S = 18$, $\bar{X}S = 6 \pm 2$, $\bar{X}H = 0.33 \pm 0.28$) (Sánchez-Ramírez & Vidal-Martínez 2002); *C. hippos* ($S = 16$, $\bar{X}S = 3 \pm 2$, $\bar{X}H = 0.55 \pm 0.4$) and *C. latus* ($S = 14$, $\bar{X}S = 2.9 \pm 1.6$, $\bar{X}H = 0.6 \pm 0.35$) (Luque & Alves 2001); *O. palometa* ($S = 13$, $\bar{X}H' = 0.79 \pm 0.38$), *O. saurus* ($S = 11$, $\bar{X}H' = 0.9 \pm 0.38$) and *O. saliens* ($S = 9$, $\bar{X}H' = 0.81 \pm 0.42$) (Takemoto et al. 1996); *Selene setapinnis* ($S = 18$, $\bar{X}S = 3.2 \pm 2.2$, $\bar{X}H = 0.32 \pm 0.15$) (Cordeiro & Luque 2004), including that parasite communities in five carangids are as rich and diverse as those of marine hosts in temperate (Châari et al. 2015) and tropical zones (Luque & Poulin 2007; Madhavi & Triveni Lakshmi, 2012).

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Assessment of the Invasive Alien Plant Species *Lantana Camara* in Nile River Millennium Park, Bahir Dar, Ethiopia

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Keywords: *assessment, invasive alien species, lantana camara, nile river millennium park.*

GJSFR-C Classification: FOR Code: 069999



ASSESSMENT OF THE INVASIVE ALIEN PLANT SPECIES LANTANA CAMARA IN NILE RIVER MILLENNIUM PARK BAHIR DAR ETHIOPIA

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Assessment of the Invasive Alien Plant Species *Lantana Camara* in Nile River Millennium Park, Bahir Dar, Ethiopia

Taye Birhanu Belay¹ & Ashenafi Ayenew Hailu²

Abstract- *Lantana camara* is the major invasive alien species in Nile River Millennium Park and it is introduced for ornamental purpose to the park intentionally. It reproduces by seed, root and vegetative multiplication, and disseminated by animals particularly birds and human intentional gardening are the major spreading mechanisms. Local communities are highly frustrated by the negative impacts of *Lantana camara* which includes competing animal feed, biodiversity loss, closing roads, and hiding harmful wild animals like rodents, snakes, mongoose, etc. According to the present study *Lantana camara* rate and scale of invasion has been increasing at an alarming rate affecting socio-economic status of the community and adverse effect to the biodiversity at large since its introduction. This is because *Lantana* spreading mechanisms are in multiple ways such as local human activity, by animals particularly birds, wind and its prolific seed production and easy dispersal, and multiplication nature. This, therefore, needs for an integrated, coordinated and multi-stakeholder and multiple level actions that the community, government and development partners shall participate in the eradication of the invasive plant *Lantana camara*. This would require the restriction of further spread of *Lantana camara* into non invaded areas, restriction use of *Lantana* in gardens and strategically controlling infestations by mechanical mechanism.

Keywords: assessment, invasive alien species, *lantana camara*, nile river millennium park.

I. INTRODUCTION

Invasive Alien Species' (IAS) are defined by the CBD (Convention on Biological Diversity) as species, subspecies or lower taxa, (including any part, gametes, seeds, eggs, or propagules of such species), introduced outside their natural past or present distribution and threaten biological diversity. Invasive alien species seriously affect many sectors of the economy; especially they are noted for being an important cause of global biodiversity loss. The impacts of alien invasive species on biodiversity have been described as "immense, insidious and usually irreversible [5].

Invasive alien species are increasingly becoming a serious environmental and development challenge in Ethiopia. Although, there is no complete account of the cost of IAS, rural communities have to endure tremendous economic and social hardships due to IAS. Many communities have lost productive assets through degradation of the natural resource base including pasture land, arable lands, plant species of medicinal, food and feed values, while many other have suffered from physical displacement and crowding into ever shrinking land area resulting in loss of ecosystem services. Invasive Alien Species has been colonizing many ecosystems of the country. Consequently, the biodiversity of Ethiopia is under increasing threat from IAS (IAS policy and strategy). The main invasion routes are: introduction by chance (unintentionally) and introduction by hand (intentional introduction of horticultural, medicinal, silvi cultural or agricultural plants for economic purpose) [3, 8].

Lantana section *Camara* is native only to the Americas, with members occurring from Florida and Texas in the north to northern Argentina and Uruguay in the south. While it is recognized that the weedy taxa of *lantana* naturalized in the Old World are of hybrid origin, and so do not have a 'native' range *per se*, the hybrids are almost certainly derived from various species within the section *Camara* that originated in the Americas [2].

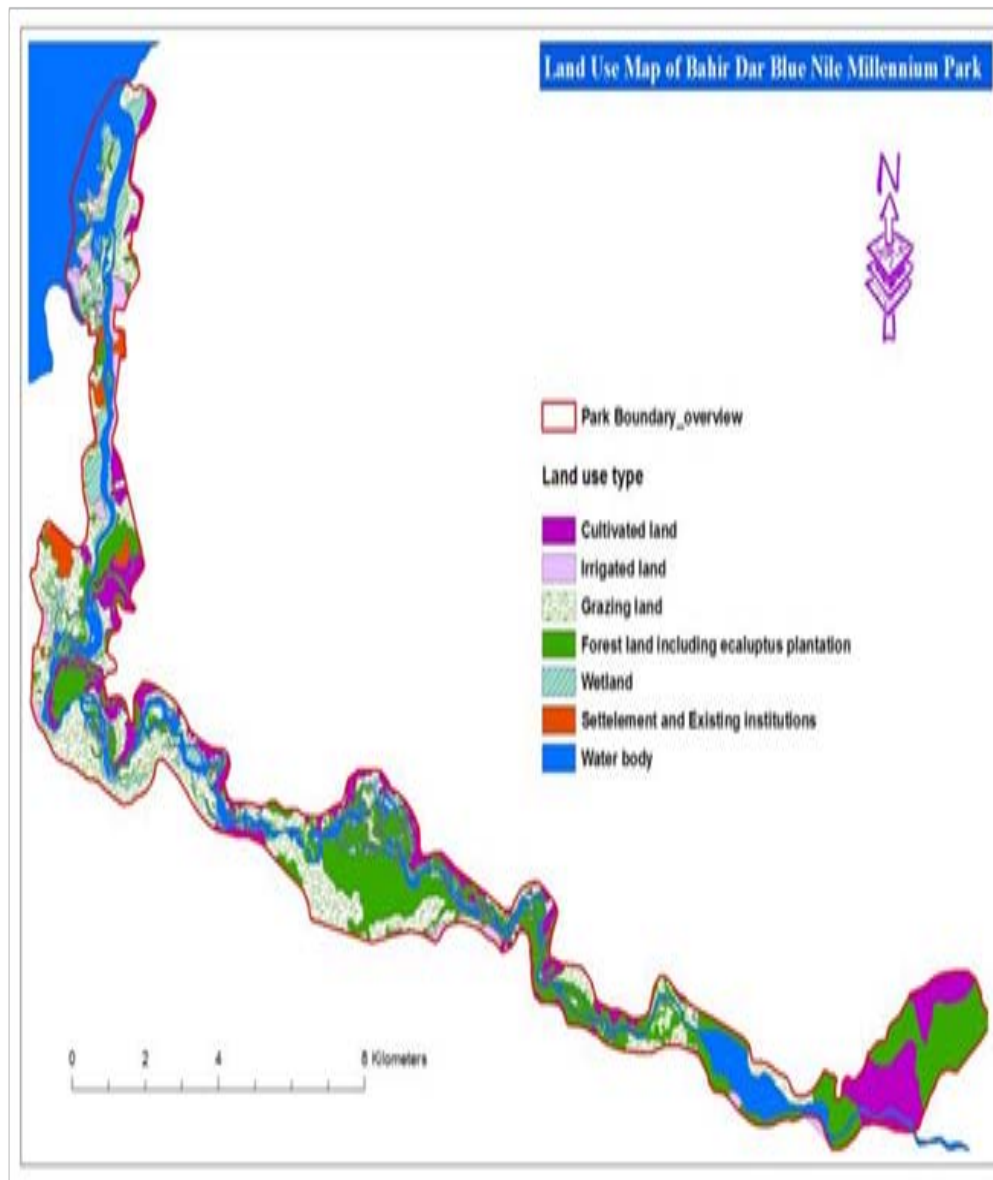
L. camara is considered a problem weed in many of the countries to which it has been introduced. It flowers prolifically and the seeds are dispersed by birds [10]. The plants can grow in individual clumps or as dense thickets, crowding out more desirable species. In disturbed native forests, it can become the dominant understorey species, disrupting succession and decreasing biodiversity. Its allelopathic qualities can reduce vigour of plant species nearby and reduce productivity in orchards [4]. It can affect agriculture by outcompeting native pastures by interfering with the mustering of cattle, and by causing death of stock by poisoning [10]. However, no adequate recent information exists about the impacts, distribution pattern, trend, status and controlling mechanisms of this species in the study area. Therefore, this research aims at assessing the impacts, distribution pattern, trend, management practices and statuses of *L. camara* in Bahir Dar Nile River Millennium Park.

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II. MATERIALS AND METHODS

a) Description of the study area



Source: Nile River Millennium Park office

Fig. 1: Land use map of Nile River Millennium Park

The geographic location of Abay riverside park (Nile River Millennium Park) is 11°29'40.2"N to 11°37'27.9" N latitudes and 37°24'37.2" E to 37°36'34.0" E longitudes starting at the source of Blue Nile out late from Lake Tana to the famous river fall of Tis Abay Fountain.

b) Method of Data Collection

Field study on the impacts, trends, mode of entry, spread, status, distribution and management practices of *Lantana camara* in Nile River Millennium Park, Bahir Dar, Amhara region was conducted. The study area was selected purposively on the basis of the level of *L. camara* invasion with the help of information

obtained from Environment and Forest office of Amhara region.

Based on the above selection criteria, 20 households from nearby kebeles of the Nile River Millennium Park were randomly selected. Accordingly, Hidar 11, Wereb kola tsion and Park Communities were selected to conduct this research.

Data was collected from primary sources and it has been collected through Rapid Ethno botanical Appraisal (REA) to obtained needed information quickly and inexpensively in a very short period of time. The primary data was collected using semi-structured interviews, discussions and direct field observations.

Secondary source of data was obtained from the agricultural, and Environment and forest office of the region, from different books and journals.

c) *Method of Data Analysis*

The collected data was analyzed by using SPSS (statistical package for social sciences). A descriptive statistical method was employed to analyze and summarize the data and to calculate percentages and frequency.

III. RESULTS AND DISCUSSIONS

a) *Invasive alien species and level of Lantana camara in the study area*

All the respondents with no variation (100%) indicated *L. camara* is the only invasive plant species in

their locality and they also indicated that *L. camara* infestation has been increasing at an alarming rate since its introduction (Figure. 2). Based on our field observation, the respondents view is quite genuine that *L. camara* makes dense and abundant thickets in Nile River Millennium Park. Not only is the geographic range of lantana still expanding in many areas, but the density of infestations within its range is increasing and it grows impenetrable thickets that can suppress the growth of native species. Similar report indicates that due to its prolific nature of flowering and dispersal, the species tends to alter the structure of the terrestrial ecosystem by gregarious presence. The species forms dense thickets and tends to eliminate the native species [7].

