Online ISSN : 2249-4626
Print ISSN : 0975-5896
DOI : 10.17406/GJSFR

Global Journal

of Science Frontier Research: B

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Assessment of Surface Water Quality

Discovering Thoughts, Inventing Future

VOLUME 19 ISSUE 1 VERSION 1.0

Global Journal of Science Frontier Research: B
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Volume 19 Issue 1 (Ver. 1.0)

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USA Tax ID: 98-0673427

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Contents of the Issue

Global Journal of Science Frontier Research: B
Chemistry
Volume 19 Issue 1 Version 1.0 Year 2019
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals
Online ISSN: 2249-4626 & Print ISSN: 0975-5896

Physicochemical Assessment of Surface Water Quality around the Sagbama Creek Water Body, Bayelsa State, Nigeria

By Iyama, William Azuka, Edori, Onisogen Simeon & Ede, Precious N

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Keywords: *gross organic pollutants, migration, sagbama creek.*

GJSFR-B Classification: *FOR Code: 039999*

PHYSICOCHEMICALASSESSMENTOFSURFACEWATERQUALITYAROUNDTHESAGBAMACREEKWATERBODYBAYELSASTATENIGERIA

Strictly as per the compliance and regulations of:

Physicochemical Assessment of Surface Water Quality around the Sagbama Creek Water Body, Bayelsa State, Nigeria

Iyama, William Azuka [α], Edori, Onisogen Simeon [σ] & Ede, Precious N [ρ]

Abstract- This study was undertaken to assess the physicochemical quality of the surface water of the Sagbama Creek, Bayelsa State, Nigeria. Samples of water were collected and preserved using trioxonitrate v acid and placed in a cooler of ice for transfer to the laboratory except for insitu measurements for temperature, turbidity, conductivity, TDS, pH, DO and Salinity using Hanna H19828 multi-parameter water quality checker. Five stations were employed correspondingly with control stations. Results were compared with both control and standard permissible limits for the following parameters for both River water samples and the control respectively; temperature (28.56±1.99 and 27.52±0.93), turbidity (225±274 and 359±318), TDS (39±29.60 and 20.80±14.81), conductivity (94±47 and 40.80±29), pH (5.55±0.6 and 5.61±0.24), Salinity (0.04±0.02 and 0.016±0.015), alkalinity (27±31 and 8.80±12.5), Hardness (20.82±22 and 6.66±7.52). Similarly the gross organic pollutants of DO, COD, and BOD recorded 2.22±0.065 and 2.26±0.15; 1.07±0.41 and 0.58±0.41; 0.72±0.49 and 0.39±0.27) respectively. All values were either below or within the permissible limits for either surface water, DPR or FMENV. This means that the Sagbama creek water is not polluted except for the high level of turbidity recorded which were above permissible limits. There is, therefore, the urgent need to de-silt the River by dredging to reduce the turbidity which could lead to migration of aquatic animals.

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I. Introduction

Water is a veritable tool of natural origin which serves for useful purposes to man. The earth's surface is made up of 70% water, which include rivers, lakes, streams, seas, oceans and ground water. All these forms are very important in life cycle (Arimieari, *et al.*, 2014).

The Pollutionor adulteration of superficial waters can also be connected with the nature of water in neighbouring water bodies. The evaluation of water quality is not meant for fitness only inhumanintake or drinking, but also for other important anthropogenic activities and recreation (Arimieari *et al.*, 2014). The need to monitor the quality of water quality becomes very necessary, both as a check on its present state and also as an instrument for management and policy implementation. Total or complete evaluation or investigation of water quality involves examination of all the components of water analysis, such as; physical, chemical and biological properties of water in comparison to set standards, which may be natural or human for proposed purpose (UNESCO/WHO/UNEP, 1996).

Despite the fact that water contamination or pollution is a universal problem, yet the nature of contamination of pollution differs, depending on the developmental stage of the area under investigation. Areas or countries with a fast growing population which do not have proper waste managerial practice or system are more likely to produce wastes which constitute pollution to the environment than those countries with slower population growth rate, that also practice proper waste management control (WHO, 2003).

The characteristic changes that is associated with the river system in the Niger Delta region of Nigeria, presently is worrisome. These changes is associated with different forms of pollution or contamination of the surface water, which serves for drinking and other purposes for the people. The discovery of oil in the region has led to increased population and pollution of the area as a result of both legal and illegal industrial activities (Adesuyi *et al.*, 2015).Therefore, this study was undertaken to investigate the physicochemical properties of surface water around the Sagbama Creek.

II. Materials and Methods

Water samples were collected from the sides of the stretch covering Bolou-rua to the Ebeni/Amasoma Bridge in Sagbama Local Government Area of Bayelsa State. Based on the topography and uniformity in the landscape, five sampling stations were made based on the traversed communities through which the road network passes. This could also mean some anthropogenic inputs from inhabitants may affect some of the water quality parameters. The simple random sampling technique was applied to create the sampling points and control. The Bolou-rua and Toru-orua samples were controlled using ground water samples whereas the Kalabiama, Amatolu, and Ebeni were controlled by the use of the adjoining river known as Sagbama Creek. Samples of water were collected from

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each station (five stations) excluding the control hence three from each and labeled accordingly.

The following parameters were measured *in situ* using the Hanna H19828 multi-parameter water quality checker: Temperature, Turbidity, Dissolved Oxygen, Total dissolved solids, conductivity, salinity, and pH were determined. All samples collected were kept in the ice chest as earlier stated to maintain them at a temperature below 4°C during transfer from the field to the laboratory within 24 hours. Similarly, in the laboratory, the samples were kept in the refrigerator under laboratory conditions until analysis was completed on them. The time between sampling and analysis of samples was kept short and between recommended times by the standard methods. To avoid contamination, the HNO_3 acid used in preservation was ultra-pure grade (J.T. Baker, Altrex).

III. Results and Discussions

The result obtained from this study is presented in Tables 3-5 under the following headings; Physical Parameters such as Temperature, Total Dissolved Solids, Turbidity and Colour, Odour, Conductivity and Chemical and Gross Organic Pollutants; DO, COD, BOD, pH, Salinity, Alkalinity, Hardness; Nutrient parameters, Sulphate, Nitrate, Phosphate, Ammonium. The levels of these parameters are measures of water quality assessment and classification which were compared with known standards to ascertain the water quality status. The geographical locations and analytical techniques/ methods are shown in Tables 1 and 2 respectively.

Table 1: Sampling Stations and Descriptions

Station	Description	Location
A_B	Bolou-rua Station	N05°05'49.5''
B_C	Bolou-rua Control	E 006° 06' 57.2''
A_T	Toru-Orua Station	N05°05'49.4''
T_C	Toru-Orua Control	E 006° 06' 57.2''
A_K	Kalabiama Station	N05°02'40.1''
K_C	Kalabiama Control	E 006° 05' 08.8''
A_A	Amatolu Station	N006°03'50.1''
A_C	Amatolu Control	E 05° 01' 43.8''
A_E	Ebeni Station	N04°59'34.6''
E_C	Ebeni Control	E 006° 04' 28.6''

Table 2: Methods and Techniques of Analysis

Parameter	Techniques
Ammonium NH_4^+	Titrimetric (APHA
Alkalinity	4500,1995)
COD	Titrimetric (APHA 2320
Hardness	B;1995)
Phosphate (PO_4^{3-})	Open reflux (APHA 5220
THC	B;1995)
BOD	EDTA Titrimetric method
	(APHA 2340 C;1995)
	Colorimetric, Ascorbic
	acid method (APHA
	4500-PE)
	APHA 507

Table 3: Physical Parameters for the Sampling Station

Parameter	Experimental Stations						Control Stations					
	A_B	A_T	A_K	A_A	A_E	Mean	B_C	T_C	K_C	A_C	E_C	Mean
Temperature(°C)	25.71	27.30	29.83	30.45	29.50	28.56±1.99	27.77	26.29	28.82	27.12	27.60	27.52±0.93
Turbidity (NTU)	92.90	670	310	25.80	28	225±274	29.60	0.20	543	558	665	359±318
TDS (ppm)	33	10	89	36	28	39±29.60	40	0.00	28	16	20	20.80±14.81
Conductivity (μS/cm	66	75	178	73	77	94±47	79	1	56	32	36	40.80±29

Table 4: Chemical parameters for sampling stations

Parameter	Experimental Stations						Control Stations					
	A_B	A_T	A_K	A_A	A_E	Mean	B_C	T_C	K_C	A_C	E_C	Mean
pH (units)	5.46	5.02	6.59	5.29	5.4	5.55±0.6	5.68	5.19	5.71	5.77	5.70	5.61±0.24
Salinity (PSU)	0.03	0.03	0.08	0.03	0.04	0.04±0.02	0.04	0.00	0.02	0.01	0.01	0.016±0.015
Alkalinity (mg/l)	5	62	60	4	4	27±31	5	4	31	2	2	8.80±12.5
Hardness (mg/l)	6.1	40	50	4.6	3.4	20.82±22	4.6	2.8	20	2.1	3.8	6.66±7.52
THC (mg/l)	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01

Table 5: Gross Organic Pollutants for Sampling Stations

Parameter	Experimental Stations						Control Stations				
	A_B	A_T	A_K	A_A	A_E	Mean	B_C T_C	K_C	A_C	E_C	Mean
DO (mg/l)	2.28	2.22	2.16	2.16	2.30	2.22±0.065	2.212.23	2.21	2.27	2.50	2.26±0.15
COD (mg/l)	0.612	1.925	1.810	0.518	0.498	1.07±0.41	0.4940.318	1.318	0.410	0.387	0.58±0.41
BOD (mg/l)	0.408	1.283	1.207	0.345	0.332	0.72±0.49	0.3290.212	0.873	0.273	0.258	0.39±0.27

BOD_5/COD= 0.67 in all the stations

The variations of measured physical parameters relative to the control stations are shown in Figures 1 and 2. The results in Figure 1 indicated that turbidity recorded the most significant variation followed by conductivity from the control which is also analogously replicated in Figure 2. The least changes were recorded for temperature and TDS. The parameters were measured in the scales shown in Table 3 correspondingly. Similarly, Figure 3 and 4 indicated that Alkalinity and Hardness observed the highest variations from the control where salinity and THC were almost negligible, but pH was the least. In a similar fashion but gross organic pollutants, DO was high but low comparative variation, COD and BOD had the highest from the control station values.