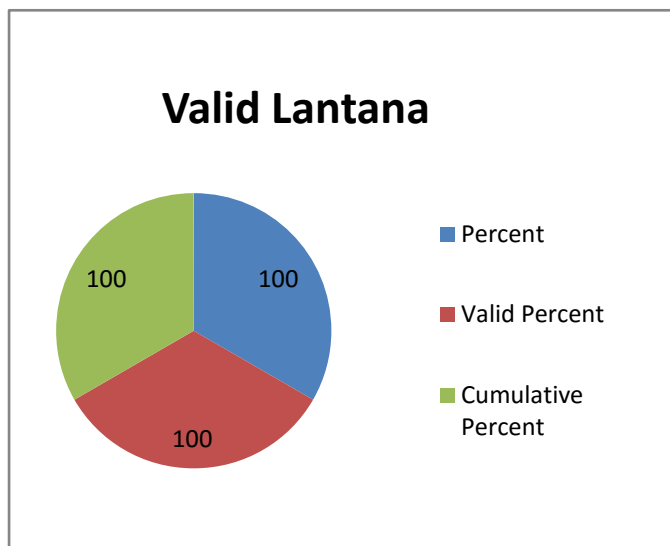


Fig. 2: List of Invasive Alien Species in the study area

b) *Modes of Introduction of Lantana camara*

The invasive plant *L. camara* is introduced intentionally. The majority of respondents (70%) indicated *L. camara* has been planted in the Bezawit palace intentionally for ornamental purpose in 1985. Bezawit palace is Emperor Hailesilassies palace where it is located inside of the Nile River Millennium Park. According to the respondents, Lantana was brought from Awash Melkasa palace by Mr. Alebel Kassa, the local administrator of Bezawit Palace, who he planted as one of the ornamental species in and around the palace. He had been an employee of Awash Melkasa palace, and he brought his garden plant, *Lantana camara* to his new work place. Some of the respondents (25%) say it is introduced by birds, others (5%) say they don't know how *L. camara* has been introduced [Fig 3]. Similar report by [1, 9] on *L. camara* reported that it is introduced intentionally to Ethiopia for ornamental purpose and also it is introduced intentionally and has covered all the park area, highly spreading and destroying the native biodiversity, converting the

beautiful attractive riverine park into homogenous and less attractive sight.

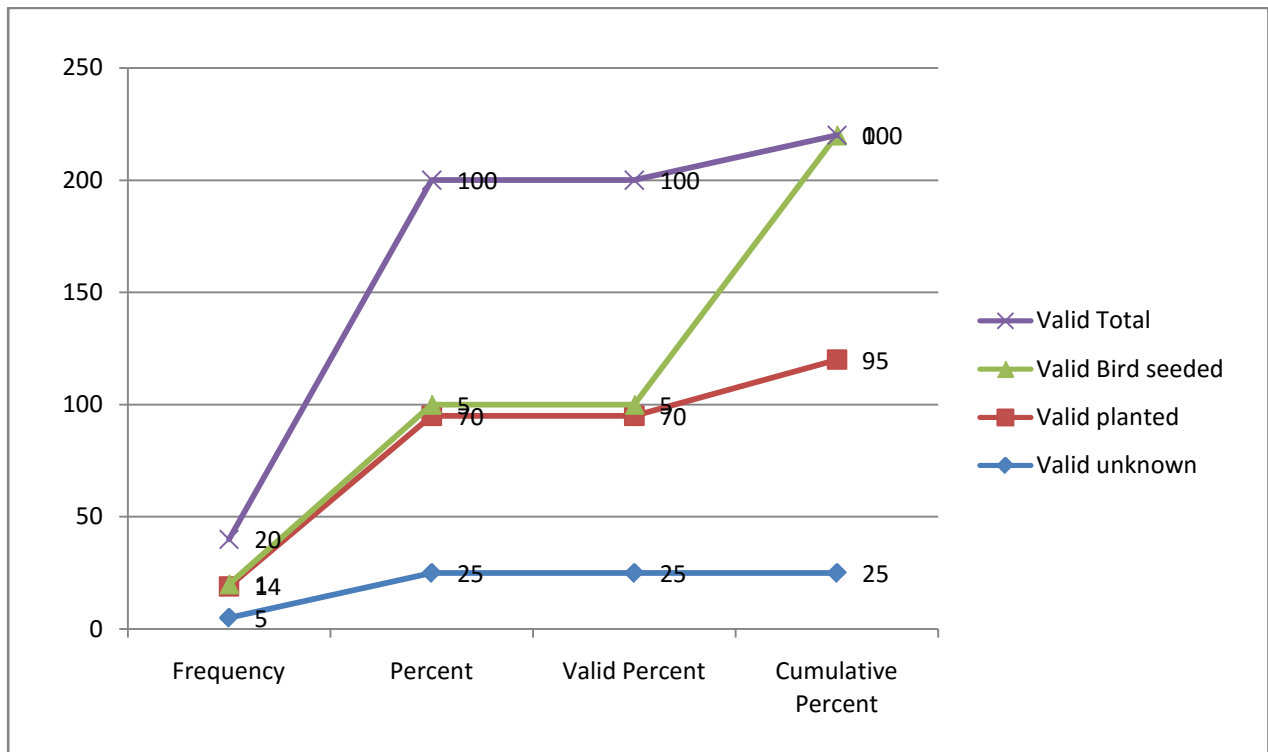


Fig. 3: Modes of introduction to the park

c) *The Modes of Dispersal of Lantana camara*

The majority of respondents (55%) indicated modes of dispersal of the invasive plant *L. camara* is by means of animals particularly birds and 15% of respondents say prolific seed production, easy dispersal, and layering are the mechanism for its high dispersal. Some (10%) say both birds and human intentional planting are the root mechanism of fast and wide dispersal. According to the respondents birds pass the seed in their droppings, potentially spreading it over

quite large distances. Others (5%) say human intentional planting is the root cause of its dispersal [Table 1]. Similar report by [2] indicates that Seeds are widely dispersed, predominantly by birds, but also by kangaroos, bearded dragons, sheep, goats, cattle, foxes, jackals, monkeys and possibly rodents. Fruit dispersal is through frugivorous birds, fox and rodents. Germination rate of fresh seed is generally low, but the germinability gets improved when the seed passes through the digestive system of birds and animals [11].

Table 1: Modes of dispersal for *Lantana camara*

	Frequency	Percent	Valid Percent	Cumulative Percent
animals(birds)	11	55.0	55.0	55.0
human	1	5.0	5.0	60.0
Valid seed multiplication	3	15.0	15.0	75.0
bird and human	3	15.0	15.0	90.0
bird and wind	2	10.0	10.0	100.0
Total	20	100.0	100.0	

d) *Highly Invaded areas in the study area*

L. camara is invading all land use. The Lion share of respondents (55%) say in their locality, the highly invaded area is grazing land while 35% of respondents informed protected area of Nile River Millennium Park is highly invaded area. The remaining 5% of respondents say *lantana camara* is invading the agricultural land. Others (5%) say *lantana camara* is invading grazing land, agricultural land and the adjacent areas of Nile River Millennium Park [Fig 4].

This is in agreement with our observation that all the areas were highly invaded by *L. camara* and in agreement with the report by [9, 11,] which states the relative abundance and invasion pattern of *L. camara* is very high in grass land followed by in the cultivated land and natural forest respectively.

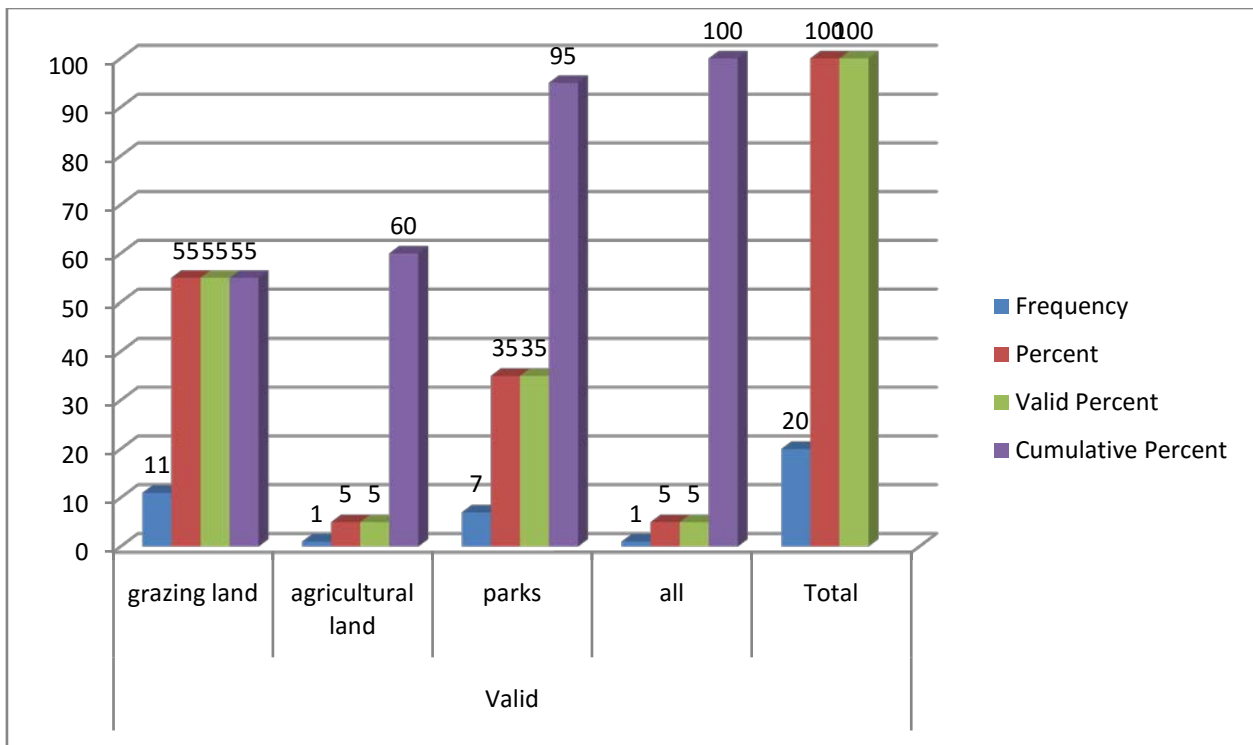


Fig. 4: Highly invaded areas in the study area

e) *Negative Impact of Lantana camara*

The effects of IAS on biodiversity have been described as “immense, insidious and usually irreversible [5]”. All informants confirmed that *Lantana* posed a very serious problem to their livelihood. They stated numerous negative effects of *L. camara* [Fig 5]. According to the majority respondents (50%), the invasive alien plant *L. camara* has a negative impact on

biodiversity in general and particularly affects animal forages, closes roads and hide harmful wild animals like snake, rodents, Egyptian mongoose [Fig 5]. Similar report by [2] indicates that once established in pastures, *lantana* forms large, impenetrable thickets, outcompeting valuable pasture species, blocking the movement of domestic stock to waterholes, poisoning stock and interfering with mustering.

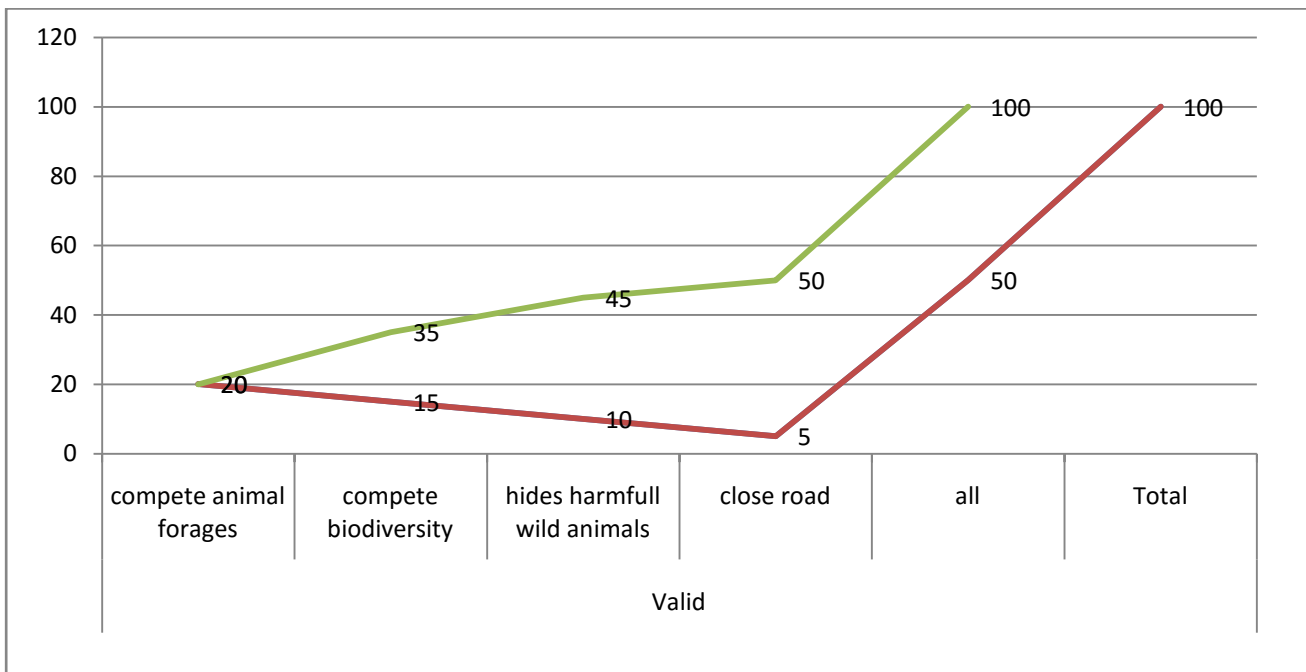


Fig. 5: Negative Impacts of *Lantana camara*

f) Control and/or Eradication Methods Introduced

L. camara is causing huge economic damage and biodiversity loss in Nile River Millennium Park. While having vast horizons of both ecological and environmental significance in the park, concerted control efforts are still very limited that majority of respondents (45%) indicated no management efforts have been made so far in the park. Some (35%) say mass cutting of *L. camara* was practiced and others (15) say both cutting and hand pulling was used to eradicate *L. camara* while the remaining (5%) say hand pulling was

practiced to manage *L. camara* [Fig 6]. If coordinated controlling mechanism had been used, it would have been effective that similar study indicated manual removal of plants minimizes disturbance to nearby vegetation and is effective in killing the plants, especially those in small, isolated clumps growing along fence lines or in public parks. Manual uprooting of lantana plants is labor intensive and costly but is often the only method available to farmers in developing countries [11].

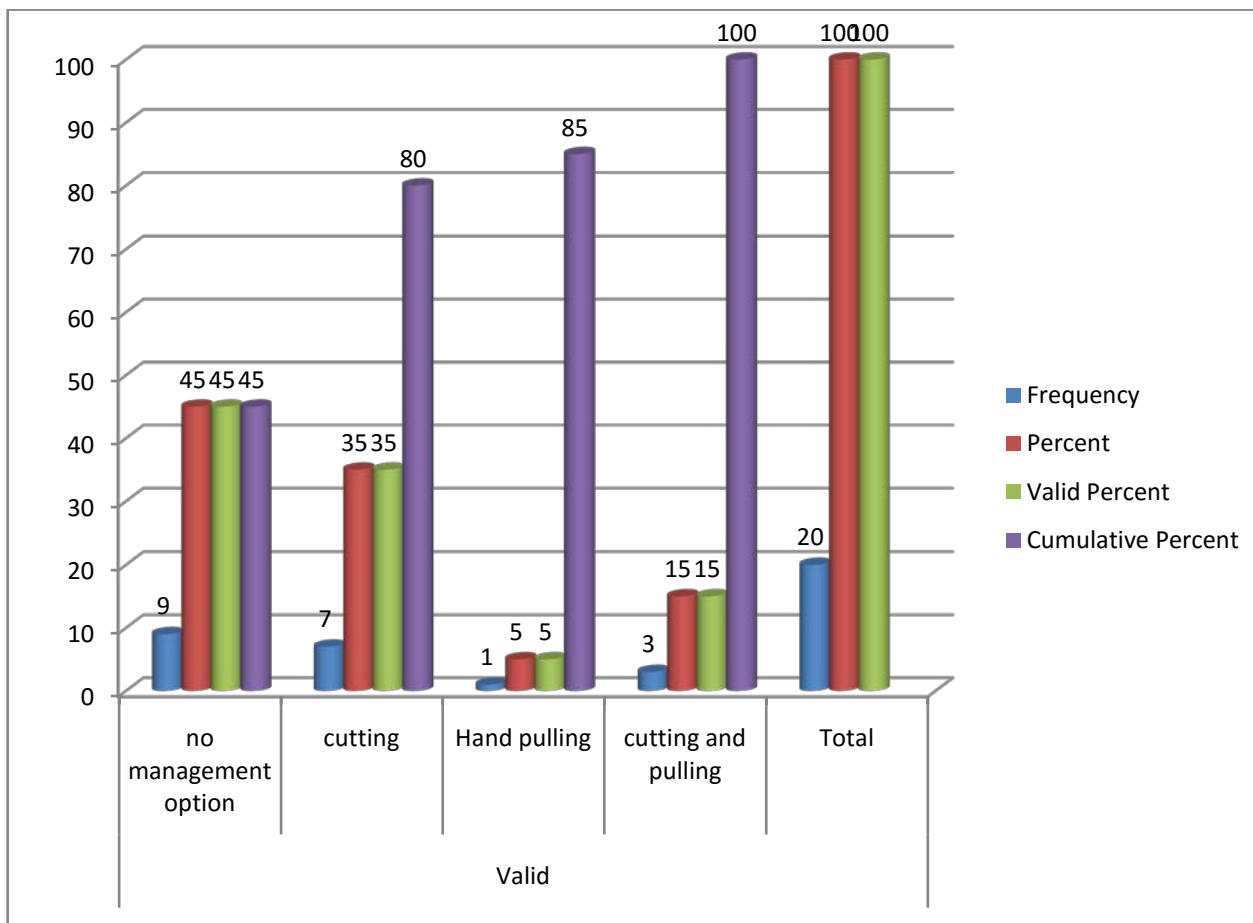


Fig. 6: Control and/or Eradication Methods Introduced

g) Suggested future actions

The majority of respondents (45%) suggested that hand pulling and burning is effective management option while 35% of respondents view is the use of chemical is the best option to manage *L. camara*. Some (15%) say combination of biological chemical and physical methods is effective management options. Others (5%) say biological control mechanism is effective technique to manage the dense *L. camara* in the study area [Fig 7].

of plants minimizes disturbance to nearby vegetation and is effective in killing the plants, especially those in small, isolated clumps growing along fence lines or in public parks. Mechanical clearing and hand pulling are suitable for small areas and fire can be used over large areas [2, 12].

The informants' perception is real that mechanical removal, using either modified bulldozers or ploughing, removes standing plants. Clearing by tractor or stick-raking is considered superior to burning when dealing with mature lantana plants [2]. Manual removal

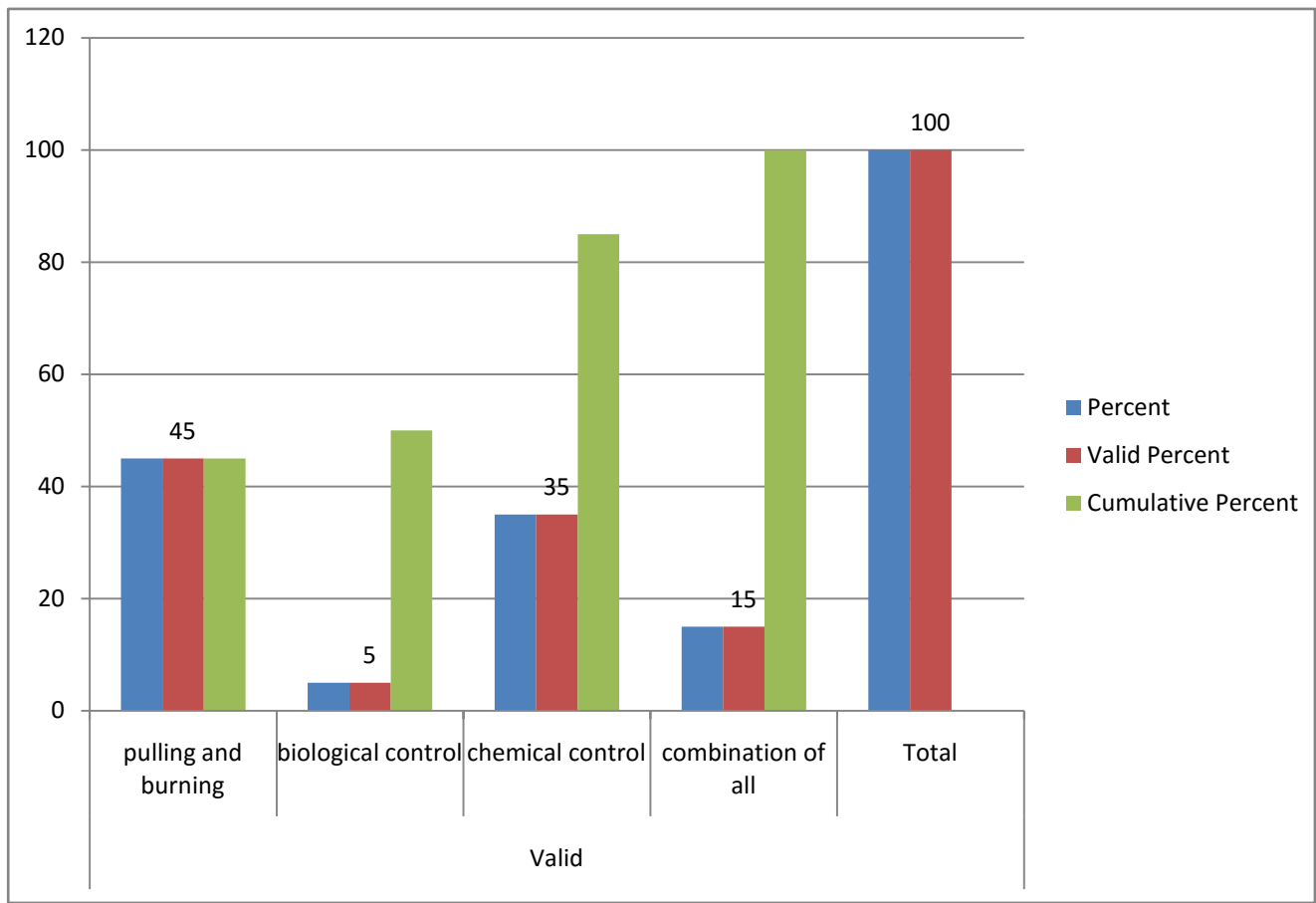


Fig. 7: Suggested future actions

h) Trends in the spread of *Lantana camara* and Major role player

According to the respondents (100%), the level of infestation in the study area has been increasing extremely and the management effort made so far were ineffective, irregular and poor. All the respondents (100%) calls for an integrated, coordinated and multi-stakeholder and multiple level actions that the community, government and development partners shall

participate in the eradication of the invasive plant *L. camara*.

Similar report by [13] describe that the major players, in management of Invasive Alien Species are the community, government and development partners. Thus it needs the coordinated measures to address invasive alien species and protect and conserve the country's biological diversity and agricultural production as well as the health of wildlife and humans, is essential.



Fig. 8

IV. CONCLUSION AND RECOMMENDATION

L. camara is the major invasive Alien species in Nile River Millennium Park and it was introduced for ornamental purpose to the park intentionally. While, Nile River Millennium Park has biodiversity conservation, water and soil conservation significance, and the ecological balance of the surrounding area of the river where the Nile River emanates from Lake Tana up to the surrounding area of Tis Abay Fountain is mostly invaded in dense stands of Lantana, the capacity of the soil to absorb rain is lower than under good grass cover. This could potentially increase the invasion impact to serious loss of biodiversity, increase the amount of run-off and the subsequent risk of soil erosion, affects animal forages, affect many sectors of the economy of local communities; especially it hide harmful wild animals like snake, rodents, Egyptian mongoose and closes roads.