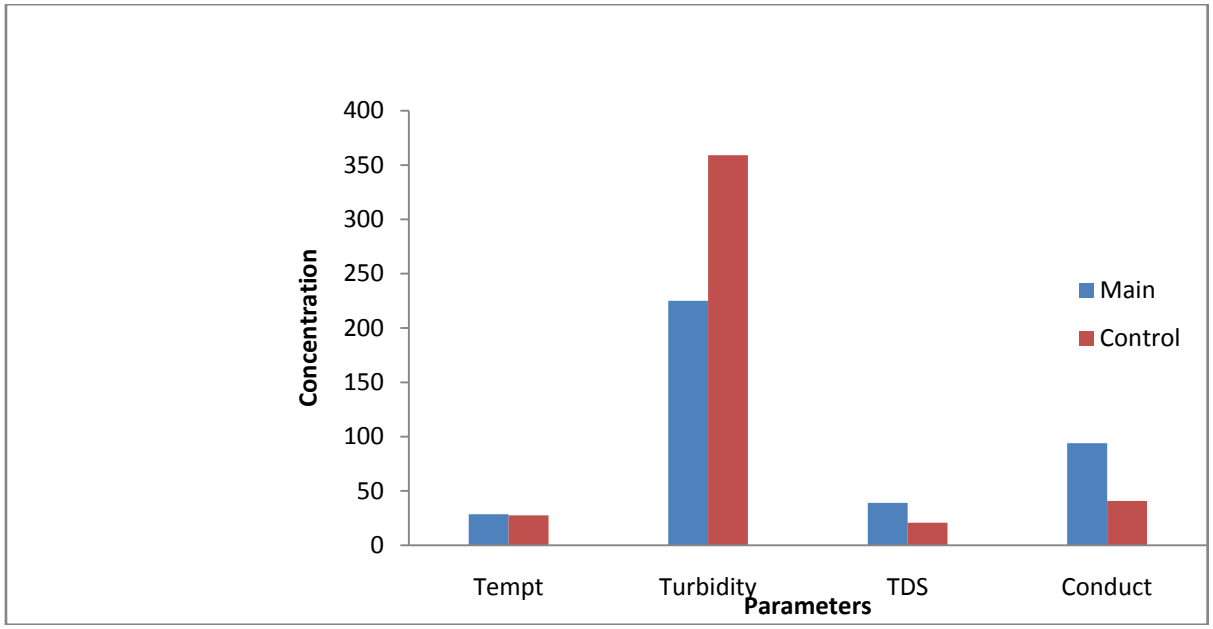

Figure 1: Variation of Physical parameters relative to the Control

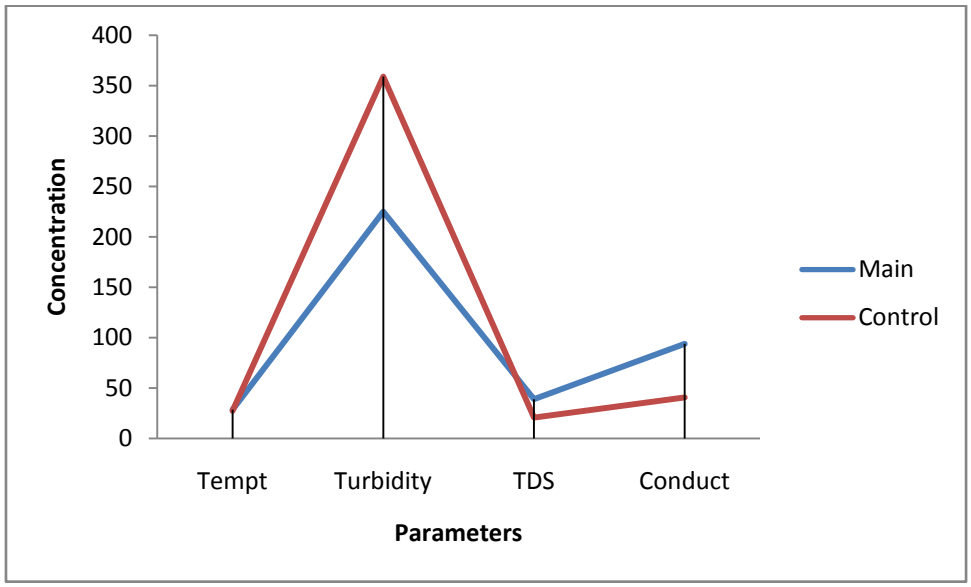

Figure 2: Line Plot of Physical Parameters against Control Values

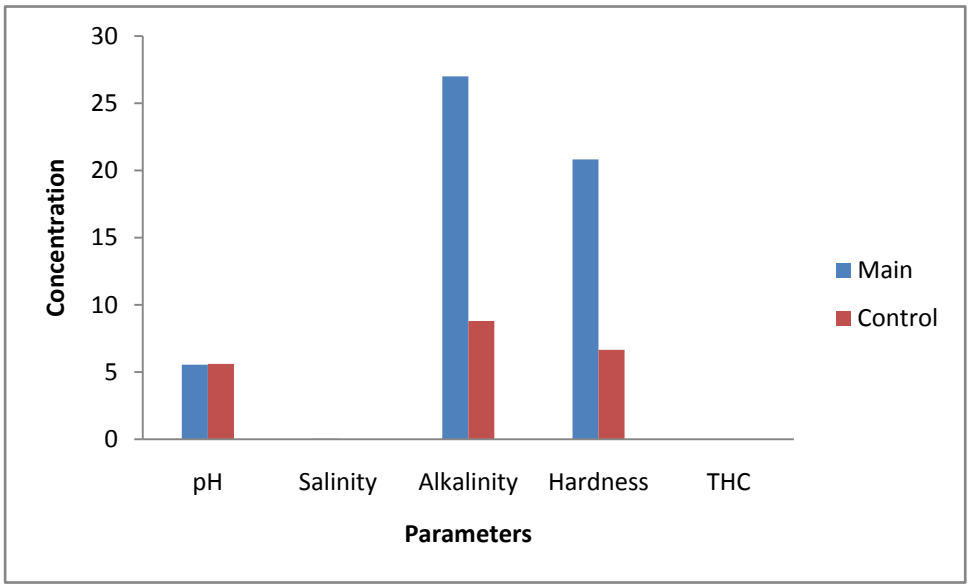

Figure 3: Chemical Parameters compared to the Control

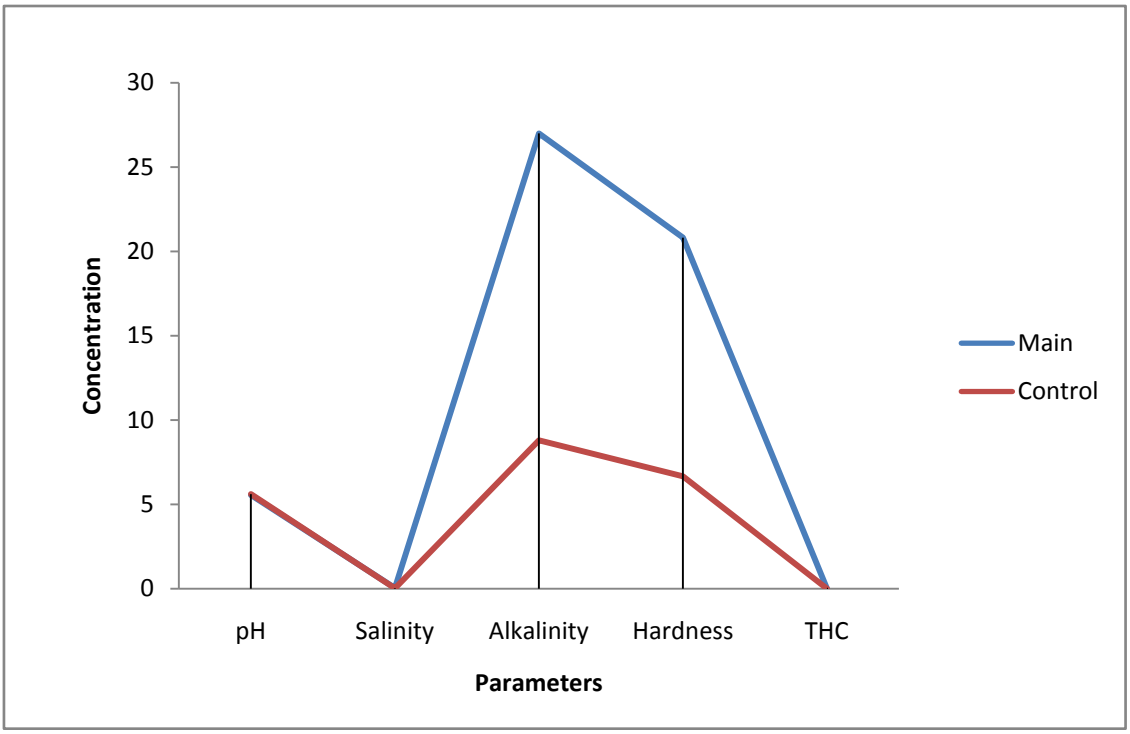

Figure 4: Line Plot of Chemical Parameters versus the Control

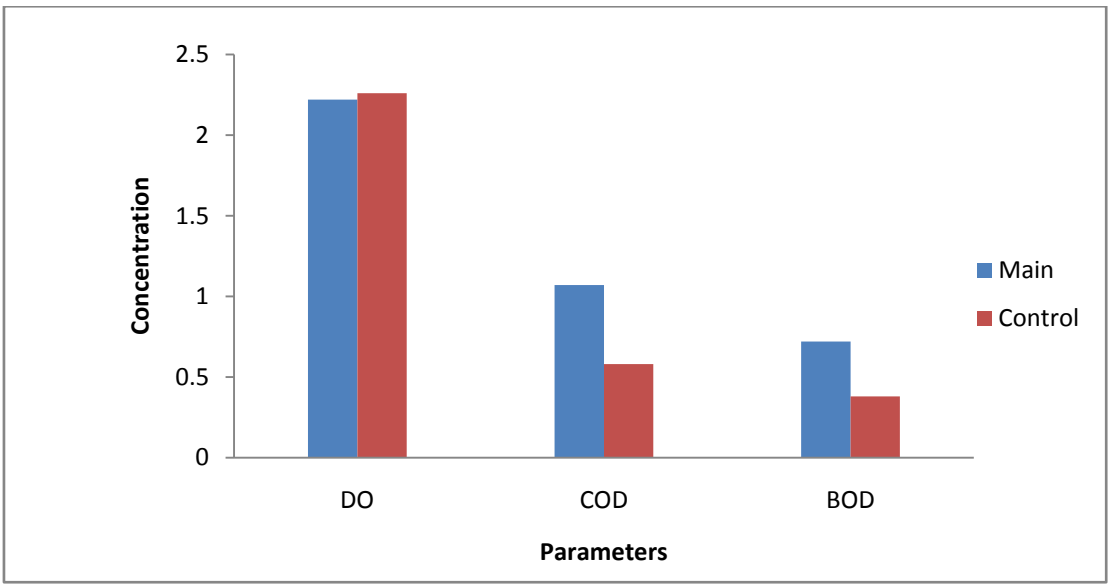

Figure 5: Variation of Gross Organic Pollutants from the Control

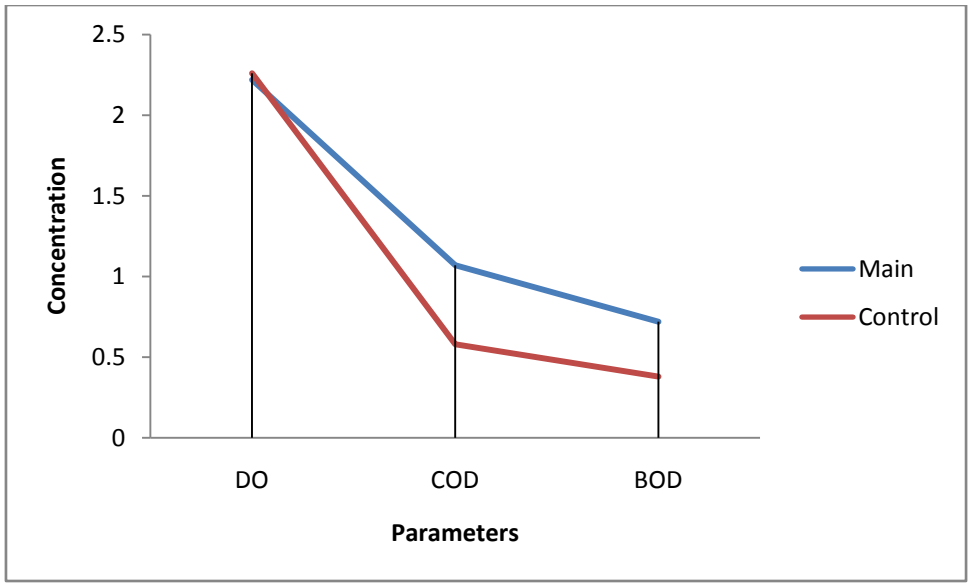

Figure 6: Variation Pattern of Gross Organic Pollutants and the Control Value

Table 8: Environmental Standards for Surface Water Quality

S/N	Parameter	Surface water standards (Drinking water)	Limit for discharge into surface water	WHO	DPR/FMENV
1	Temperature	-	<40	-	35
2	(°C) Alkalinity(mg/l)	-	200	200	200
3	Turbidity (NTU)	-	-	5	40
4	Conductivity(μs/cm)	-	-	-	10,000
5	TSS(mg/l)	-	300	-	-
6	TDS(mg/l)	250-500	-	100	500
7	pH(units)	5-9	6-9	6.5-8.5	7.0-8.5
8	Salinity(PSU)	100-300/350	-	-	-
9	Hardness(mg/l)	-	-	500-1000	100
10	DO(mg/l)	6	-	7.5-10	10
11	COD(mg/l)	5	-	500	40
12	BOD(mg/l)	1.5	50	-	10

IV. Discussion

The result shows the following parameter classified as physical, chemical and gross organic pollutants. The results are shown accordingly in Tables 3 to 5 whereas Tables 1 and 2 are descriptions of the study stations and analytical techniques and methods adopted respectively.

a) Physical Parameters

The water has an objectionable taste in all the study stations and peculiar brownish colour for the control stations of the Sagbama Creek. Temperature of the water samples has a mean of 28.56°C whereas the control stations have a mean of 27.52°C. The highest temperature of 30.45°C was recorded at Amatolu(A_A) sampling station whereas the least was at Bolou-rua station (25.71°C). This gave a range of 25.71-30.45°C. The temperature of surface water is needed to support dissolved oxygen, conductivity, pH, rate and equilibrium of chemical reactions, biological activity, fluid properties and can be used to even classify streams either as cold-water or warm-water (Natural Biological Assessment and Criteria Workshop, NBACW, 2003). The Temperature (°C) for the study stations were within the DPR/FMENV set limits of 35°C (or < 40°C) and discharged in the surface water. This agrees favourably with that recorded for the Woji Creek and Okrika Rivers (Okoye & Chukwuneke, 2008; Iyama & Edori, 2013). The

range 25.71–30.45°C fell within the temperature of 20-35°C which is most suitable for plant growth, but above 30°C may lead to regression in growth and plant decomposition (Kara*et al.,* 2004). The result also agrees with those of other researchers (Iyama & Edori, 2013; Iyama*et al*., 2014).

Turbidity being the composition of clay, silt, finely divided organic and inorganic matter, soluble,coloured organic compounds, plankton, and microscopic organisms gives an indication of the condition and productivity of a water system (NBACW, 2003). The result gave a range of 25.80-670 NTU and a mean value of 225 NTU. This is at variance with those of the control stations 0.20–665 NTU. The minimum turbidity value (25.80) was recorded at the Amatolu sampling point whereas its maximum was at Toru-Orua and the maximum of 665 NTU was at Amatolu control river station (Table 3). These values are above the WHO and DPR/FMENV permissible limits except those at Amatolu, Ebeni (sample points) and Bolou-orua/Toru-Orua control stations. This agrees with the works of different researches who also observed that turbidity is temporary and also increases near areas of turbulence (Iyama & Edori, 2014a, Iyama & Edori, 2014b). This high presence of colloidal solids gives the water the cloudy and aesthetically unattractive and the muddy nature of the area. According to Boyd (1999), the relatively high turbidity may be due to tidal flows, storm, receipt of sediments and particulates from upland (lotic river).

Total dissolved solids (TDS) were ranged between 10 and 89ppm which were recorded at Toru-orua and Kalabiama study stations. The mean TDS value was 39 which is less than the limit for discharge into surface water bodies. The range of 0.00-40 was observed by the control station at Bolou-rua and Toru-Orua control stations but of mean value 20.80mg/l. When TDS levels exceed 1000 mg/l, it is considered unfit for drinking. High TDS values indicated hard water and the presence of toxic minerals which emanated from some dissolved solids of organic origin (Iyama & Edori, 2016). The values for TDS in the study were all below the limits recommended by both DPR and FMENV. Sampling stations recorded the following as well as their corresponding controls; A_B (33:40), A_T (10:0.00), A_K (89:28), A_A (36:16), and A_E (28:20). This showed that organic solids in water were negligible compared to the standard limits.

Electrical conductivity (μS/cm) being the specific conductance is a measure of the potential a body of water has to conduct an electric current. It is a function of the types of and amount of dissolved substances dissolved in the water (NBACW, 2003). It is very important because it gives approximate measure of the groundwater intrusion, correlates with nutrients and can be an indicator of mine or waste water. The highest value of conductivity was recorded at the Kalabiama study station (A_K) as 178 while the least was at Bolou-rua (A_B) as 66. Other stations recorded A_T (75), AA (73) and A_E (77). The mean conductance was 94. In a similar fashion, but in a reverse order, Bolou-rua control station had the highest conductance of 79 (borehole) followed by Kalabiama Kc (56) whereas the least was Toru-orua (1μS/cm) which was also from a tap water. This sharp contrast in conductivity of both tap water sources showed that there were some unique features for further studies. The other control stations recorded the following concentrations of A_C (32) and E_c (36) with mean concentration of 40.80. These concentrations are well below the set limits of DPR/FMENV (Table 8). These values are lower than those recorded for Bassan Rivers, Bayelsa State (Iyama & Edori, 2016). According to Victor and AL-Mahrouqi (1996) decomposition and mineralization of allochthonous organic matter can increase the concentration of conductivity.

b) Chemical and Gross Organic Pollutant Parameters

These include pH, salinity, and alkalinity, Hardness, DO, COD, BOD and THC. The pH which is a measure of the hydrogen-ion activity of water recorded a range of 5.02-6.59. The least pH of 5.02 was recorded at Toru-Orua (A_T) whereas the highest was 6.59 at Kalabiama station. The other stations were A_B (5.46), A_A (5.29) and A_E (5.40). These values showed acidic water, but the control stations also had the following B_c (5.68), T_E (5.19), K_c (5.71), A_C (5.77) and E_C (5.70). The mean pH value for the sampling stations was 5.55 whereas that for the control was 5.50. The pH of a water body can be used for stream classification purposes (either as black water or as white water). This decrease or acidic water can be caused by several factors, including; agricultural activities and acid rain. Though the pH range fell within that recommended values for surface water standards, it is below the lower limits permissible by WHO and DPR/FMENV as shown in Table 8. This is at variance with those reported by several other researchers (Iyama & Edori, 2016; Uwadiae et al., 2009).