The means of spread of *L. camara* are multiple, including animals particularly birds, prolific seed production and easy dispersal, layering and human intentional planting is the root mechanism of fast and wide dispersal. This, therefore, it needs for an integrated, coordinated and multi-stakeholder and multiple level actions that that the community, government and development partners shall participate in the eradication of the invasive plant *L. camara*. This would require the restriction of further spread of *L. camara* into non invaded areas, restriction use of Lantana in gardens and strategically controlling infestations by mechanical mechanism such as hand pulling and burning, Stickraking, bulldozing, ploughing, grubbing, Hand cutting using brush cutters, and maintenance control involves use of techniques in a coordinated manner on a continuous basis in order to maintain *Lantana* populations at the lowest acceptable level.

V. ACKNOWLEDGMENTS

We are grateful to Ethiopian Biodiversity Institute (EBI) for financial support during field work. We are also grateful to worker of Nile River Millennium Park office who helped us in different ways. Finally, we are indebted very much to Hidar 11 and Wereb kola tсион kebeles farmers and Park Community farmers for their unreserved willingness to share their time and knowledge with us.

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Population Estimate, Group Size and Age Structure of the Gelada Baboon (*Theropithecus Gelada*) around Debre-Libanos, Northwest Shewa Zone, Ethiopia

By Kassahun Abie & Afework Bekele

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Abstract- Most of the gelada baboon's population which is endemic to Ethiopia exists on the gorges and mountain cliffs of the country including Debre-Libanos. The objective of the study was to estimate the total gelada baboons, and determine their group size and age structure. This was investigated based on direct field observation method from August 2012 to March 2013. The study area was divided into seven counting blocks (Shinkurt Mikael, Chagel, Amanuel, Abba Dinkona, Wusha Gedel, Tekle Haimanot, and Set Debre). Data were analyzed using descriptive statistics. Chi-square test was used to compare the sex and age ration, and their distribution among the counting blocks. The population size of gelada baboons was determined from direct total count. The average number of gelada baboons counted was 1608. Of these, adult males comprised 162, adult females 576, sub-adult males 121, sub-adult females 231, young 307 and infants 212. There was a significant difference among the different age groups of the total individuals counted in the study area ($\chi^2=1002.657$, $df=5$, $p<0.05$).

Keywords: *gelada baboon, population estimate, group size, debre-libanos.*

GJSFR-C Classification: FOR Code: 060799



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Kassahun Abie ^α & Afework Bekele ^σ

Abstract- Most of the gelada baboon's population which is endemic to Ethiopia exists on the gorges and mountain cliffs of the country including Debre-Libanos. The objective of the study was to estimate the total gelada baboons, and determine their group size and age structure. This was investigated based on direct field observation method from August 2012 to March 2013. The study area was divided into seven counting blocks (Shinkurt Mikael, Chagel, Amanuel, Abba Dinkona, Wusha Gedel, Tekle Haimanot, and Set Debre). Data were analyzed using descriptive statistics. Chi-square test was used to compare the sex and age ration, and their distribution among the counting blocks. The population size of gelada baboons was determined from direct total count. The average number of gelada baboons counted was 1608. Of these, adult males comprised 162, adult females 576, sub-adult males 121, sub-adult females 231, young 307 and infants 212. There was a significant difference among the different age groups of the total individuals counted in the study area ($\chi^2=1002.657$, $df=5$, $p<0.05$). There was unequal sex ratio in the individual count. The highest range of group size was recorded during the wet season (5-187 individuals) with the mean group size of 28.21; while the dry season, the range of group size was smaller (3-120 individuals) with the mean group size of 18.4

Keywords: *gelada baboon, population estimate, group size, debre-libanos.*

1. INTRODUCTION

East Africa is rich in biodiversity and abundance of large mammals (Kutilek, 1979). The extensive tropical and subtropical savanna biome provides the homeland for the variety of mammals. One of the reasons that made the African fauna so interesting and spectacular is the high degree of endemism (Hirst, 1975).

Ethiopia is internationally recognized as one of the most important conservation spots because of a great diversity of its natural ecosystem and biogeographically isolated highlands that support high species endemism. Topography ranging from 110

meters below sea level at Kobar Sink of Afar depression to a peak of 4620 meters above sea level at Ras Dashen Mountain contributes for the availability of the large diversity of ecological conditions. The existence of diverse and varied species of wildlife reflects the diversity in climate, vegetation and terrain. The highland regions, although possess fewer species than lowland parts of the country, have large number of endemic species, particularly birds, mammals and amphibians (Yalden, 1983; Kingdon, 1997). The wide range of habitats in Ethiopia, from arid desert, open grassy steppe, and semi-arid savannas to highland forests and Afro-alpine moorlands have a great positive impact on the country's biodiversity richness (Hillman, 1993).

Due to increase in human population at an alarming rate, the natural resources and ecosystems of Ethiopia have been altered. The large areas of Ethiopian lowland and highlands are changed into agricultural and pastoral lands. The vegetation is overused for fuel wood, construction, timber production and other purposes. As a result, wildlife resources of the country are now largely restricted to a few protected areas and inaccessible areas (Hillman, 1993). Most of the gelada baboon population occurs on the Ethiopian plateau, gorges and mountain cliffs which are inaccessible but with low intervention of humans. Their number is estimated around 50,000 to 60,000 individuals and the number is declining (Dunbar, 1998). The highest density of gelada baboons, and the only place where they are officially protected, occurs in the Simien Mountains National Park, especially in Sankaber and Gich areas of the Park (Beehner *et al.*, 2008). However, small populations of gelada baboons also occur in Menz (Guassa), Debresina, Wollo, and Debre-Libanos. An additional isolated population is located in the south of the Rift Valley in Arsi Province (Mori and Belay, 1990; Oates, 1996).

Even though, the existence of gelada baboon population was recorded in the northwest Shewa zone of Oromia Regional State, no research has been conducted on the population status, group size and structure of gelada baboon and other relevant issues about the species in the area. The intension of this

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research was to fill this gap and provide relevant information for local and regional administration and other conservation organizations.

II. METHODOLOGY

a) Study area

The present investigation was conducted at Debre-Libanos area, which is located in the central highlands of Ethiopia. Its geographical coordinates are 9° 43' 0" North, and 38° 52' 0" East. Debre-Libanos is found in the Oromia Regional State, within the Northwest Shewa zonal administration (Fig. 1). It is located at 104 km away from the capital city, Addis Ababa in the northwest direction, 16 km away from the zone capital (Fiche). Debre-Libanos community conservation area is designed to conserve and manage biodiversity and

wildlife. The area is also a home for a variety of wild animals including spotted hyena, anubis baboon, warthog, vervet monkey, columbus monkey, gelada baboon, different species of birds and others. It characterized by heterogeneous landscape, flora, fauna and habitat types. The area has extremely steep escarpments leading up to a strip of plateau. It is found in the altitude ranges between 2150 to 2650 meters above sea level.

It has bi-modal rainfall pattern ranging from 800 mm to 1200 mm with five months of rain (May-September). The dry season is from December to March. The annual average maximum and minimum temperature of the study area is 23°C and 15°C, respectively.

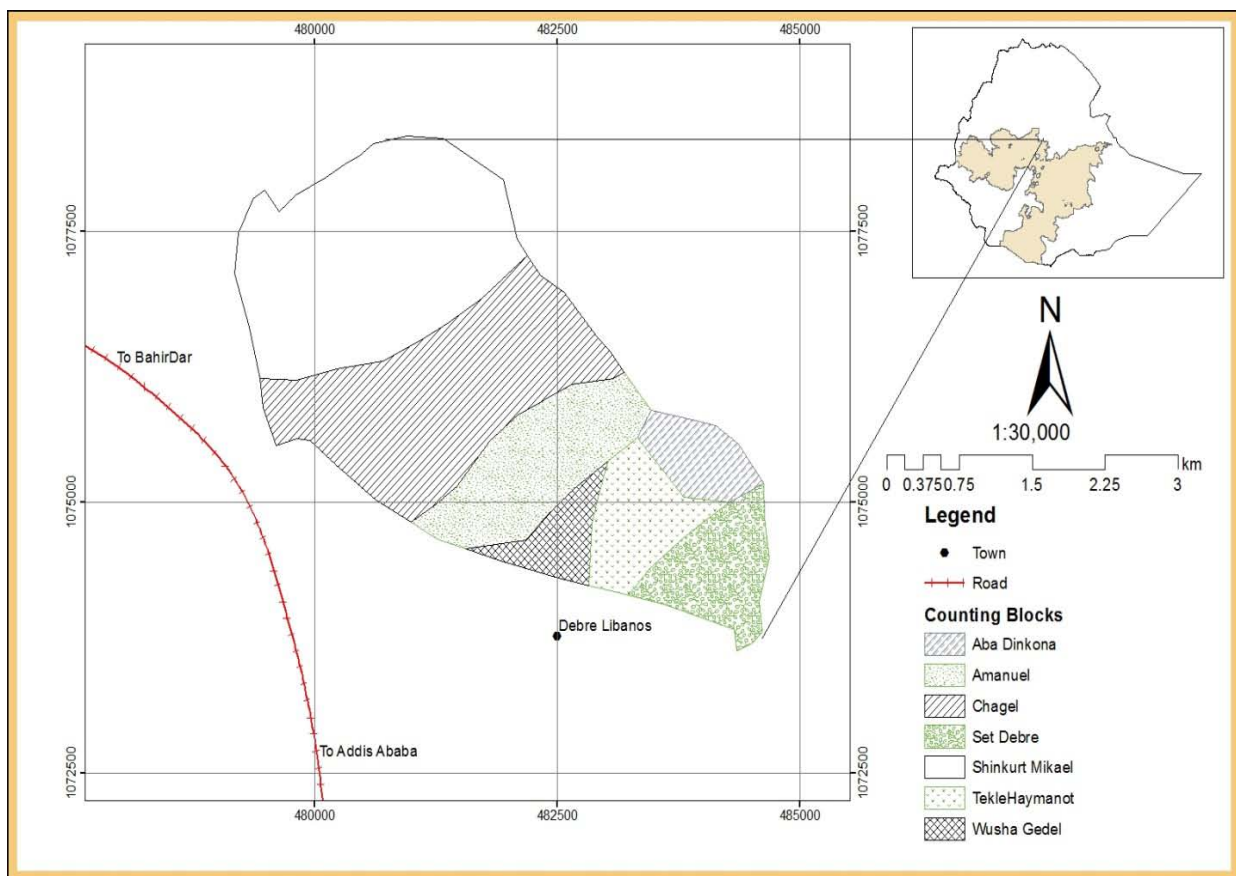


Figure 1: Map of the study area

b) Materials and methods

The present study was conducted from August 2012 to March 2013 to cover both wet and dry seasons. Quantitative and qualitative data were collected during the wet and dry seasons on the population estimate, and group size of the gelada baboon around Debre-Libanos area.

A preliminary survey was conducted in the study area in the first week of August 2012. During this period, the distribution of gelada baboon in the study

area was assessed and the classification of vegetation type was carried out. The survey revealed that the vegetation cover and topography of the area was not homogenous. For the purpose of total counting of gelada baboons, the entire study area was divided into seven blocks; these are Shinkurt Mikael, Chagel, Amanuel, Aba Dinkona, Wusha Gedel, Tekle Haimanot, and Set Debre. Division of the entire area was based on artificial boundaries like roads and bridges, and natural boundaries such as small hills and vegetation

composition of the area. 'Blocks' in this context refer to small areas with natural and artificial boundaries that can easily be identified on the map as well as on the ground. The distance and expanse of the consecutive counting blocks vary depending on the natural boundaries and the topography of the area.

Total counting of gelada baboon population based on direct observation and silent detection was carried out to estimate the number of gelada baboons, and determine their group size and structure. Direct observational technique is most appropriate and effective for medium to large sized animals that live in relatively open habitats (Norton-Griffiths, 1978; Caughly and Sinclair, 1994; Sutherland, 1996). Due to the mountainous nature of the area, almost all observations have been made on foot. A total of three counts were carried out in each of the wet and dry seasons in all the blocks. Each count was carried out simultaneously with the help of local people of the area.