Salinity is simply a measure of the salt content of a water body. The mean concentration of salinity was 0.04 PSU. The sampled stations showed that A_B, A_T, A_A had same salinity values of 0.03 but A_E was 0.04 and the highest was at the A_K station of 0.08. Similarly, the control station B_c, T_c, K_c, A_c, E_c recorded 0.04, 0.00, 0.02, 0.01 and 0.01 respectively (Table 4). This showed that the control stations had relatively lower salinity values than the actual samples from the study stations. Kalabiama study station had the highest salinity whereas the three study stations of Bolou-rua, Toru-Orua and Amatolu recorded the least. These values indicated that of a fresh water environment, which was at variance with those earlier reported by other notable researchers (Iyama & Edori, 2016; Edokpayi et al., 2010; Tait & Dipper, 1998). These values were far below those

recommended for surface water standards and even for drinking water.

Alkalinity remains the potential of a water system to neutralize strong acid and very useful for stream classification and to determine susceptibility to acidic deposition (NBACW, 2003). The study stations recorded a range of alkalinity 4-62. The least value was in stations A_A (Amatolu) and A_E (Ebeni) whereas the highest was found in A_T (62) while A_K (60), A_B (5) and mean alkalinity was 27. The control stations of B_C, T_C, K_C, A_C, and E_C recorded 5, 4,31,2,2,12 respectively, whereas their mean concentration was 8.8. These values compared to the sampled stations indicated that there are some significant factors or conditions responsible for the high variation (see Table 4). Alkalinity is mostly produced by the action of ground water on limestone.

Hardness (total hardness) is a measure of the presence of certain insoluble and soluble salts in water, which may be products of calcium and magnesium salts. This may affect the use of soap. The study recorded hardness level in the sampling points for A_B, A_T, A_k, A_A, A_E as 6.1, 40, 50, 4.6, 3.6, 3.4 respectively. The mean values of the samples and control stations were 20.82 and 6.66 respectively. The control stations recorded 4.6, 2.8, 20, 2.1, 3.8 for B_C, T_C, K_C, A_C, E_c respectively. These values were below the permissible limit for water by WHO (1993, 2003), but some stations A_T (40) and A_K (50) had higher total hardness values above the DPR (1991) and FMENV (2001) permissible limits (Table 8). Result of hardness is shown in Table 4. Hardness is generally caused by the presence of Ca^{2+}, Mg^{2+}, $Fe^{2+}$and Sr^{2+}ions in water as they are usually associated with HCO_3^-, SO_4^{2-}, Cl^-, $NO_3^-$and expressed in terms of $CaCO_3$.

Dissolved oxygen (DO) is the amount of oxygen in a body of water which is available for biochemical activities. Result in Table 4 showed that the study stations A_B, A_T, A_k, A_A, A_E, recorded 2.28, 2.22, 2.16, 2.16, 2.30 respectively. Similarly, the control stations B_c, T_C, K_C, A_C, E_C have 2.21, 2.23, 2.21, 2.27, 2.50 correspondingly. These values are practically below that recommended for surface water standards and potable water. This is also below WHO limit of 7.5mg/L (Table 8). The mean values for the sampling stations and the control are 2.22 and 2.28. Water saturated with oxygen is usually of a pleasant taste while the reverse have insipid taste. Clean surface waters are normally saturated with DO, but such DO is readily exhausted by the oxygen demand of organic wastes. These values were far below those of Mustapha *et al*.,, (2013) on similar environment. So many factors affect DO concentration such as temperature which has an inverse relationship.

Biological Oxygen Demand (BOD) refers to the amount of oxygen required for the biodegradation or decomposition of organic matter by micro-organisms. The results from the sampled stations are shown in Table 1 and the results in Table 4 indicated that A_B (0.408), A_T (1.283), A_K (1.207), A_A (0.345), A_E (0.332) and their control stations recorded 0.329, 0.212, 0.873, 0.73 and 0.258 respectively. The mean values for the sampled and control stations were 0.72 and 0.39 correspondingly. These values when compared to permissible standards for surface water (1.5), limit for discharge into surface water (50) and DPR/FMENV of 10mg/l, shows that they are lower, as shown in Table 8. These values are also below those reported by some other studies (Okoye & Chukwuneke, 2008; Iyama *et al*., 2017) but similar to those reported by Iyama and Edori, (2014a) on the water quality of the Imonite Creek, Rivers State.

Chemical Oxygen Demand (COD) which is another form of oxygen demand gives a measure of the oxygen required or demand for the chemical oxidation using $KCrO_4$ and concentrated H_2SO_4. From the sampling stations of A_B, A_T, A_K, A_A, A_E, the COD concentration were respectively 0.612, 1.925, 1.810, 0.518, and 0.498 with their corresponding controls as 0.494, 0.318, 1.310, 0.410 and 0.387 (mg/L). This is shown in Table 4, with mean sampling control station values as 1.07 and 0.58mg/L. The COD values recorded during the study were all below the recommended limits both for discharge into surface water and surface water standards by WHO and DPR/FMENV (WHO, 2003) as shown in Table 8. These results are far lower than those reported by Okoye & Chukwuneke (2008), and Marila & Tamuno-Adoki (2007), on the Woji Creek and Okrika River. Total Hydrocarbon Content (THC) was all through the sampled stations and control less than 0.001mg/L. This showed the near absence of THC in the water bodies of the entire study area. These values are below those recorded for the Ekerikana River as posited by Iyama *et al*., (2017) on similar environment.

c) Nutrient Parameter (mg/L)

Sulphate (SO_4^{2-}), nitrate (NO_3^-), Phosphate (PO_4^{3-}) and Ammonium (NH_4^+) were sampled for and analyzed. The concentration of sulphate in the sampled area is shown in Table 5. The sampled stations and their concentrations are A_B (2.0), A_T (210), A_K (130), A_A (2), A_E (2) whereas their corresponding control stations are B_C (2), Tc(1), K_C(120), Ac(1), E_C(2). The mean concentrations for the sampled stations and controls are 69.2 and 5.04 respectively. The range of sulphate is 2-130mg/L. The high concentration recorded at A_T (Toru-Orua) and A_K (Kalabiama) shows some naturally occurring tendencies by the presence of coal seams or sulphur containing rocks or soils and from acid rains (NBACW, 2003). Sulphate concentration can affect taste and odour of water, changes in surface water, Chemistry and aquatic biota (NBACW, 2003). When compared to limit for discharge into surface water, WHO and DPR/FMNV; the sulphate concentration is relatively

lower, but those of A_T and A_K need be checked to avoid nutrient enrichment (eutrophication) around the rivers and lake. These values were lower than the recommended standard limits and guidelines by WHO and DPR/FMENV. These values were lower than those reported earlier in researches (Ikem *et al.*, 2002; Orebiyi *et al.* 2010; Iyama *et al.*, 2016; Iyama and Edori, 2014a) on similar ecosystems. This was though higher than those reported by other researchers in similar environments (Lahlurja, 2005; Edet, 1993; Olabaniyi & Owoyemi, 2006).

Nitrate (NO^-_3) concentration (mg/L) recorded in the study is shown in Table 4 and are A_B (1.2), A_T (8.2), A_K (2.7), A_A (1.8) A_E (2.1) while the control readings were correspondingly B_C (1.8), Tc(1.7), K_C (5.3), A_C(2.0) and E_C (2.20). The mean for both the sampled stations and the controls are 3.2 and 2.6 mg/L respectively. The range of nitrate concentrations in the study area was 1.2-8.2.The least value of 1.2mg/l was recorded in the sample from Bolou-rua, whereas the maximum was from Toru-Orua. The increased variation may emanate from anthropogenic inputs from natives as nitrate concentration can be affected by combustion of fossil fuels and Agricultural activities. This sharp increase can affect trophic dynamics, increase higher turbidity, decrease DO concentration, increase algal and macrophyte production, as nitrogen-ammonia is also toxic to fish (NBACW, 2003). When compared to both WHO and DPR/FMENV standards, nitrates was found to be lower but close to less than 5mg/L for surface water standards. This result agrees with earlier ones recorded in similar ecosystems by different authorities and researchers (Mustapha *et al.,* 2013; Manila &Tamuno-Adoki, 2007; Emovin, Akporhonor, Akpoborie & Adaikpoh, 2006).

Phosphate (PO_4^{3-}) (mg/L) recorded spatial variations in the study stations as A_B(0.34), A_T (0.73), A_K (0.63), A_A(0.41), A_E(0.28), whereas the control stations have B_C(0.28), T_C (0.18), K_C(1.21), A_C(0.31), E_C(0.19). The mean concentration of phosphates for both sampling stations and control are 0.48 and 0.43 respectively. These phosphate concentrations are lower than those recommended as standard limits by both WHO and DPR/FMNV. These values are in consonance with those earlier reported by other authorities in similar environments (Mustapha *et al.*, 2013; Osibanjo & Majolagbe, 2012; Iyama *et al.*,, 2014).The values obtained showed that the water body meets the standards for salmonids (Mustapha *et al.,* 2013).These values were lower than those recorded by Okoye *et al.*,(2008) which were higher than the DPR/FMENV limit. These concentrations of phosphates are shown in Table 5.Similarly, Ammonium ion concentrations as shown in Table 5 are relatively higher compared to the DPR/FMENV limits for surface water which agreed favourably with earlier researches on similar terrains (Iyama, Eze & Ede, 2014). These concentrations are A_B (0.5), A_T (1.90), A_K (5.9), A_A (0.40), A_E (0.20) whereas their corresponding controls are B_C (0.30), T_C (0.20), K_C(1.20), A_C (0.30) E_C(0.30). The mean ammonium (NH_4^+) concentration for the sampling stations and controls are 1.78mg/L and 0.46mg/l respectively.

V. Conclusion

This study was to determine the surface water quality of a stretch of road from Toru-Orua community through Bolou-rua to Ebeni by the Amasoma Bridge in Bayelsa State. Surface water quality was collected from five sampling stations with corresponding control stations using boreholes and river water. From the results of physical, chemical and gross organic pollutants relevant concentrations levels of specific water quality were determined and compared to WHO, DPR and FMENV standards where applicable.

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Global Journal of Science Frontier Research: B
Chemistry
Volume 19 Issue 1 Version 1.0 Year 2019
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals
Online ISSN: 2249-4626 & Print ISSN: 0975-5896

Glutathione@Fe3O4 Nanoparticles as Efficient Material for the Adsorption of Mercury(II)from Water at Low Concentrations

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Abstract- The application of magnetic nanoparticles (MNPs) in environmental and chemical engineering is progressively increasing, leading to the development of new surface modifications using green methodologies. Magnetite nanoparticles functionalized with Glutathione (Glutathione@MNPs) were synthesized by a novel eco-friendly method using glutathione as a reductant and stabilizer agent.Mercury adsorption was investigated at different initial pH values, contact time, temperatures and adsorbate- adsorbent concentrations. Maximum Hg2+ removal took place at pH 7.5. Adsorption dynamic data were best described by pseudo-second order rate equation, and adsorption equilibrium data were best fitted to Langmuir equation.Maximum adsorption capacities of 34.48 mg/g at 1 mg/L of initial conditions and 25°C was obtained. Regeneration of Glutathione@MNPs and recovery of Hg2+ was demonstrated using 0.1 M KI and HCl up to ten cycles of adsorption.

Keywords: *magnetite nanoparticles, green synthesis, reduced glutathione, adsorption, mercury.*

GJSFR-B Classification: *FOR Code: 030299*

Strictly as per the compliance and regulations of:

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Wilfredo Marimón-Bolívar [α] & Edgar E. González [σ]

Abstract- The application of magnetic nanoparticles (MNPs) in environmental and chemical engineering is progressively increasing, leading to the development of new surface modifications using green methodologies. Magnetite nanoparticles functionalized with Glutathione (Glutathione@MNPs) were synthesized by a novel eco-friendly method using glutathione as a reductant and stabilizer agent.Mercury adsorption was investigated at different initial pH values, contact time, temperatures and adsorbate- adsorbent concentrations. Maximum Hg^{2+} removal took place at pH 7.5. Adsorption dynamic data were best described by pseudo-second order rate equation, and adsorption equilibrium data were best fitted to Langmuir equation.Maximum adsorption capacities of 34.48 mg/g at 1 mg/L of initial conditions and 25°C was obtained. Regeneration of Glutathione@MNPs and recovery of Hg^{2+} was demonstrated using 0.1 M KI and HCl up to ten cycles of adsorption.

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I. Introduction

The contamination by heavy metals in bodies of water is animportant environmental problem since there is a facility of dispersion in great distances due to its biogeochemical cycle, which leads to affect the integrity of the organisms of several ecosystems [1]. Many countries in the world have been highly affected by one of them: Mercury. Contamination due to this metal is a problem that compromises at a global level the food safety and the quality of water for human consumption.[2–4].

Although mercury is released into the environment from natural sources, coal-fired power plants and gold mining have been identified as the largest source of mercury[5,6].For example, 2493.8 tons of mercury were released into the environment from coal combustion between 1978 and 1995. On the other hand, mercury is used in open-pit mining by 15 million miners in over 70 countries in artisanal and small-scale gold mining[7], as it happens in Peru where gold production in Madre de Dios mine was 1583 kilograms of gold in 2016 but that process released an estimated 30–40 tonnes of mercury each year into water[8].