Information was provided for data collectors on how to identify and categorize gelada baboons into adult males, adult females, sub-adult males, sub-adult females, young and infants. Adult males were defined as males with visible manes and overall size about twice that of the adult females. Sub-adult males were males similar in size to adult females with initial development of manes. Adult and sub-adult females were identified based on body size. All other individuals considered as young and infant are based on their body size (Beehner *et al.*, 2008). Because of the smaller size of young and infants, identification of sex was difficult from a distance. During total counting of the gelada baboons, their sex

and age composition were also recorded. Age and sex determination were carried out based on body size, presence or absence of mane, and by looking at their genital organs, following Mori *et al* (19990), and Kingdon (1997).

During each total count, the total number of individuals encountered in a group was recorded on a prepared data sheet before further subdividing into respective age and sex categories. When the distance between them was less than 50 meters, the animals were considered as the same group, following Lewis and Wilson (1979), Befekadu Refera and Afework Bekele (2004).

c) *Data analysis*

The data were pooled together, and SPSS software for Windows Evaluation Version 20 was used for statistical analysis using descriptive statistics and chi-square test. Statistical tests used were two-tailed with 95% confidence intervals. Chi-square test was used to compare between the sex ratio of gelada baboon, and their distribution among the counting blocks between wet and dry seasons.

d) *Results*

The total count of gelada baboon individuals in the study area for both wet and dry seasons is given in Tables 1 and 2. The average number of gelada baboons observed in the entire study area was 1608 individuals. The total number of gelada baboon counted was 1642 during the wet season, and 1573 during the dry season. There was no significant difference between the dry and wet seasons count ($\chi^2= 1.481, df=1, p>0.05$).

Table 1: Number of gelada baboon counted in each counting blocks during the wet season (AM, adult male; AF, adult female; SAM, sub-adult male; SAF, sub-adult female)

Blocks	AM	AF	SAM	SAF	Young	Infants	Total	Percent
Abba Dinkona	5	27	7	14	15	13	81	4.9
Amanuel	10	41	15	18	20	23	127	7.7
Chagel	21	90	31	44	63	39	288	17.5
Set Debre	52	153	23	62	80	56	426	25.9
Shinkurt Mikael	62	207	23	56	85	51	484	29.5
Tekle Haimanot	7	30	6	16	18	11	88	5.4
Wusha Gedel	12	39	15	25	30	27	148	9.0
Total	169	587	120	235	311	220	1642	

In the total population of gelada baboons counted in the study area, adult females comprised the largest proportion. Next to adult females, the largest proportion was young individuals.

During the wet season, the total population was composed of 10.29% adult males, 35.75% adult females, 7.31% sub-adult males, 14.31% sub-adult females, 18.94% young and 13.39% infants. There was a

significant difference among the age groups of the population counted during the wet season of the study period ($\chi^2=506.146, df=5, p<0.05$).

During the wet season, the number of adult females was higher than adult males, and showed significant difference ($\chi^2= 231.116, df=1, p<0.05$). There was also significance difference between sub-adult males and sub-adult females ($\chi^2=37.254, df=1,$

$p < 0.05$). Comparison of young individuals with infants showed a significant difference ($\chi^2 = 15.595$, $df = 1$,

$p < 0.05$). Out of the total individuals counted during the wet season, 29.5% was from Shinkut Mikael.

Table 2: Total count of gelada baboon during the dry season across blocks (AM, adult male; AF, adult female; SAM, sub-adult male; SAF, sub-adult female)

Blocks	AM	AF	SAM	SAF	Young	Infants	Total	Percent
Abba Dinkona	6	29	8	16	18	13	90	5.7
Amanuel	8	36	12	18	16	14	104	6.6
Chagel	15	72	29	37	48	32	233	14.8
Set Debre	54	161	29	68	90	62	464	29.5
Shinkurt Mikael	57	201	23	54	85	49	469	29.8
Tekle Haimanot	5	29	6	10	15	7	72	4.6
Wusha Gedel	9	38	14	24	30	26	141	9.0
Total	154	566	121	227	302	203	1573	

During the dry season, a total of 1573 gelada baboons were counted. Out of these, there were 9.8% adult males, 35.98% adult females, 7.69% sub-adult males, 14.43% sub-adult females, 19.20% young and 12.91% infants. Adult females were higher in number when compared to other groups. The number of adult females was significantly higher than adult males during the dry season ($\chi^2 = 235.756$, $df = 1$, $p < 0.05$).

During the dry season, there was a significant difference between sub-adult male and sub-adult female ($\chi^2 = 32.287$, $df = 1$, $p < 0.05$); and young and infants ($\chi^2 = 19.408$, $df = 1$, $p < 0.05$). Based on counting blocks of the study area, the largest count during the dry season, 29.8% was from Shinkut Mikael.

Among the counting blocks, the highest population was recorded from Shinkurt Mikael counting block both during wet and dry seasons, 484 and 469 individuals, respectively. The smallest number of gelada baboon was recorded from Abba Dinkona (81 individuals) during wet season, and Tekle Haimanot (72 individuals) counting block during the dry season. Out the total gelada baboon individuals counted during both the wet and dry seasons, 29.55% was from Shinkurt Mikeal, 16.10% from Chagel, 7.15% from Amanuel, 5.3% from Abba Dinkona, 8.95% from Wusha Gedel, 5% from Tekle Haimanot and 27.65% from Set Debre (Figure 2).

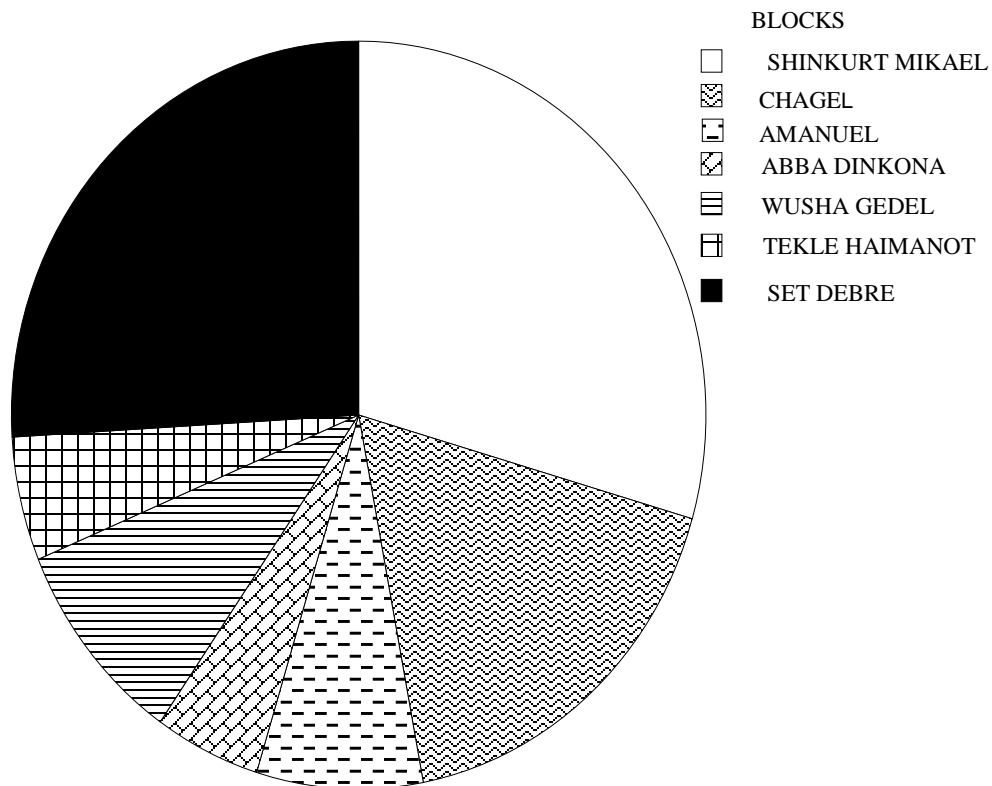


Figure 2: Percentage count of individuals during wet and dry seasons

The age structure and sex ratio showed 10% adult males, 35.8% adult females, 7.5% sub-adult males, 14.5% sub-adult females, 19.0% young and 13.1% infants. There was a significant difference among the different age groups of the total individuals counted in the study area ($\chi^2=1002.657$, $df=5$, $p<0.05$). The age and sex ratio given in Table 3 shows the ratio of adult male to adult female 1:3.47 (during the wet season) and 1:3.68 (during the dry season). The ratio of sub-adult male to adult female is 1:4.89 (during the wet

season) and 1:4.68 (during the dry season). The ratio of adult males to sub-adult females was (1:1.47 during the dry season and 1:1.39 during the wet season). The ratio of young to adult male was (1:1.96 during the dry season and 1:1.84 during the wet season) and to adult female was (1:1.89 during the wet season and 1:1.87 during the dry season). The ratio of infants to adult females was (1:2.79 during the dry season and 1:2.67 during the wet season) and to adult male was (1: 1.32 during the dry season and 1:1.3 during the wet season).

Table 3: Age and sex categories during wet and dry seasons

Sex	Wet season	Dry season	Mean	Percent
Adult male	169	154	161.5	10.0
Adult female	587	566	576.5	35.8
Sub-adult male	120	121	120.5	7.5
Sub-adult female	235	227	231	14.4
Young	311	302	306.5	19.0
Infant	220	203	211.5	13.1
Total	1642	1573	1607.5	

The group size of gelada baboons ranged from 5 to 187 individuals during the wet season. Group size differed within the wet and dry seasons. The highest range of group size was recorded during the wet season (5-187) with the mean group size of 28.21. During the dry season, the range of group size ranged from 3 to 120 with the mean group size of 18.4. Large group sizes of gelada baboons (up to 187) were aggregated during the wet season, while during the dry season, they split into smaller number of groups over wider areas. The density of gelada baboons in the entire study area (84.3km²) during the wet season was 19.49 individuals/km² and 18.67 individuals/km² during the dry season. Their density was higher during the wet season than the dry season.

III. DISCUSSION

In the present study, the total number of individual gelada baboons counted in the study area was 1642 and 1573 during the wet and dry seasons, respectively. The insignificant variation in the number of gelada baboons may be due to the migration of gelada baboons to adjacent areas to get better feeding site and reduce the effect of food scarcity. This result is similar with the finding of Hailu Beyene, 2010. According to Zewdu Kifle, Gurja Belay and Afework Bekele (2013), the population estimate of gelada baboons in Wenchit valley area was around 1525 individuals that was more or less closer to the total population counted around Debre-Libanos. The number of gelada baboons varied among the counting blocks during the wet and dry seasons. This variation of gelada baboons among counting blocks may due to human disturbance, food availability, and variation in the size of counting blocks. Yonatan Ayalew (2009) also obtained similar variation of gelada baboon among counting blocks.

The present study showed that the population composition of gelada baboons consisted of unequal sex and age ratio. There was significant difference in age group and sex ratio between wet and dry seasons of the study period. It was also happened in the work of Yonatan, 2009 and Mussa Adem, 2009. The number of adult females was significantly greater than adult males. Adult male to adult female ratio was similar with report of Beehner et.al., (2008) and Habtamu Asfafaw and C. Subramanian (2013) where an adult sex ratio of 1:3.40 was recorded. The possible reasons for an unequal sex ratio may be due to an increased predation pressure on males, and the emigration of subordinate males to less favourable habitats (Estes, 1974).

There was a large group size during the wet season of the study period, and this decreased during the dry season. The same group size variation in the wet and dry seasons was observed in the study of Mussa Adem (2009). The aggregation of large group of gelada baboon population during the wet season in a limited area may be due to lack of space and presence of ample food in their living habitat. During the wet season, the local people farmed the area and chased gelada baboons to protect their crops, restricting gelada baboon population to the cliffy parts of the study area. Increment of human population causes high demand of land for farming, livestock grazing, settlement and other purposes to fulfill the basic daily requirement of people, and it results to restriction of animals into small areas (Siex and Struhsaker, 1999). But, during the dry season, they were distributed in different habitat types of the study area. This distribution in wider area might be to avoid the unpalatable foods and due to shortage of foraging access resulting the reduction of group size during the dry season. Gelada baboons migrate to different areas adjacent to the study site as food and

water can be limited at the edges of the cliff during the dry season. Iwamoto (1993) revealed that due to the shortage of food resources, baboons move greater distance in search of their food during the dry season. The Distribution of gelada baboon population is based on the availability of food availability and quality, and distance from human (Wallace, 2006).

IV. CONCLUSION AND RECOMMENDATION

Most of the gelada baboon's population exists on the gorges and mountain cliffs of Ethiopia. The present study provided relevant information on gelada population and group size, and age structure in Debre-Libanos area. The average number of gelada baboons counted was 1608. There was a significant difference among the different age groups of the total individuals counted. Unequal sex and age ratio among the individual count was recorded. The highest range of group size was recorded during the wet season with the mean group size of 28.21.

The following points are suggested to reduce the problems and conserve the population of gelada baboon properly:

- Develop sense of ownership among the local community through awareness creation
- Set clear demarcation of conservation area for better conservation of gelada baboons
- Prior to this study, no comprehensive gelada baboon census of the area has been carried out. So, gelada baboon population censuses should be carried out in the future at specific duration to determine the population trends at Debre-Libanos.

Conflict of interests

The authors did not declare any conflict of interest.

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Microorganisms and Aflatoxin Content in Ready-To-Eat Groundnut Paste from Some Markets in Anambra and Edo States, Nigeria

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Abstract- This study was undertaken to quantitatively and qualitatively estimate microbial and aflatoxin content in ready – to – eat groundnut pastes sold in some markets in Anambra and Edo States, Nigeria. A total of 100 samples of ready- to eat – groundnut pastes packaged in plastic cans and low density polyethylene were purchased from some markets in Anambra State (Head Bridge, Ogbaru, Agulu, Mgbuka and Awka) and Edo State (Oba market, Santana, New Benin, Uselu and Oregbeni). The samples were analyzed microbiologically and physicochemically using standard procedures. Aflatoxin detection was done using Enzyme-Linked Immunosorbent Assay (ELISA). Bacteria species associated with the samples were identified as *Staphylococcus aureus*, *Bacillus cereus*, *Bacillus subtilis*, *Micrococcus roseus*, *Escherichia coli* and *Pseudomonas aeruginosa* while fungi include *Aspergillus flavus*, *Aspergillus tamarii*, *Aspergillus niger*, *Aspergillus fumigates* and species of *Penicillium* and *Fusarium*.

Keywords: ready-to-eat groundnut paste, microbial quality, aflatoxin content, ELISA method.

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Microorganisms and Aflatoxin Content in Ready-To-Eat Groundnut Paste from Some Markets in Anambra and Edo States, Nigeria

Okwu, Grace Ifeoma ^α, Akpe, Azuka Romanus ^σ, Amhanre, Idi Napoleon ^ρ & Ogbon-Ogieva, Edosa ^ω

Abstract- This study was undertaken to quantitatively and qualitatively estimate microbial and aflatoxin content in ready – to – eat groundnut pastes sold in some markets in Anambra and Edo States, Nigeria. A total of 100 samples of ready- to eat – groundnut pastes packaged in plastic cans and low density polyethylene were purchased from some markets in Anambra State (Head Bridge, Ogbaru, Agulu, Mgbuka and Awka) and Edo State (Oba market, Santana, New Benin, Uselu and Oregbeni). The samples were analyzed microbiologically and physicochemically using standard procedures. Aflatoxin detection was done using Enzyme-Linked Immunosorbent Assay (ELISA). Bacteria species associated with the samples were identified as *Staphylococcus aureus*, *Bacillus cereus*, *Bacillus subtilis*, *Micrococcus roseus*, *Escherichia coli* and *Pseudomonas aeruginosa* while fungi include *Aspergillus flavus*, *Aspergillus tamarii*, *Aspergillus niger*, *Aspergillus fumigates* and species of *Penicillium* and *Fusarium*. Total bacterial count ranged from $2.50 \pm 0.61 \times 10^4$ – $5.54 \pm 0.50 \times 10^4$ cfu/g. Lowest bacterial count ($2.50 \pm 0.61 \times 10^4$ cfu/g) was obtained in Awka market while the highest ($5.54 \pm 0.50 \times 10^4$ cfu/g) was from Oba market. The fungal counts ranged from $1.70 \pm 0.99 \times 10^3$ – $5.60 \pm 0.65 \times 10^3$ cfu/g with Uselu market having the lowest counts ($1.70 \pm 0.99 \times 10^3$ cfu/g) and Ogbaru market in Anambra State having the highest counts ($5.60 \pm 0.65 \times 10^3$ cfu/g). Aflatoxin content of the samples ranged from 1.1 ± 0.07 – 143.9 ± 2.72 ppb. The presence of pathogenic bacteria and aflatoxin in the ready-to-eat groundnut paste pose a potential health challenge to the consumers in some parts of Anambra and Edo States, Nigeria.

Keywords: ready-to-eat groundnut paste, microbial quality, aflatoxin content, ELISA method.

I. INTRODUCTION

Groundnut (*Arachis hypogaea*) which is also known as peanuts, monkey-nut and goobers, are edible seeds of legume plant that grow to maturity in the ground rather than on aerial part of the plant. As a legume, groundnut belongs to the botanical family Fabaceae (also known as Leguminosae, and commonly known as the bean or pea family). The initial domestication of groundnut may have taken place in North – Western Argentina or in South – Eastern Bolivia where the peanut landraces with the most wide – like features are grown today (Hepper, 2001).

Groundnut is widely grown in the tropics and sun tropics, being important to both small holder and

large commercial producers. It was introduced into Nigeria in the 16th century and it has been estimated that about 1.4 million hectares is cultivated for groundnut in Nigeria. Nigeria is the 4th largest producer of groundnut with a proportion of 4.5% of the total world production. It follows China, India, and USA with 45.5%, 18.2% and 6.8% respectively of total world groundnut production. In West Africa, Nigeria produces 41% of the total groundnut production (Taru *et al.*, 2008).

Groundnut contains high quality edible oil (50%), easily digested protein (25%) and carbohydrate (20%) (Taru *et al.*, 2008; Muhammad – Lawal *et al.*, 2012). Groundnut seeds are nutritionist source of vitamin E, folic acid, calcium, zinc, iron, phosphorus, niacin, magnesium, potassium, riboflavin, and thiamine (Surendranatha *et al.*, 2011). It is widely consumed in Nigeria as roasted or boiled nuts. Groundnut processing is basically the transformation of raw groundnuts and would yield edible oil which can be refined to get vegetable oil.

Groundnut paste is a food paste made primarily from ground dry roasted groundnut and is popular in the North America, United Kingdom, Phillipines and Netherlands. It is mainly used as sandwich spread, sometimes in combination (peanut butter and jelly sandwich – sandwich). In Nigeria, ready – to – eat groundnut paste is consumed in combination with bread, kolanuts and garden egg (*Solanum melongena*). It is rich in protein, fat and oil and is a very good source of vitamins (Achu *et al.*, 2005). Groundnut paste has been frequently associated with food illness in which initial contamination is traceable to food handlers. Numerous epidemiological reports and studies have implicated food of ready – to – eat origin as the major vehicles associated with illness caused by food – borne pathogen (Sokari, 1991).

Due to unhygienic practices during processing, groundnut pastes can be contaminated with microorganisms. Species of *Proteus*, *Serratia*, *Micrococcus*, *Bacillus*, *Staphylococcus*, *Salmonella*, and *Escherichia* were isolated from peanut butter samples obtained in Port Harcourt (Odu NN and Okonko *et al.*, 2012). Also, Elzupir *et al.*, (2011) reported total aflatoxin concentration in peanut butter ranging from 26 .6 – 853 μg /Kg in Sudan. Microbial and aflatoxin contamination is a major concern in food safety as they

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affect humans, animal and economic growth of any nation (Hwang *et al.*, 2004).

II. MATERIALS AND METHODS

Samples of ready – to – eat groundnut pastes were collected randomly between December, 2015 and February, 2016 from five (5) major markets each in Anambra State namely Head Bridge, Ogbaru, Agulu, Mgbuka and Akwa and in Edo State which include Oba market , Santana , New Benin, Uselu and Oregbeni in 2015 and 2016, respectively. Ten samples were bought from each market making the total number of samples collected 100. They were packaged in low density polyethylene.

a) Bacteriological Analysis

Ten (10) grams of ready – to – eat groundnut paste was weighed aseptically into a conical flask and 90ml of sterile distilled water was added. A ten – fold serial dilution was made using 1ml aliquot from stock solution. Pour plate method was used to cultivate the organism. An aliquot of 1ml of the ten – fold serial dilution was poured into the petri – dishes before the molten agar was poured over it. The plates were incubated at 37°C for 24 hours. At the end of incubation, total viable counts were expressed in cfu/g and colonies of isolates were purified by sub-culture into fresh plates of nutrient agar and thereafter stored in nutrient agar slant for characterization and identification using the method of Holt (1994).

b) Mycological Studies

A ten - fold serial dilution of 1ml of each of the sample was made. An aliquot of 1ml was plated on Sabouraud dextrose agar (SDA) amended with chloramphenicol to inhibit bacteria growth and alcohol for fungi isolation. All plates were incubated at room temperature (28°C) for seven (7) days. After incubation, the colonies were counted and expressed in cfu/g. Isolated pure fungal colonies were identified according to Robert *et al.*, (2004).

c) Physicochemical Analysis

The physicochemical parameters analysed were pH and titratable acidity. pH value of the groundnut samples was determined with a single electrode pH meter (Hanna calibrated pH Tester). The sample was prepared by homogenizing 10g of ready – eat groundnut paste in 90ml of sterile distilled water. The pH of the solution was taken.

The titratable acidity of the sample (ready – to – use groundnut paste) was determined by introducing 10ml of the sample solution into a conical flask with three (3) drops of phenolphthalein indicator. Sodium hydroxide (0.1M solution) was placed in a burette and titrated against the supernatant in the flask. The appearance of a pinkish colour indicated the end point

of titration. The procedure was carried out in triplicates and average values noted.

d) Determination of Aflatoxin Content In The Ready – To – Eat Groundnut Paste Samples

Aflatoxin content of samples was determined using Enzyme – Linked Immunosorbent Assay (ELISA) method. The analysis was done using competitive ELISA method and Agraquant Total Aflatoxin Assay Kit (from Roman Singapore Company).

e) Elisa Testing Procedure

i. Extraction of ready – to – eat groundnut paste samples

5gm of sample was weighed into a suitable plastic container that was covered and 25ml of ethanol / tween water (70: 30) was added. It was placed on the shaker set at 250 rotations for 110 seconds and was filtered using the folded filter paper.

ii. Sample Analysis

A multichannel pipette set at 200µl was used to pipette aflatoxin enzyme conjugate into the uncoated antibody microplate wells. 100µl of each standard solution and samples extract were added to the coated antibody microplated wells and incubated at 20 – 25°C for 10minutes.

Toxins in samples and control standards compete with enzyme conjugate for binding to solid phase antibody. Content were discarded and washed with de – ionized water for 5minutes and dried using absorbent paper towel. Thereafter, 100µl of enzyme substrate was added to wells and incubated at room temperature for 5minutes until blue colour was observed in wells. Then 100µl of stop solution was added to the wells and the blue colour change into yellow. Dye concentration is inversely related to concentration of toxin in the sample and standard. ELISA reader in the wavelength of 450 – 630nm was used to analyzed toxin concentration and wells absorbance. Toxin concentration in the sample was compared with standard concentration curve.