This situation is alarming because mercury does not need to be present in large quantities to generate health risks since it proved to be carcinogenic to mammals due to theirA capacity of accumulation in organisms, biomagnification through the trophic chains and their resistance to biodegradation [9,10]. Methylmercury effects in humans include severe damage to the nervous system, congenital malformations, and even death.These affectations take place becausethe multiple Hg chemical species have hydrophobic properties and a strong affinity for the biological compounds in the sulfhydryl groups as well as DNA binding[11]. Moreover, conventional techniques for the remediation of heavy metals in water such as chemical precipitation, adsorption, ion exchange, membrane filtration, reverse osmosis, coagulation and flocculation, electrochemical treatment techniques, advanced oxidation processes, and adsorption processes have some disadvantages. Problems such as high energy demand, a large amount of chemical compounds required, production of large volumes of waste, lead to the search for optimization or development of these processes with new materials.[12].

Within a large numbers of new materials developed, Iron Oxide Nanoparticles, especially Magnetite (Fe_3O_4), are shown as a promising alternative for the application in environmental remediation since they have a superparamagnetic behavior. This behavior allows controlling the material in such a way that it minimizes its dispersion to bodies of water and therefore possible contamination[13,14]. However, it is necessary to consider that the production of this material could generate a high environmental impact to ensure responsible application.

For this purpose, the green synthesis of magnetic nanoparticles has been studied where plant extracts, marine algae and biomolecules are used as reducing, and stabilizing agents.Around the optimization of magnetic properties necessary for the correct implementation of these materials on a

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large scale, biomolecules have shown better results. However, these green materials have not been applied for metal removal at levels of concentrations like the levels found in the effluents to establish real adsorption potentials.

For these reasons, the adsorption of mercury (II) on magnetic nanoparticles modified with Glutathione (*Glutathione@MNPs*) was studied under different pH values, low concentrations, temperature, and ionic strength conditions. Although there are previous studies[15–17] where glutathione is used as an agent modifier of magnetic nanoparticles, these studies synthesize the magnetic material through the traditional coprecipitation route and then perform a process of functionality. This way to obtain the magnetic material leads to little feasibility in large scale application due to the environmental impact generated, so that this work allows to use glutathione as a synthesis agent and modifier in a single step which would allow the implementation of this process in environmental remediation tasks.

II. Experimental

a) Materials

Iron (III) chloride hexahydrate ($FeCl_3 \cdot 6H_2O$), sodium hydroxide (NaOH) and $HgCl_2$ standard solution (1000 mg/L) were purchased from Merck. Reduced L-Glutathione ($C_{10}H_{17}N_3O_6S$) was purchased from Sigma-Aldrich. All chemicals were analytically pure and used as received without further purification, and all solutions were prepared with double-distilled water.

b) Synthesis of Fe_3O_4 nanoparticles by a green method using glutathione

Fe_3O_4 NPs obtained through the green method (*Glutathione@MNPs*) were synthesized using a methodology described in a previous work[18]. Briefly, 10 mL of a $FeCl_3$ (0.1 mol/L) solution was added to a 100-mL beaker under vigorous mechanical stirring at 75°C. On reaching this temperature, 20 mL of an aqueous solution of L-Glutathione (0.214mol/L) was added dropwise into the beaker with the pH value of the mixed solution adjusted to 10. A temperature value of 85°C was reached and it remained in agitation for one hour. Then the nanoparticles were separated by magnetic field application, washed several times with deionized water and alcohol and dried in vacuum at 40°C for 12 hours.

c) Adsorption studies

Glutathione@MNPs obtained by a green route were added at 25°C to aqueous solution of mercury with concentration of 1 ppm prepared from Cl_2Hg standard solution at different pH values (2-10) adjusted with solutions of (0. 1M) NaOH or HCl with concentration of nanoparticles of 100 mg/L and a stirring speed of 140 rpm for a standard time of 2 hours. The final concentration of the Hg(II) ions was measured using atomic absorption spectroscopy. The adsorption capacity is calculated on the difference of concentrations at the beginning and the end of the process[19, 20]:

$$q_e\left(\frac{mg}{g}\right) = \frac{V(C_0 - C_f)}{m}$$

Where q_e is the adsorption equilibrium of the capacity (mg/g), Co and C_f are the initial concentrations and the equilibrium (mg/l) of the ion in the solution, V is the volume (L) of solution and m is the mass (g) of used adsorbent. Considering the favorable conditions of adsorption tests, kinetics and adsorption isotherms were studied.

d) Adsorption kinetics and adsorption isotherm

The adsorption capacity of Hg (II) ions was studied as a function of time. Therefore, an optimal contact time of the adsorption of Hg (II) on the nanoparticles was established and under this equilibrium time condition, the adsorption kinetics was studied. To determine an adequate kinetic model, the adsorption was evaluated in four equations (Table 1)[21,22].

Table 1: Kinetic and isothermal models used

Kinetic model	Mathematical expression
Pseudo-First Order	$q_t = q_e(1-e^{-k.t})$
Pseudo-Second Order	$q_t = \frac{t}{\left(\frac{1}{(k_2 \cdot q_e^2)}\right) + (t/q_e)}$
Elovich	$q_t = \frac{1}{\beta}\log(\alpha\beta) + \frac{1}{\beta}\log(t)$
Intraparticular diffusion	$q_t = k\sqrt{t} + C$

Isotherm	Mathematical expression
Langmuir	$\frac{C_e}{q_e} = \frac{C_e}{q_m} + \frac{1}{K_L q_m}$
Freundlich	$q_e = K_F C_e^{1/n}$
Temkin	$q_e = Bln(A) + Bln(C_e)$
Sips	$q_e = \frac{q_m (K_F C_e)^{1/n}}{1 + (K_F C_e)^{1/n}}$

Where k_1 is the constant of Pseudo-First Order (min^{-1}), k_2 is the kinetic constant of Pseudo-second order (g/(mmol·min)), α constant of Elovich's equation (mmol/(g·min)); (g/mmol), k diffusion constant, q_e adsorption capacity at equilibrium (mmol/g), q_t adsorption capacity at the measured time (mmol/g)[19, 23], C equilibrium concentration (mmol/L), q_m amount of metal or contaminant needed to form a surface monolayer (mmol/g), K_L is the equilibrium constant of Langmuir, and K_F is the equilibrium constant of Freundlich. N is the exponent of each characteristic equation [24–27].

Alternatively,adsorption isotherms were carried out by changing the initial concentration of nanoparticles, while the initial metal concentration was 1 mg/L and a constant temperature value was 20°C. The experimental isotherms used for the study of the elimination of metal ions are shown in Table 1.

e) Adsorption thermodynamics

To establish parameters for process scaling studies, the enthalpy change (ΔH^0), free energy of Gibbs (ΔG^0) and entropy (ΔS^0) were established by the measurement of the adsorption capacity at different temperatures and the subsequent application of the following equations[28]:

$$\Delta G^0 = -RT \cdot lnk_L$$

$$lnk_L = \frac{\Delta S^0}{R} - \frac{\Delta H^0}{RT}$$

$$\Delta S^0 = (\Delta H^0 - \Delta G^0)/T$$

f) Competitive study with ions

To determine the involvement of ions in the adsorption of mercury on the nanoparticles, lots of tests were carried out with concentrations (0-30 mg/L) of 4 ionic species that are frequently found in bodies of water (magnesium, calcium, zinc).The addition of the ions was carried out with sulfate salts because the sulfate ions are frequent in wastewater. The experiments were carried out in triplicate, and the favorable conditions found in the previous sections were kept constant (pH, time and dose of the material).

g) Study of adsorption capacity in real water

For the application in real water, samples from strategic sites in Colombia were selected, specifically in the department of Bolívar and Cundinamarca. The samples were filtered with a 450 mm filter to simulate the water pre-treatment processes that eliminate the suspended material.

h) Adsorption cycles

Desorption of heavy metals was studied with acid and saline eluents (Hydrochloric Acid HCl, Sodium Chloride NaCl, potassium iodide KI) as a function of its concentration according to the *Lehningerprinciple* for the precipitation of proteins using high ionic strength. The nanoparticles used for the adsorption of the mercury ions are diluted in a volume equal to that of the adsorption batches at a stirring speed of 130 rpm and for a time of 1 hour. The degree of desorption was calculated from the following expression:

$$\%Desorption = \frac{Adsorbed\ Metal}{Recovered\ Metal} \times 100\%$$

III. Results & Discussion

a) Characterization of the nanomaterial

The nanoparticles obtained by a green synthesis using Glutathione were characterized to establish the magnetic properties, size, surface chemistry, crystallographic properties, and elemental composition. FTIR analysis showed characteristic peaks of magnetite. The peaks assigned to the iron oxide core can be observed at 580 cm^{-1} (FeO). Moreover, a low-intensity band was evidenced at 3477 cm^{-1}, which is attributed to amine groups (NH and NH_2), and the peak recognised to the SH group was detected at ~2500 cm^{-1} which is typically very weak. Furthermore, bands were perceived at 2920 cm^{-1}, which is associated to CH sp3 bonds[29,30]. Instead, it is important to note that the CO stretch band of the carboxyl group, which is representative of the Glutathione spectrum at 1710 cm^{-1}, is absent. This result can be explained by the bonding pattern of the carboxylic acids on the surface of the nanoparticles. Strong adsorption at 1000 cm^{-1} arises from the stretching of the CO single bond. These results revealed that glutathione was attached to the Fe_3O_4 nanoparticles as a carboxylate [31].

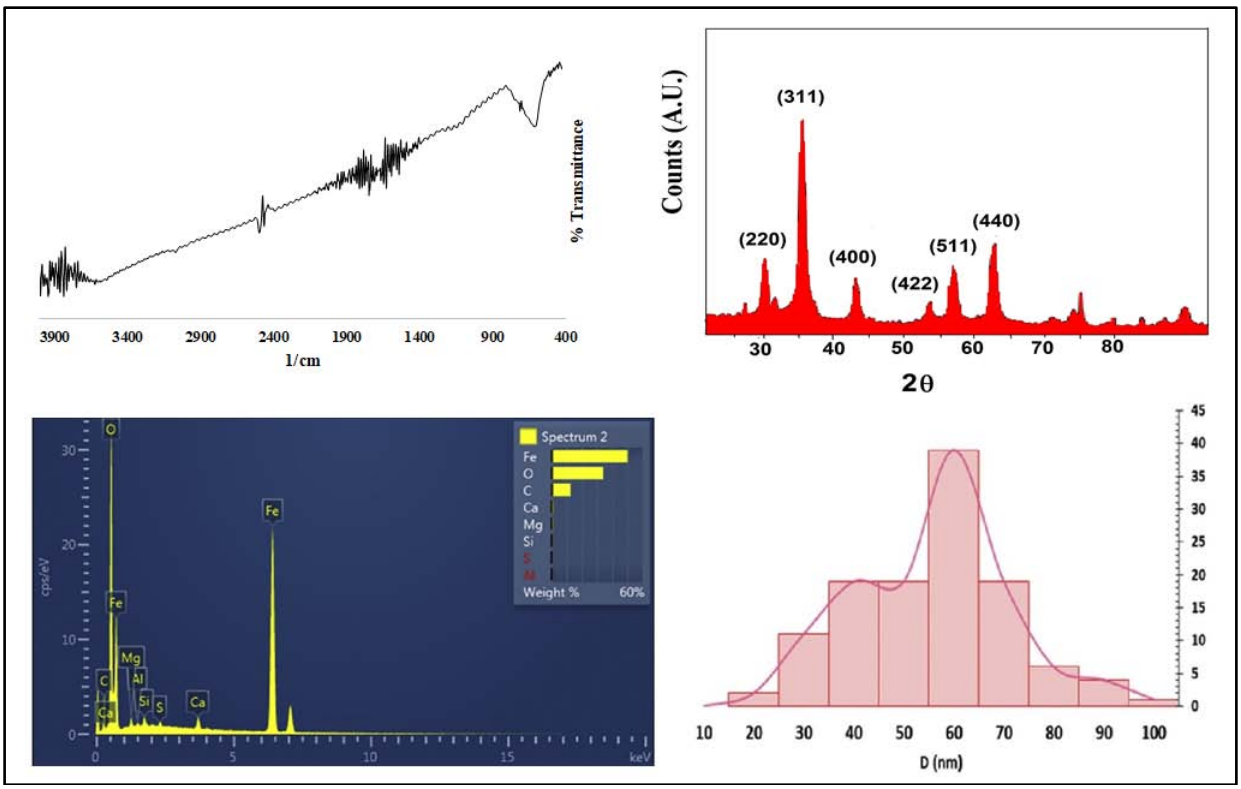

Figure 1: Characterization (FTIR, XRD, EDS, DLS) of Glutathione@MNPs.

XRD characterizations were carried out with CrKα and Cu radiation (1.5406 Å). The angular range was set between 20 and 80° with a scanning speed of 0.04°/s and a step size of 0.04°/s. Fig. 4a and b show the diffractogram of the Glutathione@MNPs. This diffractogram shows indexed peaks as (220), (311), (400), (422), (511) and (440). These peaks correspond to a cubic spinel structure. The crests of XRD were indexed using data from the International Centre for Diffraction Data (ICDD) database. The calculation of the lattice constant yields a value of 8. 30 Å which is close to the value for magnetite: 8. 39 Å. Furthermore, this structure is recognized for having the peak (311) with the highest intensity.

The EDS spectrum is shown evidence the presence of sulfur, which is part of the thiol groups active on the surface according to the FTIR spectrum. The hydrodynamic size was∼ 60 nm, while it can be deduced that a greater dispersion in the size of nanoparticles is obtained through green synthesis in comparison to chemical co-precipitation method. Also, this material did not show a reduction in its magnetic properties (Saturation magnetization of 84.5 emu/g) as is usually seen in magnetic nanoparticles obtained by green synthesis, which would make it possible to be used in scale processes [32].

b) Effect of pH

pH is asignificant variable in the behavior of the compounds in the aqueous medium and therefore for the adsorption process since it affects both the protonation of the functional groups of the surface of the adsorbent nanomaterial and the predominance of the chemical species present in aqueous solution. For this reason, adsorption batch studies were performed in a pH range of 3 to 10 (Figure 2).

The adsorption process is favored when the pH values are close to 7 and 8 (neutral) while in very acidic and basic conditions it was reduced. This occurs because an interaction with protonated mercury ions (Hg^{2+}) takes place when the surface charge of the material is negative (-35mV), as in this case when the pH is close to 7.5[18]. Likewise, in these pH values the functional groups SH-, NH-, NH_2, and COOH- are negatively charged, which favors the electrostatic interactions. Otherwise, in acid pH values, amino and carboxyl groups tend to acquire a neutral or positive charge that gives rise to repulsive forces. For high pH values, the adsorption potential decreases due to the abundance of OH- ions that react with the Hg^{2+} ions to form precipitable compounds, which is not conducive to the adsorption process[33–35].

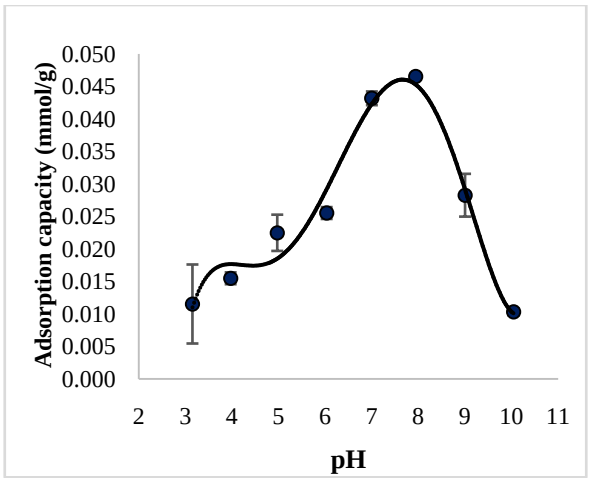

Figure 2: Effect of pH on Hg (II) ions adsorption with *Glutathione@MNPs* (Initial concentration of Hg (II) 1 mg/L; dosage *Glutathione@MNPs* 100 mg/L; T= 25°C; time 60 minutes).

c) Adsorption kinetics

Equilibrium time and adsorption kinetic of Hg(II)on *Glutathione@MNPs* were determined. Figure 3 shows how equilibrium time for the adsorption of Hg(II) was reached in 30 minutes approximately since there is no significant difference with adsorption times after 5 hours.This equilibrium time shown is greater than somereported studies [36,37], which would imply a lower adsorption rate.

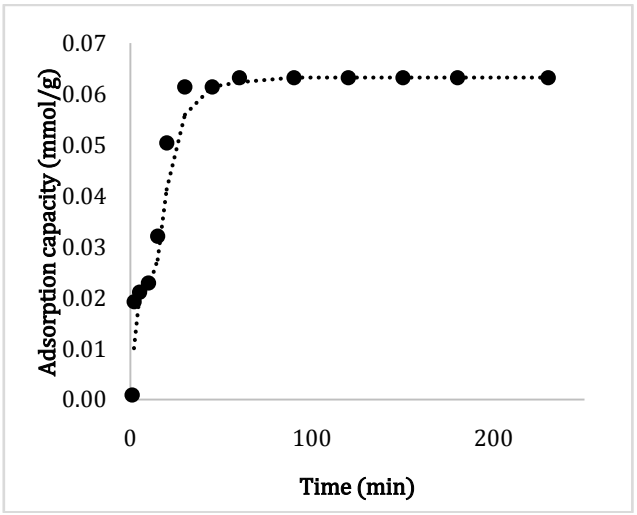

Figure 3: Effect of contact time on the adsorption efficiency of Hg(II). Experimental conditions (1 mg/L Hg, nanoparticle dose 10 mg/L, pH = 8, 140 RPM, 25 ° C).

However, the experimental conditions are different, since the initial concentrations of the metals used are higher compared to the studies reported, especially in the case of mercury. This difference leads taking more time in the process of reaching the steady state since there is a very high amount of metal in the aqueous medium compared to the concentration of the nanomaterial.

To give clarity to the kinetic phenomenon, an experimental data model with kinetic models (Supplementary Figure 1) in its linear form was established. From the linearized adjustment, it can be observed that the kinetic model that best adapts to the experimental data and that can be used to describe the kinetics of adsorption of Hg (II) on *Glutathione@MNPs*is the Pseudo Second Order model (Table two). This means that the speed at which the interaction between the adsorbate and the adsorbent occurs is dominated by the chemical interactions (chemisorption), which prevail over the transport of adsorbate from the liquid phase to the hydrodynamic layer located around the particle, thus as the transport of this hydrodynamic layer to the surface of the adsorbent[38].

Table 2: Kinetic model parameters for adsorption of Hg (II) andCr(VI) by *Glutathione@MNPs*.

		Value
Pseudo-first order	q_e (mmol/g)	0.075
	K_1 (min^{-1})	0.036
	R^2	0.9271
Pseudo- Second order	q_e (mm/g)	0.066
	K_2 (mmol $g^{-1}min^{-1}$)	1.828
	R^2	0.995
Elovich	α (mmol/(g.min))	1.4e-5
	β (g/mmol)	2.9274
	R^2	0.980
Intraparticular Diffusion	K_i (mmol·g^{-1}·$min^{-1/2}$)	2.554
	C (mmol/g)	12.63
	R^2	0.872

It should be noticed that the diffusion limiting effects (mass transfer) could also be minimized by the agitation applied to the system so that transport of ions from the liquid to the surface of the material was facilitated. Similarly, Pseudo-second order model describes how adsorption occurs only at localized sites and does not imply an interaction between the adsorbed ions and that the adsorption energy does not depend on the surface coverage, as well as the absorption of metallic ions in the coals activated is governed by a second order rate equation [39]. From the above, it can be established interaction of Hg ions with the (-SH) groups on the surface of the nanomaterial are the dominant ones in the process, which would serve as an ideal adsorbent for removal contamination in waters by mercury ions.

d) Isothermal studies of the adsorption

Adsorption isotherms describe the equilibrium of the adsorption of a material on a surface at a constant temperature. At high proportions of metal concentration relative to the nanomaterial, the concentration gradient serves as a driving force to overcome the resistance to mass transfer that promotes a favoring of adsorption[40]. The effect of the nanomaterial dose on the efficiency of heavy metal adsorption was carried out at 20°C with different amounts of*Glutathione@MNPs* (10-100mg/L) using a 50 ml batch with an initial concentration of 1 mg/L of mercury (figure 4). As can be seen, the adsorption capacity of the material decreases with the increase of the adsorbent dose.

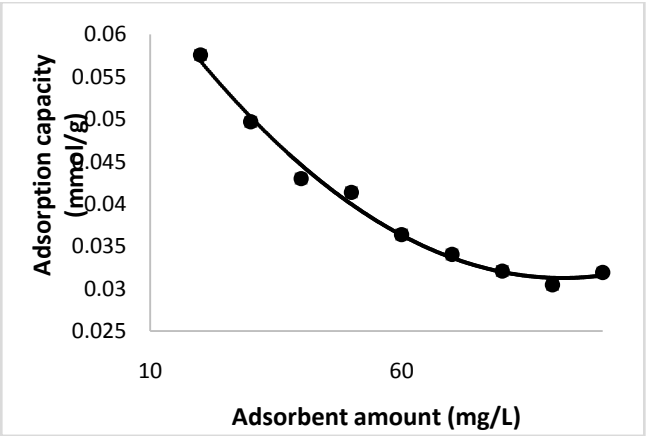

Figure 4: Adsorption isotherm for a) Hg (II)

This phenomenon happens because when the initial concentration of metal used is high, MNPs with the availability of active functional groups decreases concerning the mercury ions. Conversely, when there is a large amount of MNPs, and the final concentration of the metal is lower, the saturation of the active sites is not reached and, therefore, a poorer capacity of adsorption is expressed. For example, a 50% decrease in the dose of nanomaterials (from 100 to 50 mg/L) only leads to an increase in the final mercury concentration from 1.8 to 2.9 mmol/L, but the adsorption capacity of 33 increases at 43 mmol/L.

To determine the adsorption mechanism and the maximum adsorption capacity, the linear adjustments of the isothermal models were made (Supplementary Figure 2). It can be seen that for mercury adsorption, Langmuir equation fits better to the experimental data, which suggests monolayer adsorption on a homogeneous surface. This adsorption is associated with thiol groups, and there is negligible interaction between the species adsorbed[41,42]. These results lead to the fact that,

due to the adsorbate /adsorbent interaction, the adsorption energy decreases linearly as the deposition of metal ions increases on the active sites [26,27,35]. The parameters resulting from these linear models are shown in Table 3.

Table 3: Parameters of adsorption isotherms

	Parameter	Value
Langmuir	Q_m	34.843
	KL	0.479
	R^2	0.9827
Freundlich	KF	11.648
	N	1.257
	R^2	0.9822
Temkin	A	1.714
	B	10.82
	R^2	0.9775
Sips	q_m	36.135
	KF	0.0007
	N	0.991
	R^2	0.981

Here we can notice that all correlation coefficients were higher than 0.92, so that, to a certain extent, the four adsorption isotherm models could be used to describe the Hg(II) adsorption equilibrium, but results would be found more in line with the experimental with Langmuir. From the parameters obtained, it is detected that the maximum adsorption capacity (q_m) of *Glutathione@MNPs* on Hg (II) is 36,126 with a Freundlich coefficient of *1/n* of 0.795 that is less than 1, which expresses an outstanding affinity between the adsorbent and metal. Compared with other studies (Table 4), the adsorption capacity shown by the material is satisfactory since, although it is not one of the highest, the initial concentration of mercury for the studies is lower and this is reflected in the adsorption capacity[43,44].

In other words, for the application in remediation, where mercury contamination values are around 0.1 ppm, working with concentrations higher than 10 ppm gives an erroneous adsorption capacity so, when repeating the studies for the materials compared with contractions equal to the one used in this study, the reported adsorption capacities will be much lower. A proof of this is the capacity of adsorption shown in [36] is 74.85 mg/g being the MOF (Metal Organic Framework) one of the materials with the largest surface area and hence very high adsorption capacities but with a high environmental impact when this material is obtained.

Table 4: The Maximum adsorption capacity of several adsorbents on Hg(II).

Material	q_m/C_0 (L/g)	Ref
Dendrimers of polyamidoamine oxide on magnetic graphene	1.162	[45]
Chelation fiber functionalized with thiourea	0.20	[46]
Modifiedtitaniumdioxidenanotubes	0.05	[34]
Chitosan stabilized magnetic iron sulfide nanoparticles	4.42	[47]
Nano-absorbent PGMA (poly glycidyl methacrylate) magnetic functionalized triethyleneteramin	1.08	[48]
Exhausted coffee waste	6.35	[49]
Alkynyl carbon materials	1.61	[50]
Modified silica gel surface with chelating ligand	1.14	[51]
l-Cysteine functionalized bagasse cellulose nanofibers	0.58	[52]
Synthetic FeS and natural pyrite	7.69	[53]
Aminophosphonic acid functionalized polyacrylonitrilefiber	3.68	[54]
This work	34.88	-

e) Adsorption thermodynamics

The study of adsorption thermodynamics offers more information regarding the viability of the adsorption process and the evaluation of thermodynamic parameters such as the enthalpy energy change (ΔH^0), entropy (ΔS^0), Gibbs free energy (ΔG^0) which are significant parameters for the engineering application of the treatment system [55]. For this, adsorption tests were carried out at different temperatures (20-30 °C) as shown in (Supplementary Figure 3). Adsorption capacity was not favored by the increase in temperature. For the determination of the thermodynamic parameters, the graphs of Ln *K* were made concerning the inverse of the temperatures, where the enthalpy and entropy changes are obtained from the intersection, and the slope of the straight line acquired. These values are shown in Table 5. The positive values of ΔH^0 reveal the endothermic nature of the adsorption process. Likewise, these enthalpy givesindicative of

the adsorption mechanisms since it can be used to determine the source of the interaction force that exists between the adsorbent and the adsorbate, indicatingthe binding strength.

Many studies show a decreasing behavior of the capacity of adsorption when the temperature increases, which leads to ΔH^0 being negative. However, other studies have shown an increase in the ability of adsorption when the temperature increases leading to ΔH^0 being positive [56–60]. This difference in behavior is due to the chemical nature of the species that take place in the adsorption[61].

Values of ΔH^0 between 4 and 40 kJ/mol are characteristic of physical interactions, while values of 40 to 800 kJ/mol correspond to chemical interactions [62].

The negative ΔG^0 for the adsorption of Hg (II) suggest that this process is spontaneous and favorable. The positive ΔS^0 indicated the increase in randomness during the adsorption or disorder in the solid-liquid interface that is a sample of the dynamic phenomena of desorption adsorption even at equilibrium.

Table 5: Thermodynamic parameters of the adsorption of Hg on *Glutathione@MNPs*

T (K)	$\Delta G^0(\mathbf{kJ \cdot mol^{-1}})$	$\Delta H^0(\mathbf{KJ \cdot mol^{-1}})$	$\Delta S^0(\mathbf{KJ \cdot mol^{-1} \cdot K^{-1}})$
293	-11.021	45.702	0.118
298	-10.404		
303	-9.828		

f) Adsorption Mechanisms

Based on the previous results (nanomaterial characteristics and the thermodynamic study), the adsorption mechanism of Hg(II) ions on *Glutathione@MNPs* could be established. The process was carried out mainly by ion exchange on the surface of the nanoparticles due to the resulting loads and chemical formations categories of both the metal and the nanomaterial in their dependence on the pH. This formations happens because sulfur is considered to be an electron donor atom that can present complexes with weak metal acids such as mercury[63,64]. Since, a pH close to the neutral values, the mercury species formed ($Hg(OH)_2$, $HgOH^+$ y Hg^{2+}) interacts with the thiol groups in this way:

$$-SH + Hg^{2+} \rightarrow SH - Hg^+$$

$$-SH + HgOH^+ \rightarrow SH - HgOH$$

$$-SH + Hg(OH)_2 \rightarrow -SH \cdots Hg(OH)_2$$

$$-OH + Hg(OH)_2 \rightarrow -O \cdots Hg(OH)_2^{1-} + H^+$$

Because the surface of the nanomaterial is homogeneous (thiols, hydroxyls, amines and carboxyl groups) different adsorption mechanisms take place, giving priority to the route governed by the chemical affinity of the thiols towards mercury ions, but when these are saturated adsorption processes physical in the other ligands take place. The oxygen atoms attached to the hydroxyl groups attached to the iron atoms may also behave as weak bases that interact with the mercury ions. [65]:

$$2(-OH) + Hg^{2+} \rightarrow (-O)_2 \cdots HgOH + H^+$$

$$-OH + HgOH^+ \rightarrow -O \cdots HgOH + H^+$$

This proposed mechanism can be supported in the same way by the interpretation of FTIR spectra before and after adsorption (Supplementary Figure 4). There is no remarkable difference in the peaks expressed at~ 1628 cm^{-1} (carboxyl groups) or 3480cm^{-1} (amino groups) before and after adsorption. For this reason, it is proposed that the active adsorption sites active for the adsorption of mercury would be the- SH groups (Figure 5 and Supplementary Figure 5).