III. RESULTS AND DISCUSSION

The microbial and aflatoxin contamination in food commodities and its association with health risk in both animals and humans continue to raise increasing concern over years. Groundnuts pastes are a rich source of protein and edible oil; it has been reported to be mostly contaminated by microorganisms and aflatoxin. In this study, microbial counts as well as aflatoxin content of this product were determined. The bacterial counts ranged from $2.50 \pm 0.61 \times 10^4$ – $5.54 \pm 0.50 \times 10^4$ cfu/g and fungi counts ranged from $1.7 \pm 0.99 \times 10^3$ – $5.6 \pm 0.65 \times 10^3$ cfu/g as shown in Table 1. Fungal load of the ready – to – eat groundnut pastes had lower counts than bacterial counts. Work done by Adebisin *et al.* (2001) indicated similar microbial load

from groundnut products hawked in Bauchi. The bacterial count ranged from $1.16 \pm 0.35 \times 10^5$ to $5.92 \pm 0.59 \times 10^5$ (cfu/g), while the fungal counts ranged from $1.91 \pm 0.32 \times 10^4$ - $8.60 \pm 0.22 \times 10^4$ cfu/g in the samples. However, Akinnibosun and Osawaru, 2015 reported higher fungal than bacterial load in Benin Metropolis, Edo State, Nigeria.

Samples from Anambra had higher microbial contamination than samples from Edo State. This could be attributed to poor processing and prolonged storage as the pastes were produced in bulk in Anambra State and might not be readily sold out because of bulk product overshadowing the demand, forcing the bulk producers to embark on long storage which can be termed lag storage resulting in proliferation of organisms contrary to the practice in Edo State, where due to low patronage, the pastes were produced in low quantities and almost on demand.

The bacteria and fungi isolates from the samples are shown in table 2. The presence of these bacterial species in the product was of particular interest because of their possible involvement in food infections. The presence of *Staphylococcus* sp and *Micrococcus* sp in samples could be attributed to contamination from human handling, the surrounding air and environment during processing and display for sale in the markets. Most groundnut pastes samples are stored at temperature between 26°C – 30°C which favours the growth of the organisms. *Staphylococcus* sp in the groundnut pastes may result if the grinding machine and other utensils are contaminated. The sanitary conditions of the environment may also lead to contamination of the products. The presence of these microbes is an indication of the use of non – portable water, which is mostly used in local food processing (Adebayo-Tayo *et al.*, 2009; Adebayo-Tayo *et al.*, 2012). *S. aureus* is known for production of heat stable enterotoxin and has potentials for multiple antibiotic resistances when they get into living tissue (Scot, 2002) making the product an immense epidemiological danger (Adebayo-Tayo *et al.*, 2009; Adebayo-Tayo *et al.*, 2012). Heat stressed microorganisms that survived roasting process are capable of growing if samples were not preserved under appropriate temperature (Sokari, 1991).

Bacillus sp and *Pseudomonas* sp are normal inhabitants of the soil and capable of causing disease in humans and animals. These organisms are capable of surviving harsh conditions such as roasting and blending processes (Doyle, 2007). The presence of *E.coli* indicated unhygienic handling of the products right from the source, storage and packaging. This might have adverse effect on the health of the consumers (Okonko *et al.*, 2008a,b,c). The isolation of *E.coli* in the pastes was in line with the findings of Odu and Okonko, 2010 who also isolated *E.coli*, *Bacillus* sp,

Serratia sp, *Proteus* sp, *Micrococcus* sp, and *Staphylococcus* sp.

All the samples showed fungal growth with Ogbaru market being the most contaminated sample ($5.6 \pm 0.65 \times 10^3$ cfu/g) although this is below the maximum tolerance limit of 10^4 cfu/g recommended by International Commission on Microbiological Specification for foods (Elliot, 1980; Da Silver, 2000). Fungal species isolated includes; *Aspergillus flavus*, *A. niger*, *A. fumigatus*, *A. tamarii*, *Fusarium* sp and *Penicillium* sp. *Fusarium* species are plant pathogens that contaminate crops in the field or immediately after harvest while *Aspergillus* species are predominantly storage contaminants (Sweeney and Dobson, 1999). The presence of these organisms in the groundnut pastes might be due to improper handling during processing, hawking and display in open trays for sales. Some of these fungi especially *Aspergillus* sp is able to survive in situations where free water is not available (Reddy *et al.*, 2012). Their presence in the product may result in production of toxic substances such as aflatoxins and fumonisins.

Table 3 revealed the pH and titratable acidity values of the samples. The pH values of the groundnut paste samples ranged from 5.9 ± 0.01 (Oba market in Edo State) to 6.9 ± 0.03 (Agulu market in Anambra State). The pH values obtained could be as a result of the presence of bacteria in the groundnut samples whose pH optimal for growth is near neutrality (7.0), showing no erosive potential to the body as acidic pH in the body can occur from an acidic forming diet which negatively affects blood body's ability to absorb minerals and other nutrients, decrease the energy production in cells, and decrease its ability to repair damaged cells.

Titratable acidity measurement has also been used to assess food erosive potential. The titratable acidity ranged from 0.02 to 0.05 with Oba markets having the highest titratable acidity. This may be due to temperature in storage material which may have increased the activities of lactic acid bacteria breaking down sugars to produce lactic acid among the groundnut paste samples.

Total aflatoxin content (Table 4) obtained from samples from different open markets ranged from 1.1 ppb – 143.9 ppb. The samples obtained from Anambra State indicated high levels of aflatoxin B1 compared to samples from Edo State. This could be attributed to groundnut species, processing, handling and storage. The mean aflatoxin concentration of the samples obtained from Anambra State was five times higher than the specification of National Agency for Food, Drug Administration and Control in Nigeria whose permitted aflatoxin limit is 4 ppb in ready – to – eat foods. Akano and Atanda (1990) found aflatoxin B1 concentrations in the range of 20 - $455\mu\text{g}/\text{kg}$ in groundnut cake purchased from market in Ibadan, Oyo State, Nigeria.

Similarly, Adebajo and Idowo (1994) reported that most of the corn groundnut snacks, contained aflatoxins above 30µg /kg immediately after preparation. Isibor *et al.* (2010) also reported high levels of aflatoxin contamination in groundnut and groundnut products while Okwu *et al.*, (2010) revealed high incidence and alarming level of naturally produced aflatoxin in ready – to –use food thickeners sold in South – East geopolitical zone in Nigeria. The fungi that produce mycotoxins proliferate in the tropics where climatic and crop storage such as temperature, humidity, and water activity are conducive for fungal growth (D Mello and Mavdonald, 1997). The incidence of fungal contamination has also been linked to high rainfall and high relative humidity (Gamanya and Sibanda, 2001).

High levels of aflatoxin and fungal contamination result in a decrease in quality and nutritional value of ready – to – eat groundnut pastes (Gong *et al.*, 2002). Furthermore, children are most vulnerable to the detrimental effects of aflatoxin (Williams *et al.*, 2004; Cullen and Newberne, 2003). Hence the detection of high levels of aflatoxins in ready – to – eat groundnut is a cause for concern. Also the threshold dose for aflatoxin leading to malnutrition and growth stunting in children is very low (Shephard, 2003). In Togo and Benin, chronic aflatoxicosis was also linked to growth stunting and underweight in infants under 5 years old (Gong *et al.*, 2002). It should also be noted that underweight children are also prone to child mortality and acute morbidity due to diarrhoea, malaria, measles, pneumonia and other selected infectious diseases (Williams *et al.*, 2004).

The commercial ready – to – eat groundnut pastes were contaminated. This might be due to several factors including poor policing of set regulations by the authorities and lack of effective quality control systems to cover unpackaged food products (e.g. factory inspection for aflatoxin contamination, hand sorting to remove shriveled nuts, and proper cleaning of

equipments). However, manufacturing process can reduce aflatoxin contamination to levels that are acceptable in many countries. It is imperative that effective system for policing unregistered and unpackaged food products be put in place by constituted authorities. This will prevent manufacturers from churning out ready – to – eat groundnut pastes that have aflatoxin levels above the maximum tolerable limits.

IV. CONCLUSION

Tackling the problem of microbial contamination and aflatoxin in developing countries is very difficult and complex. Policy makers and the general public in the regions with the highest contamination levels generally lack full knowledge of aflatoxins and the scale of adverse health effects they cause (Gong *et al.*, 2004). A vast majority of the people mostly affected produce and consume their own food, rendering regulatory measures to control exposure ineffective (Shephard, 2003). In general, public health programmes like vaccination, malaria prevention and control, improved sanitation and clean drinking water supply are perceived to be more valuable than aflatoxin control (Gong *et al.*, 2003). Furthermore, the aflatoxin problem sits at the interface of agriculture, health and international trade and it is impossible to tackle it without first tackling the insurmountable problems affecting these sectors in the developing countries (Gong *et al.*, 2003; Shephard, 2003.).

However, manufacturing process can reduce microbial and aflatoxin contamination to levels that are acceptable in many countries. It is imperative that effective systems for policing unregistered and unpackaged food products be put in place by constituted authorities. This will prevent manufacturers from churning out ready – to – eat groundnut paste that has microbial and aflatoxin levels above the maximum tolerable limits.

Table 1: Total Microbial Counts In Ready –To – Eat Groundnut Paste Samples Obtained From Ten Open Markets In Anambra And Edo States, Nigeria

Sample	Market	Total Bacterial Counts CFU/g ± SEM x 10 ⁴	Total Fungal Counts CFU/g ± SEM x 10 ³
Anambra	Head Bridge	4.20 ± 0.42	4.0 ± 0.45
	Ogbaru	5.00 ± 0.40	5.6 ± 0.65
	Agulu	4.80 ± 0.48	4.63 ± 0.57
	Awka	2.50 ± 0.61	5.20 ± 0.61
Edo	Mgbuka	3.66 ± 0.32	4.20 ± 0.70
	Oba	5.54 ± 0.50	4.03 ± 0.50
	Satana	2.90 ± 0.18	1.78 ± 0.13
	Oregbeni	3.53 ± 0.24	2.71 ± 0.18
	Urelu	2.99 ± 0.32	1.70 ± 0.99
	New Benin	3.75 ± 0.32	2.29 ± 0.18

SEM = Standard Error Mean

Table 2: Bacteria and Fungi Isolated From Ready – To – Eat Groundnut Pastes From Ten Markets

Market	Bacterial Isolates	Fungal Isolates
Head Bridge	<i>Staphylococcus aureus</i> , <i>Bacillus</i> sp <i>E.coli</i>	<i>A. flavus</i> , <i>Penicillium</i> sp <i>A. tamarii</i>
Ogbaru	<i>Bacillus</i> sp, <i>Staphylococcus aureus</i>	<i>A. flavus</i> , <i>Penicillium</i> sp <i>A. tamarii</i>
Agulu	<i>Pseudomonas</i> sp, <i>Bacillus</i> sp, <i>Staphylococcus aureus</i>	<i>A. flavus</i> , <i>A. niger</i>
Mgbuka	<i>Staphylococcus aureus</i> , <i>Bacillus</i> sp, <i>Pseudomonas</i> sp	<i>A. flavus</i> , <i>Penicillium</i> sp, <i>A. niger</i>
Akwa	<i>Pseudomonas</i> sp, <i>Bacillus</i> sp	<i>A. flavus</i> , <i>A. niger</i>
Oba	<i>Staphylococcus aureus</i> , <i>Micrococcus roseus</i> , <i>Bacillus cereus</i>	<i>A. niger</i> , <i>Fusarium</i> sp, <i>A. flavus</i>
Satana	<i>Bacillus cereus</i> , <i>Bacillus subtilis</i>	<i>A. flavus</i> , <i>A. fumigates</i>
Oregbeni	<i>Staphylococcus aureus</i> , <i>Bacillus subtilis</i> , <i>Bacillus cereus</i> , <i>Micrococcus roseus</i>	<i>A. niger</i> , <i>A. flavus</i>
Uselu	<i>Staphylococcus aureus</i> , <i>Bacillus subtilis</i> <i>Pseudomonas aeruginosa</i>	<i>A. niger</i> , <i>Fusarium</i> sp <i>A. flavus</i>
New Benin	<i>Micrococcus roseus</i> , <i>Bacillus cereus</i> , <i>Staphylococcus aureus</i>	<i>A. niger</i> , <i>Fusarium</i> sp, <i>A. flavus</i>

Table 3: Physicochemical Characteristics (Mean Ph And Titratable Acidity) Of The Ready – To – Eat Samples From Ten Different Open Markets In Anambra And Edo States.