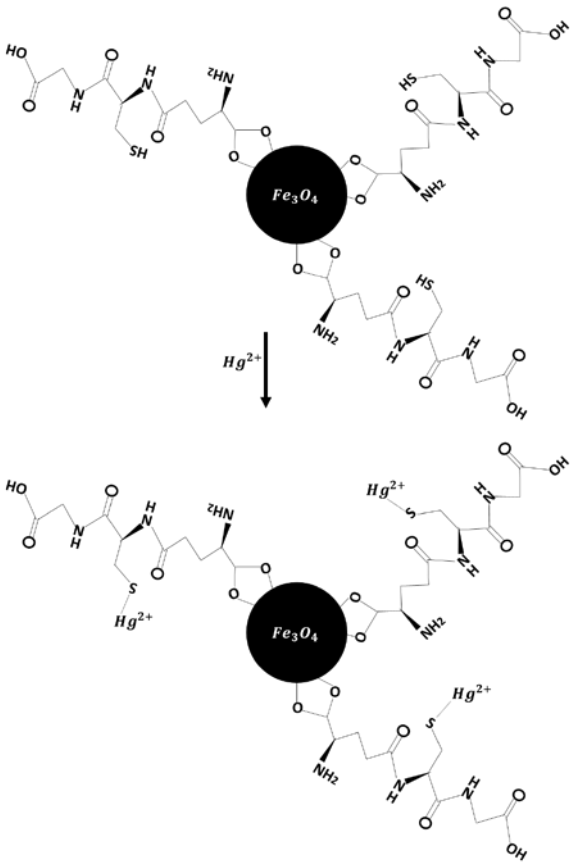

Figure 5: Proposed mechanism adsorption for Hg(II) on *Glutathione@MNPs*.

g) Ion interference

The competitive adsorption of ions coexisting to the binding sites is usually a problem when conventional adsorbents are used for the removal of heavy metals. To investigate the effect of coexisting ions, mercury solutions were prepared with cations of an equal or similar charge.In Figure 6, the behavior of nanomaterial is presented in the presence of calcium, magnesium, sodium and zinc ions concerning the adsorption of mercury. As evidenced in [66], the improvement in the capacity of adsorption of mercury ions occurs with the increase of the ionic strength with cations such as Na^{+}, Zn^{+}, Mg^{2+}, Ca^{2+}.

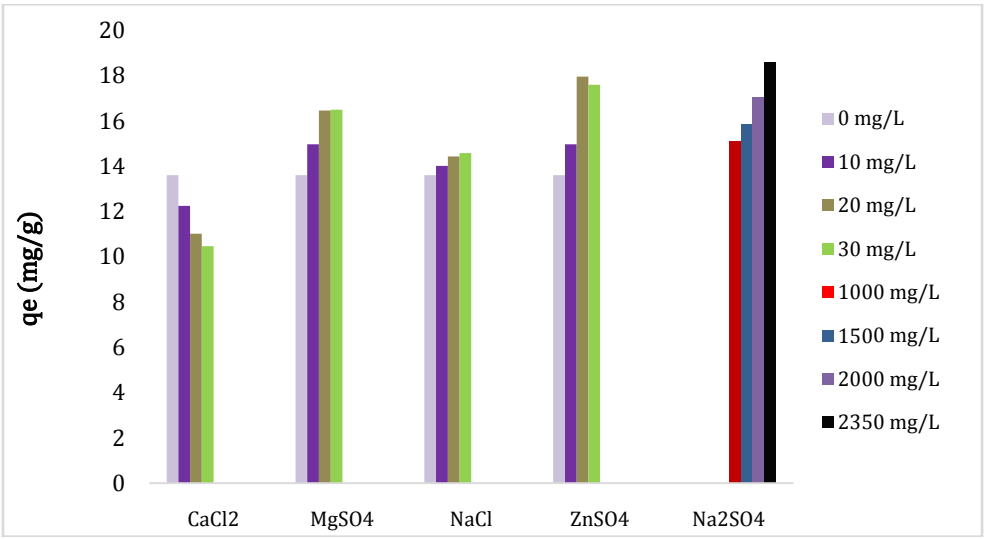

Figure 6: Metal ion interference in Hg (II) adsorption

In this case, the presence of calcium cations interfered in the adsorption process decreasing up to 10%. This could be due to the agglomeration of the nanoparticles which leads to a decrease in the available surface area and the presence of Cl^{-} anions from the calcium precursor salt used, which form complexes with the mercury ions, thus decreasing the interaction with thiol groups [67]. In the case of the influence of sulfate ions, interference is studied with concentrations around the limit of allowed discharges (~1500 mg/L). It can be seen that when sulfates coexist with mercury ions, the adsorption process is not affected, but on the contrary, there is a favoring of the interactions that

can be associated with the fact that sulphate ions could eliminate electrostatic repulsions that mercury could have with the positive charges of the surface of the material, decreasing the entropy of the system and thus optimize the adsorption.

h) Desorption and reusability

To evaluate the possibility of regeneration and reusability of the *Glutathione@MNPs* as an adsorbent, batch desorption experiments were conducted. From Figure 7, it can be observed that for the concentrations studied potassium iodide (KI) is the one that best performs the process. This favourability in the process shown by KI is given that the iodine ions have a high affinity for the Hg(II) atoms that cause the phenomenon of saline displacement in comparison with the dissociation and union of mercury with the Chlorine or hydroxyl ions.

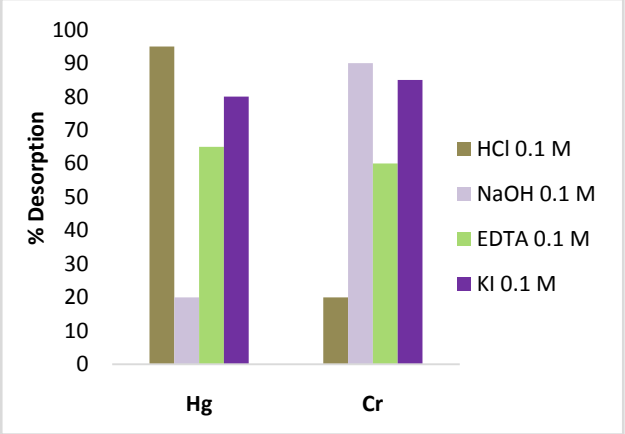

Figure 7: Percentages of recovery of Hg and Cr using different eluents.

Reuse tests showed a decrease in the percentage of adsorption higher than 20% only after the tenth cycle of mercury adsorption (Figure 8). These results could be an indicative for the favorability in the application of this material in process at scale.

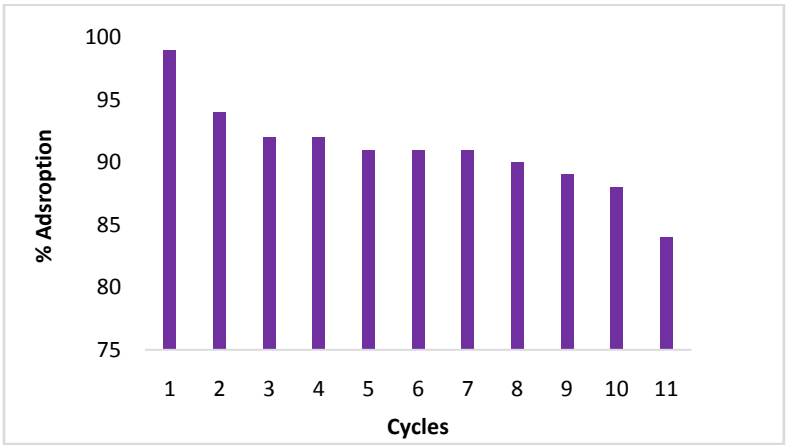

Figure 8: Consecutive adsorption cycles for the adsorption of Hg on *Glutathione@MNPs*.

i) Tests with real water

To survey capability of *Glutathione@MNPs* for removal of Hg (II) from real water, some batch experiments were conducted. Concentrations of the initial physicochemical parameters before adsorption were analyzed, and the results were summarized in Table 6. It should be noticed that the assessment of the presence of mercury and the best evaluation of the adsorption potential of *Glutathione@MNPs* on mercury ions, made an addition of 100 ppb of the metal before treatment, which is a value 10 times higher than the average of the limits of wastewater discharges allowed by resolution 0631 from Colombia for the different economic sectors[68]. The number of nanoparticles added to each treatment was 40 mg/L with an adsorption time of 1 hour at 25°C and a stirring speed of 130rpm.

Table 6: Characteristics of surface and residual water before and after treatment

		Bogotá River	Wetland	San Francisco River	WWTP	San Jorge River
pH		6.30	7.36	5.99	5.32	6.05
Dissolved oxygen (mg/L)		3.4	4.9	6.3	1.5	4.01
DQO (mg/L)		58	45	143	459	50
Conductivity (μS/cm)		272	501	681	747	303
Sulphate (mg/L)		5	11	1	8	6
Nitrates (mg/L)		0.1	0.1	ND	0.3	0.1
Hg (II) (ppb)	Before	1928	1067	1116	984	582
	After	246	153	156	565	78

The results indicated that Hg (II) in real water was removed, and the removal efficiency of Hg(II) reached an average of 78%. It can be observed that in rivers and wetland the percentage of removal is greater than 85% indicating capacity of adsorption. In the case of WWTP, adsorption capacity was not equally favorable (less than 50%), may be due mainly to the physicochemical conditions found, especially the acid character that is lower than the other trials. Likewise, the conductivity value of the sample is higher, which indicates a presence of ions in solution that could interfere in the adsorption process, so an initial pre-treatment of pH adjustment and a decrease of conductivity could increase the efficiency of the process.

These results show that, despite the coexistence of other compounds and non-ideal laboratory characteristics, the function of adsorption of material towards mercury ions is not affected, which would indicate an implementation potential in this type of waters.

IV. Conclusions

The results obtained confirmed that the material of *Glutathione@MNPs* can eliminate mercury (II) ions from aqueous solutions efficiently. This adsorption depends strongly on parameters such as initial metal concentration, pH, contact time and coexisting ions.

The adsorption process follows an isotherm behavior of Langmuir for mercury ions with maximum adsorption capacity on mercury ions of 34.8 mg/g under conditions of mercury concentration of 1 mg/L.

The adsorption kinetics follows a Pseudo-second order regime for mercury with an equilibrium time of Less than 30min. The coexistence of metals from real waters such as magnesium and zinc does not interfere in the removal of mercury. The thermodynamics of the process shows that the interaction with the mercury ions is of chemical order (especially with the functional groups's thiols and hydroxyls) since it expresses binding energy of 45.7kJ/mol.

Acknowledgments

The authors would like to thank the Geophysical Institute of the Faculty of Engineering of the Pontificia Universidad Javeriana for providing support with experimental infrastructure; the Government of the Department of Bolivar.Colombia and its program "Bolivar Ganador" for financing doctoral studies programs; and Dr. Adriana Herrera for her contributions in the characterization of Magnetite Nanoparticles.

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Global Journal of Science Frontier Research: B
Chemistry
Volume 19 Issue 1 Version 1.0 Year 2019
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals
Online ISSN: 2249-4626 & Print ISSN: 0975-5896

A Review Exploring the Coordination Chemistry of Active Methylene Groups Hydrazones

By B. A. Salah, A. T. Kandil & M. G. Abd-El-Nasser
Helwan University

Abstract- Compounds pos sessing a m ethylene bridge located between two strong electron withdrawing groups (MACs,e.g., diketones, dinitriles, benzoyl acetonitriles, cyanothioacetamide, ethyl acetoacetate, diethyl malonate ethyl cyanoacetate, cyanoanilides, etc.) have also been of significant interest in coordination and organomettalic chemistry of hydrazones, for instance, -diketones are important starting materials in many organic synthetic reactions, particularly in the perfume and cosmetic industries.

GJSFR-B Classification: *FOR Code: 030207*

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A Review Exploring the Coordination Chemistry of Active Methylene Groups Hydrazones

B. A. Salah [α], A. T. Kandil [σ] & M. G. Abd-El-Nasser [ρ]

Abstract- **Compounds possessing a methylene bridge located between two strong electron withdrawing groups (MACs, e.g., diketones, dinitriles, benzoyl acetonitriles, cyanothioacetamide, ethyl acetoacetate, diethyl malonate ethyl cyanoacetate, cyanoanilides, etc.) have also been of significant interest in coordination and organometallic chemistry of hydrazones, for instance, -diketones are important starting materials in many organic synthetic reactions, particularly in the perfume and cosmetic industries.**

I. Introduction

Aryl azocompounds (AACs) have ubiquitous motifs in heterocyclic chemistry and are widely utilized as azo dyes, PH-indicators, pigments, food additives, initiators for some radical reaction, therapeutic agents, etc. [H. Zollinger, et al., (1995), H. Nishihara, (2005)]. The coordination science of AACs has been extensively studied throughout the last decades on account of potential combination of important functional properties with a diversity of molecular structure of their complexes. Thereby, these complexes have attracted attention in variuos fields, such as synthesis of conducting and magnetic equipments, Light-by-Light Scattering equipments, supramolecular chemistry, biological activity, catalysis and bioinorganic chemistry of some complexes, among others [B.G. Gowenlock, et al., (2005), Roglans, et al., (2006), Pettinari, et al., (2003)].

Compounds possessing a methylene bridge located between two strong electron withdrawing groups (MACs, e. g., diketones, dinitriles, benzoyl acetonitriles, cyanothioacetamide, ethyl acetoacetate, diethyl malonate ethyl cyanoacetate, cyanoanilides, etc.) have also been of significant interest in coordination and organometallic chemistry of hydrazones, for instance, -diketones are important starting materials in many organic synthetic reactions [B.G. Gowenlock, et al., (2005), Roglans, et al., (2006)], particularly in the perfume and cosmetic industries [Pettinari, et al., (2003)]. Moreover, several representatives of MACs have been widely used in organic and organometallic chemistry for a long time and have recently been the object of increasing attention as components of multidentate ligands in metallo-supramolecular chemistry. Thus, the rich coordination chemistry of -diketones has been reviewed recently. The chemistry of cyano and ester-substituted MACs concerns such organic reactions as dimerization, hydrolysis, halogenation, reduction, carbonyl condensation hydrazones, ortho-ester and ylide formation; they are widely applied in industrial synthetics of, e.g., herbicides, corrosion inhibitors, dyes, polymers, washing and bleaching compositions, catalysis, lubricants, and optical sensitizers [Z. Rappoport, (1990),]. Numerous inorganic template transformations [G. Aromi, et al (2008), P. A. Vigato, et al., (2009), F. Hibbert, et al., (1990)] added an exicting interest on cyano-substituted MAC for metal-mediated preparation and coordination science.