Market Place Sample Collected	pH ± SEM	Titrateable Acidity ±SEM
Head Bridge	6.0 ±0.03	0.03 ±0.00
Ogbaru	6.7 ±0.00	0.04 ±0.005
Agulu	6.9 ±0.03	0.03 ± 0.01
Mgbuka	6.5 ±0.03	0.02 ± 0.02
Awka	6.4 ±0.29	0.02 ± 0.01
Oba	5.9 ±0.01	0.05 ± 0.00
Santana	6.2 ±0.01	0.03 ± 0.05
Oregbeni	6.5 ± 0.03	0.02 ± 0.00
Uselu	6.7 ± 0.11	0.02 ±0.01
New Benin	6.3 ±0.02	0.04 ±0.01

SEM = Standard Error Mean

Table 4: Aflatoxin Content (Ppb ± Sem) In Ready – To – Eat Groundnut Paste Samples Sold In Markets In Anambra and Edo States.

Sample Location	Market	Total Aflatoxin content (ppb) ± SEM
Anambra	Head Bridge	143.9 ± 2.72
	Ogbaru	108.8 ± 1.59
	Agulu	55.0 ± 1.80
	Awka	43.3 ± 0.73
Edo	Mgbuka	76.8 ± 1.38
	Oba	1.1 ± 0.07
	Santana	27.8 ± 0.21
	Oregbeni	3.0 ± 0.07
	Uselu	43.3 ± 1.80
	New Benin	3.5 ± 0.23

SEM = Standard Error Mean

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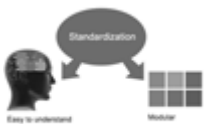
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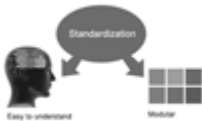
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Final Points:

A purpose of organizing a research paper is to let people to interpret your effort selectively. The journal requires the following sections, submitted in the order listed, each section to start on a new page.

The introduction will be compiled from reference matter and will reflect the design processes or outline of basis that direct you to make study. As you will carry out the process of study, the method and process section will be constructed as like that. The result segment will show related statistics in nearly sequential order and will direct the reviewers next to the similar intellectual paths throughout the data that you took to carry out your study. The discussion section will provide understanding of the data and projections as to the implication of the results. The use of good quality references all through the paper will give the effort trustworthiness by representing an alertness of prior workings.



Writing a research paper is not an easy job no matter how trouble-free the actual research or concept. Practice, excellent preparation, and controlled record keeping are the only means to make straightforward the progression.

General style:

Specific editorial column necessities for compliance of a manuscript will always take over from directions in these general guidelines.

To make a paper clear

- Adhere to recommended page limits

Mistakes to evade

- Insertion a title at the foot of a page with the subsequent text on the next page
- Separating a table/chart or figure - impound each figure/table to a single page
- Submitting a manuscript with pages out of sequence

In every sections of your document

- Use standard writing style including articles ("a", "the," etc.)
- Keep on paying attention on the research topic of the paper
- Use paragraphs to split each significant point (excluding for the abstract)
- Align the primary line of each section
- Present your points in sound order
- Use present tense to report well accepted
- Use past tense to describe specific results
- Shun familiar wording, don't address the reviewer directly, and don't use slang, slang language, or superlatives
- Shun use of extra pictures - include only those figures essential to presenting results

Title Page:

Choose a revealing title. It should be short. It should not have non-standard acronyms or abbreviations. It should not exceed two printed lines. It should include the name(s) and address (es) of all authors.



Abstract:

The summary should be two hundred words or less. It should briefly and clearly explain the key findings reported in the manuscript-- must have precise statistics. It should not have abnormal acronyms or abbreviations. It should be logical in itself. Shun citing references at this point.

An abstract is a brief distinct paragraph summary of finished work or work in development. In a minute or less a reviewer can be taught the foundation behind the study, common approach to the problem, relevant results, and significant conclusions or new questions.

Write your summary when your paper is completed because how can you write the summary of anything which is not yet written? Wealth of terminology is very essential in abstract. Yet, use comprehensive sentences and do not let go readability for briefness. You can maintain it succinct by phrasing sentences so that they provide more than lone rationale. The author can at this moment go straight to shortening the outcome. Sum up the study, with the subsequent elements in any summary. Try to maintain the initial two items to no more than one ruling each.

- Reason of the study - theory, overall issue, purpose
- Fundamental goal
- To the point depiction of the research
- Consequences, including definite statistics - if the consequences are quantitative in nature, account quantitative data; results of any numerical analysis should be reported
- Significant conclusions or questions that track from the research(es)

Approach:

- Single section, and succinct
- As an outline of job done, it is always written in past tense
- A conceptual should situate on its own, and not submit to any other part of the paper such as a form or table
- Center on shortening results - bound background information to a verdict or two, if completely necessary
- What you account in an abstract must be regular with what you reported in the manuscript
- Exact spelling, clearness of sentences and phrases, and appropriate reporting of quantities (proper units, important statistics) are just as significant in an abstract as they are anywhere else

Introduction:

The **Introduction** should "introduce" the manuscript. The reviewer should be presented with sufficient background information to be capable to comprehend and calculate the purpose of your study without having to submit to other works. The basis for the study should be offered. Give most important references but shun difficult to make a comprehensive appraisal of the topic. In the introduction, describe the problem visibly. If the problem is not acknowledged in a logical, reasonable way, the reviewer will have no attention in your result. Speak in common terms about techniques used to explain the problem, if needed, but do not present any particulars about the protocols here. Following approach can create a valuable beginning:

- Explain the value (significance) of the study
- Shield the model - why did you employ this particular system or method? What is its compensation? You strength remark on its appropriateness from a abstract point of vision as well as point out sensible reasons for using it.
- Present a justification. Status your particular theory (es) or aim(s), and describe the logic that led you to choose them.
- Very for a short time explain the tentative propose and how it skilled the declared objectives.

Approach:

- Use past tense except for when referring to recognized facts. After all, the manuscript will be submitted after the entire job is done.
- Sort out your thoughts; manufacture one key point with every section. If you make the four points listed above, you will need a least of four paragraphs.



- Present surroundings information only as desirable in order hold up a situation. The reviewer does not desire to read the whole thing you know about a topic.
- Shape the theory/purpose specifically - do not take a broad view.
- As always, give awareness to spelling, simplicity and correctness of sentences and phrases.

Procedures (Methods and Materials):

This part is supposed to be the easiest to carve if you have good skills. A sound written Procedures segment allows a capable scientist to replacement your results. Present precise information about your supplies. The suppliers and clarity of reagents can be helpful bits of information. Present methods in sequential order but linked methodologies can be grouped as a segment. Be concise when relating the protocols. Attempt for the least amount of information that would permit another capable scientist to spare your outcome but be cautious that vital information is integrated. The use of subheadings is suggested and ought to be synchronized with the results section. When a technique is used that has been well described in another object, mention the specific item describing a way but draw the basic principle while stating the situation. The purpose is to text all particular resources and broad procedures, so that another person may use some or all of the methods in one more study or referee the scientific value of your work. It is not to be a step by step report of the whole thing you did, nor is a methods section a set of orders.

Materials:

- Explain materials individually only if the study is so complex that it saves liberty this way.
- Embrace particular materials, and any tools or provisions that are not frequently found in laboratories.
- Do not take in frequently found.
- If use of a definite type of tools.
- Materials may be reported in a part section or else they may be recognized along with your measures.

Methods:

- Report the method (not particulars of each process that engaged the same methodology)
- Describe the method entirely
- To be succinct, present methods under headings dedicated to specific dealings or groups of measures
- Simplify - details how procedures were completed not how they were exclusively performed on a particular day.
- If well known procedures were used, account the procedure by name, possibly with reference, and that's all.

Approach:

- It is embarrassed or not possible to use vigorous voice when documenting methods with no using first person, which would focus the reviewer's interest on the researcher rather than the job. As a result when script up the methods most authors use third person passive voice.
- Use standard style in this and in every other part of the paper - avoid familiar lists, and use full sentences.

What to keep away from

- Resources and methods are not a set of information.
- Skip all descriptive information and surroundings - save it for the argument.
- Leave out information that is immaterial to a third party.

Results:

The principle of a results segment is to present and demonstrate your conclusion. Create this part a entirely objective details of the outcome, and save all understanding for the discussion.

The page length of this segment is set by the sum and types of data to be reported. Carry on to be to the point, by means of statistics and tables, if suitable, to present consequences most efficiently. You must obviously differentiate material that would usually be incorporated in a study editorial from any unprocessed data or additional appendix matter that would not be available. In fact, such matter should not be submitted at all except requested by the instructor.



Content

- Sum up your conclusion in text and demonstrate them, if suitable, with figures and tables.
- In manuscript, explain each of your consequences, point the reader to remarks that are most appropriate.
- Present a background, such as by describing the question that was addressed by creation an exacting study.
- Explain results of control experiments and comprise remarks that are not accessible in a prescribed figure or table, if appropriate.
- Examine your data, then prepare the analyzed (transformed) data in the form of a figure (graph), table, or in manuscript form.

What to stay away from

- Do not discuss or infer your outcome, report surroundings information, or try to explain anything.
- Not at all, take in raw data or intermediate calculations in a research manuscript.
- Do not present the similar data more than once.
- Manuscript should complement any figures or tables, not duplicate the identical information.
- Never confuse figures with tables - there is a difference.

Approach

- As forever, use past tense when you submit to your results, and put the whole thing in a reasonable order.
- Put figures and tables, appropriately numbered, in order at the end of the report
- If you desire, you may place your figures and tables properly within the text of your results part.

Figures and tables

- If you put figures and tables at the end of the details, make certain that they are visibly distinguished from any attach appendix materials, such as raw facts
- Despite of position, each figure must be numbered one after the other and complete with subtitle
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Discussion:

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- Make a decision if each premise is supported, discarded, or if you cannot make a conclusion with assurance. Do not just dismiss a study or part of a study as "uncertain."
- Research papers are not acknowledged if the work is imperfect. Draw what conclusions you can based upon the results that you have, and take care of the study as a finished work
- You may propose future guidelines, such as how the experiment might be personalized to accomplish a new idea.
- Give details all of your remarks as much as possible, focus on mechanisms.
- Make a decision if the tentative design sufficiently addressed the theory, and whether or not it was correctly restricted.
- Try to present substitute explanations if sensible alternatives be present.
- One research will not counter an overall question, so maintain the large picture in mind, where do you go next? The best studies unlock new avenues of study. What questions remain?
- Recommendations for detailed papers will offer supplementary suggestions.

Approach:

- When you refer to information, differentiate data generated by your own studies from available information
- Submit to work done by specific persons (including you) in past tense.
- Submit to generally acknowledged facts and main beliefs in present tense.



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<i>Introduction</i>	Containing all background details with clear goal and appropriate details, flow specification, no grammar and spelling mistake, well organized sentence and paragraph, reference cited	Unclear and confusing data, appropriate format, grammar and spelling errors with unorganized matter	Out of place depth and content, hazy format
<i>Methods and Procedures</i>	Clear and to the point with well arranged paragraph, precision and accuracy of facts and figures, well organized subheads	Difficult to comprehend with embarrassed text, too much explanation but completed	Incorrect and unorganized structure with hazy meaning
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<i>References</i>	Complete and correct format, well organized	Beside the point, Incomplete	Wrong format and structuring



INDEX

A

Amphipolycotyle · 8, 10
Aspergillus · 34, 36

C

Carolinus · 8, 9, 10, 13, 14, 15
Catarinensis · 8, 10, 13, 15
Contraecum · 15

D

Demarcation · 32

M

Mesenteries · 9
Militating · 1

O

Oligoplites · 8, 9, 17

T

Trachinotus · 8



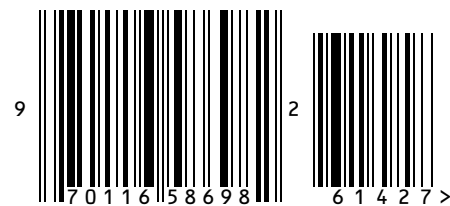
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