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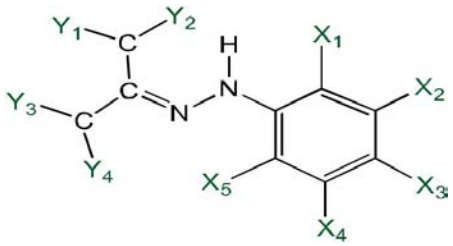
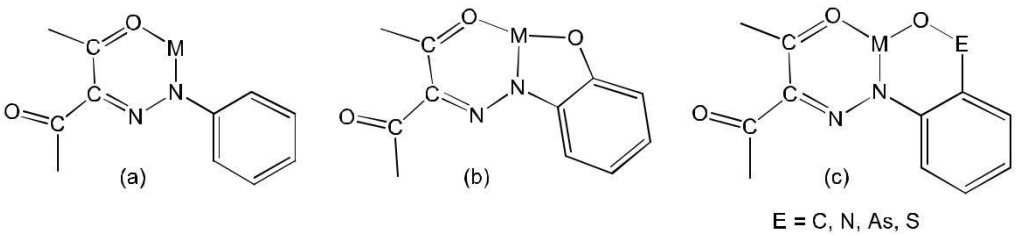
II. Literature Survey

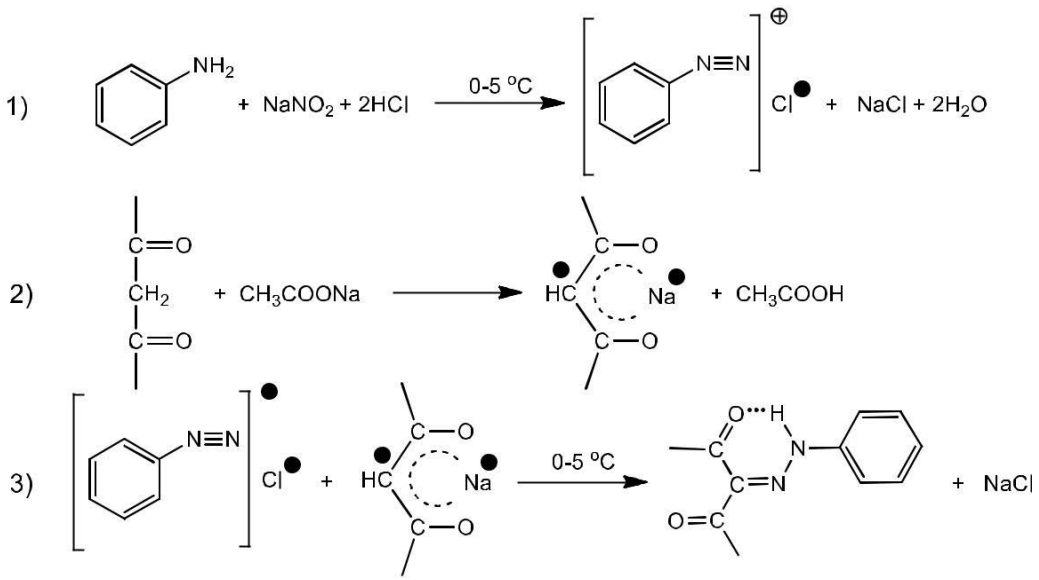
The first methylene active arylhydrazones (Scheme 1) were reported as early as 1883, by Richter and Müntzer, then selected as "benzolazoacetone" [M.N. Kopylovich, et al., (2011)] that was five years later represented by Japp and Klingemann to be an aryl-hydrazone [M.N. Kopylovich, et al., (2011)]. Until now, the synthesis of this category of AHMACs consists in the coupling of a MAC with an aromatic diazonium salt, mostly performed in methanolic, aqueous or ethanolic solution in presence of acetate as catalyst (Scheme 2) [J. Sokolnicki, et al., (1999)]. This reaction was considered very important that the title Japp–Klingemann became a standard reaction name [F. Huang, et al., (2005)]. In its course, the amine requires an early diazotization with an aqueous solution of sodium nitrite in acidic medium at very low temperature to produce the corresponding diazonium salt, which is very important part of the full Japp–Klingemann conversion. The earlier scheme underwent many modifications and improvements, e.g., higher yields of more pure products can be obtained when the coupling is undertaken in an alkaline medium such as solution of sodium hydroxide instead of sodium acetate (Scheme 3). On the other hand, the acidic medium used in the diazonium salt formation process strongly affects the yield and stability of diazonium salt.



Scheme 1: Aromatic hydrazones of methylene active compounds (AHMACs).

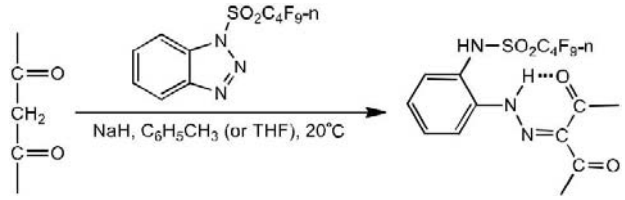


Scheme 2: Possible chelating modes of coordination within the AHBD complexes.



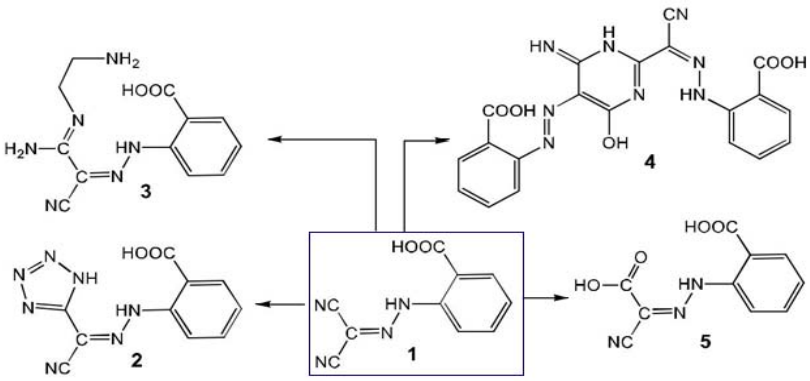
Scheme 3: Japp – Klingemann synthesis of AHBD [W. Kuznik,et al., (2011)].

The Japp–Klingemann method has been modified to an immense variety of AHMACs derived from various aromatic amines and MACs. The AHMAC products act as anintermediates in a large number of synthetic reactions of organic molecules or as ligands in coordination chemistry [R.A. Aliyeva, et al., (2009), L. Hao, et al., (2008)].



Scheme 4: Synthesis of AHMAC with N- nonafluorobutyl Sulfonyl benzotriazole [M. Uhde,et al.,(2010), M.N. Kopylovich, et al.,(2013)].

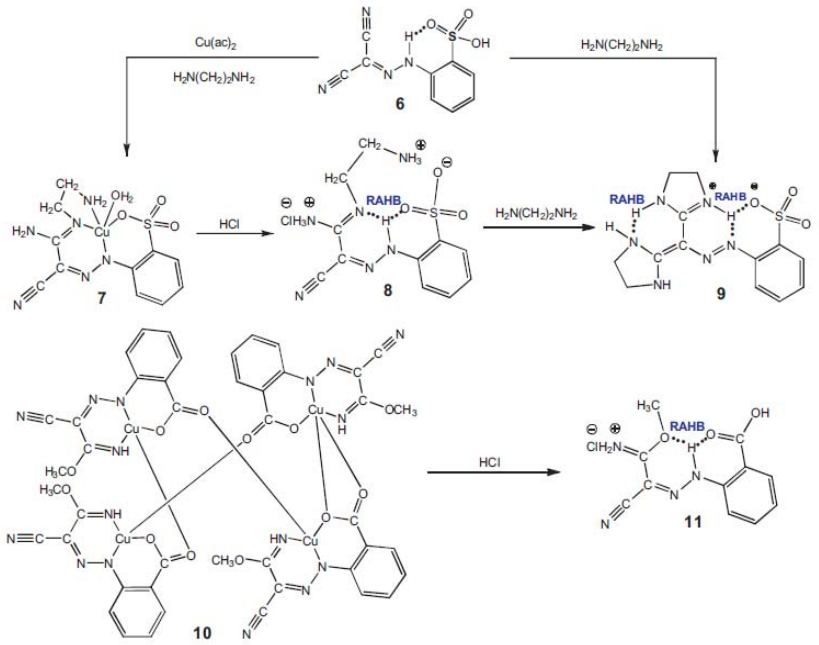
[Z. Chen, etal., (2006)] provided a different approach to the synthesis of AHMACs includes the reaction of a MAC with n-nonafluoro butylsulfonylbenzotriazole in the presence of sodium hydride in an inert medium (Scheme 4). However, the synthesis of n-nonafluoro butylsulfonylbenzotriazole is not trivial, and this preparative procedure has not been frequently used.



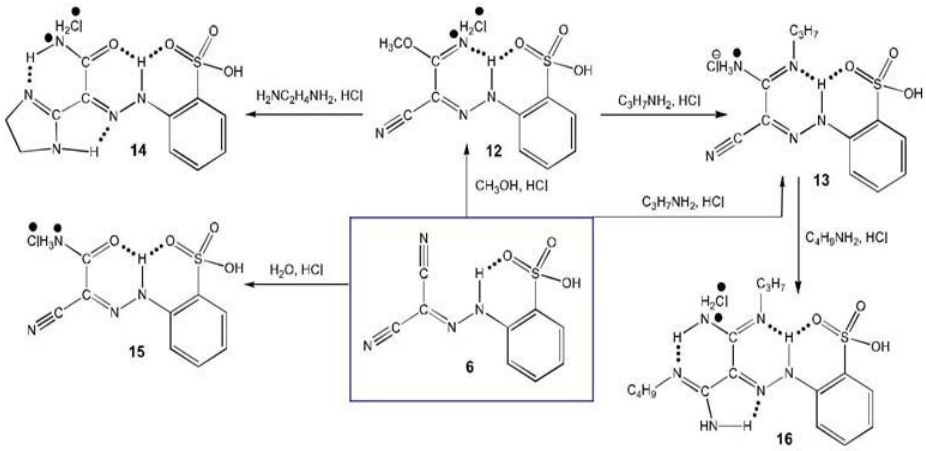
Scheme 5. Template metal-mediated formation of AHMAC ligands from 1.

Scheme 5: Template metal- mediated formation of AHMAC ligands Form 1.

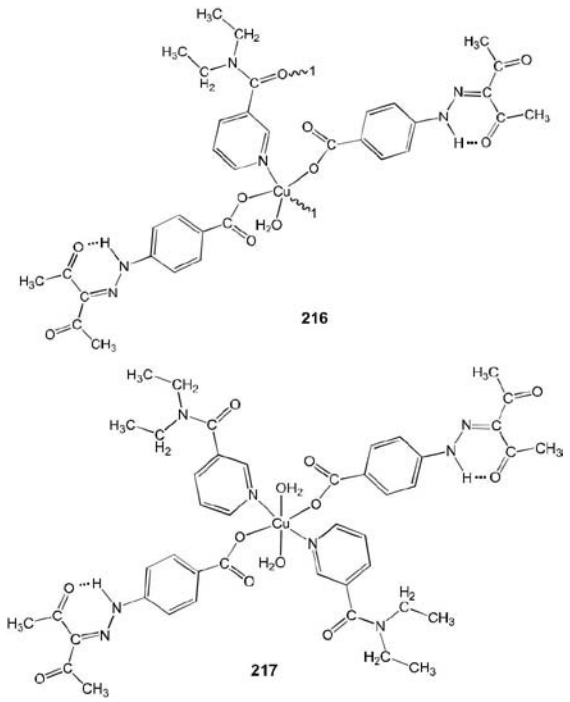
A new template synthesis of AHMAC ligands has recently been reported (Scheme 5), where one cyano group of a starting dinitrile **1** undergoes a metal (Mn)-assisted Nu reaction with different nucleophiles producing the corresponding unsymmetric products **2**–**5**. Nevertheless, the thus prepared ligands **2**– **5** were not separated and fully characterized. Later, it was concluded that similar AHMAC ligands can be synthesized upon copper(II)-mediated synthesis and easily separated on account of the formation of resonance assisted hydrogen bond (RAHB) (Scheme 6) [M.N. Kopylovich, et al (2011)]. The RAHB can also facilitate different organic preparation of symmetric and unsymmetric AHMAC compounds (Scheme 7). In this Case the cyano group of the starting compound **6** is easily hydrolyzed to amidine, carboxamide and iminoester derivatives **12**–**16** depending on the nucleophiles attacking the cyano moieties and conditions used.



Scheme 6: Template synthesis and RAHB- promoted liberation of AHMAC ligands



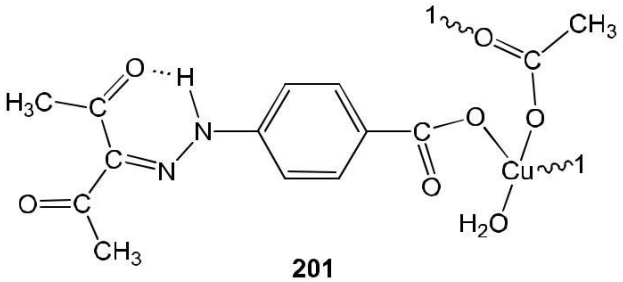
Scheme 7: Synthesis of AHMAC Via RAHB



Scheme 8

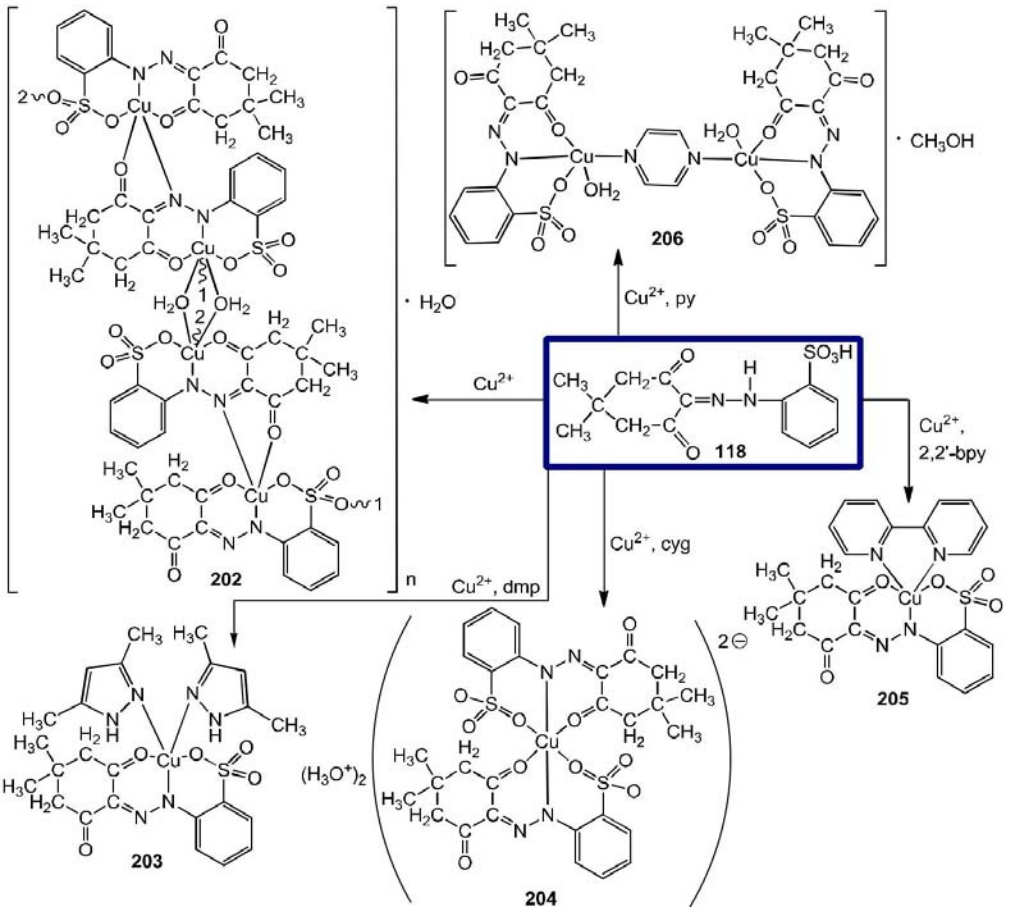
[M.N. Kopylovich, et al., (2011)] prepared new copper (II) complexes **216** and **217** (Scheme 8) by reaction of copper II acetate hydrate with N,N-diethylpyridine-3-carboxamide (cardiamine) in the presence and absence of Na_2CO_3, respectively. The Cu^{II} atom in **216** exist in the plane defined by the oxygen atoms of the carboxylate ions, the nitrogen atom of pyridine moiety and the water molecule, while the apical coordination to the amido oxygen atom of an adjacent N-heterocycle leads to the polymeric chain formation.

Reaction of Cu^{II} acetate with **50** gave the polymer complex **201** (Scheme 9) [D.Z. Mijin, et al., (2010)]. The RAHB system stays intact and the Cu^{II} ions are complexed through coordination bonds by three carboxylate oxygen atoms from one AHBD ligand and two acetates.



Scheme 9: Ref. [M.N. Kopylovich, et al., (2012)].

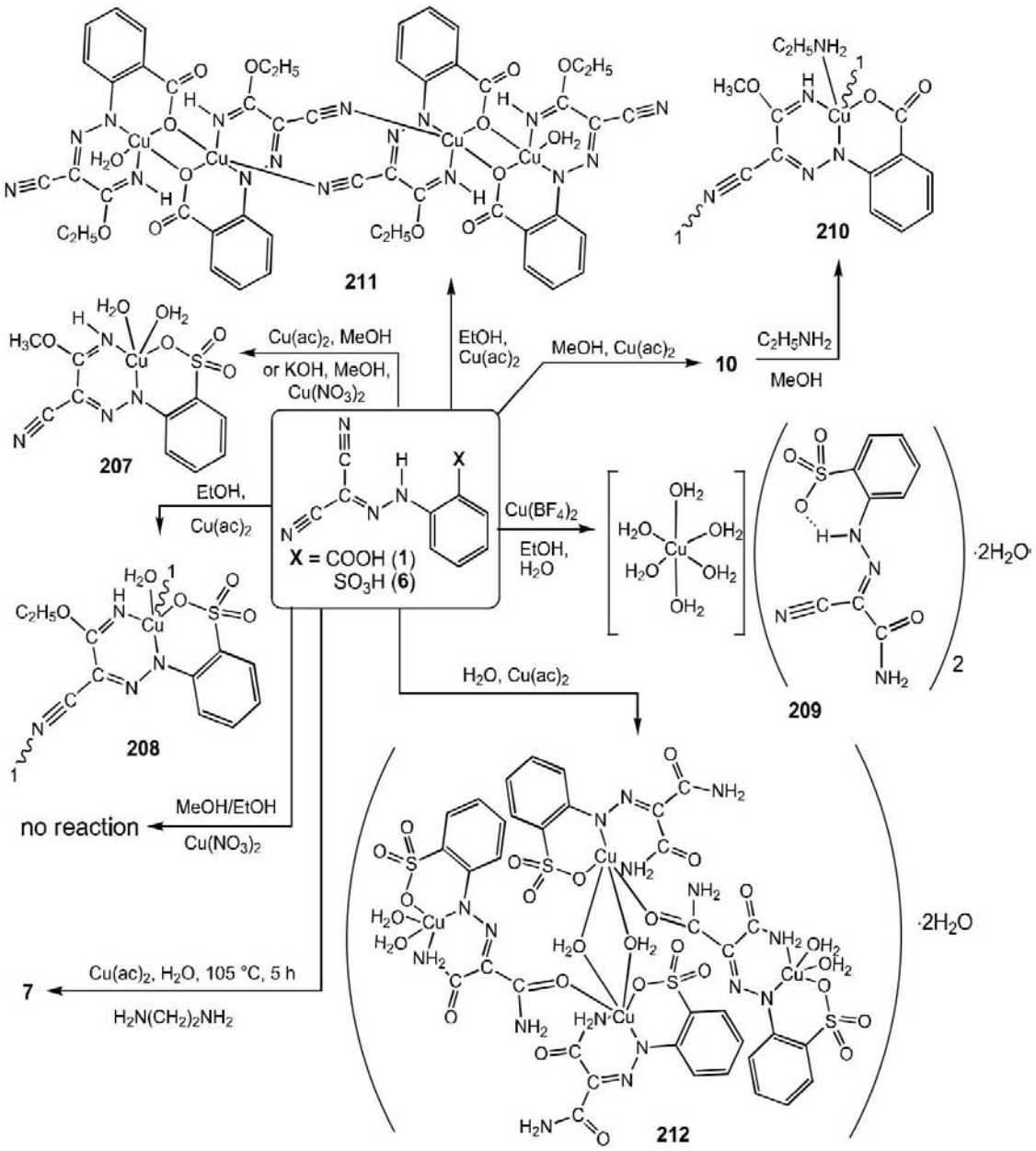
The Cu^{II} –AHBD complexes **202**–**206** were prepared by reacting **118** with a Cu^{II} source in the presence of auxiliary ligands (Scheme 10). In **202**, water molecules act as bridges connecting two dimeric structural units, while a coordination polymer is formed by ligation of the sulfo- group to Cu^{II}. **202** is the first example in which both nitrogen atoms of the former hydrazone group are simultaneously coordinated to Cu^{II}. Pyrazine bridges two monomers forming **206**. **202**–**206** were applied as catalysts in the oxidation of cyclohexane to cyclohexanol and cyclohexanone [X. Li, et al., (2006)].



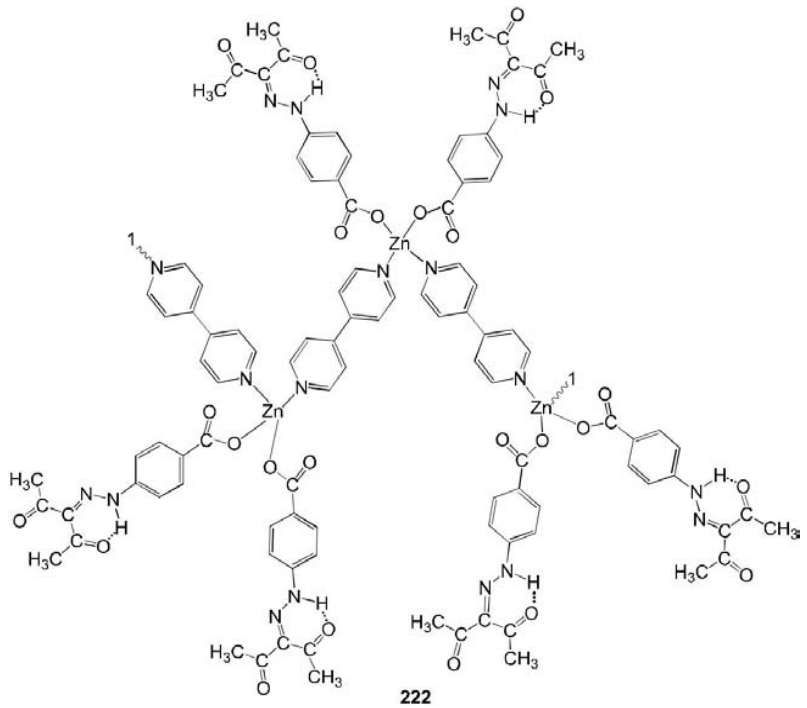
Scheme 10

[M.N. Kopylovich, et al., (2011)] studied the reactions of Cu^{II} with **1** and **6** in the presence of methyl alcohol, ethyl alcohol, water, ethylamine and ethylenediamine, nucleophilic attacks to the cyano moieties occur leading to a variety of different ligated amidines, carboxamides and iminoesters depending on the starting ligands, attacking nucleophiles, solvent and conditions used. Mononuclear **7**, **207–209**, tetranuclear **10**, **211**, **212** and polymeric **208** and **210** complexes were thus synthesized (Scheme 11) Selective oxidation of primary and secondary alcohols to the corresponding carbonyl compounds, as well as diastereoselective nitroaldol (Henry) reaction catalyzed by the complexes were studied, affording typical yields of 80–99% .

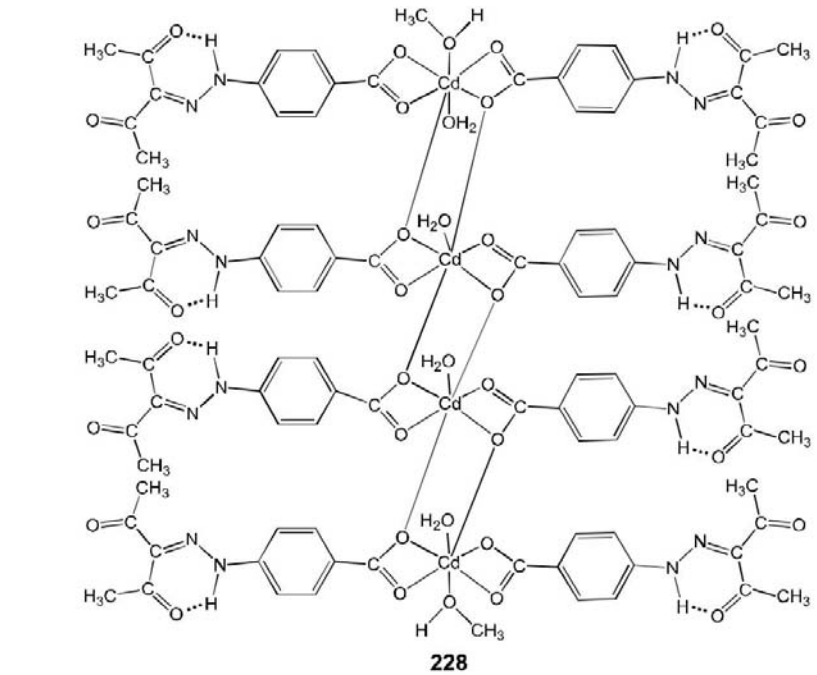
The coordination polymer **222** (Scheme 12) has been prepared by the reaction of the sodium salt of **50** with Zn^{II} sulfate and 4, 4 -bipyridine [D.Z. Mijin, et al., (2010). The Zn^{II} ions are coordinated by two carboxylate oxygen atoms from two monodeprotonated **50** and two nitrogen atoms from two 4, 4 -bipyridines. The metal ions are linked Treatment of **50** with cadmium II nitrate and cadmium II acetate afforded complexes **228** and **229**, respectively (Scheme 13) [Bustos, et al., (2007)]. In **228**, four Cd^{II} ions are bridged by bridging monodeprotonated **50** leading to a tetranuclear core. In the polymer **229**, the CH_3COO^- anions bridge the Cd^{II} ions forming 1D chain involving chelating monodeprotonated **50** units in the side chain. Each Cd^{II} atom is seven-coordinated, and is in a single-cap triangular prism environment with three oxygen atoms from two $OOCCH_3$ units, two oxygen atoms from one chelating **50** and two oxygen atoms from two water molecules. Thus, in the solid state structure of **229**, linear chains are bonded by intermolecular hydrogen bonds forming a 2D sheet. These 2D sheets were packed each other through Van der Waals forces forming a 3D supramolecular structure.

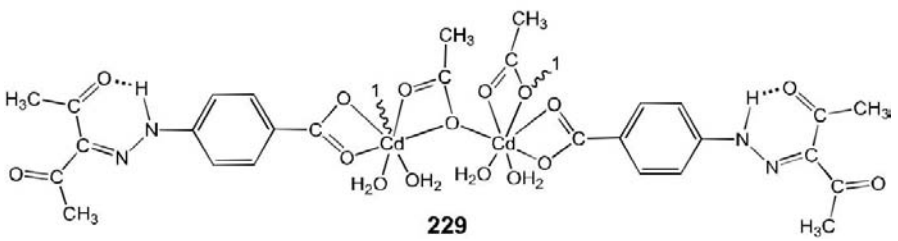


Scheme 11: Reactions of 1 and 6 with Cu^{II} [J.Marten, et al.,(2010)].



Scheme 12: Ref. [J.Marten, et al.,(2010)].





Scheme 13: Ref.[J. Marten, et al., (2008)].

III. Conclusion

Arylhydrazones complexes have a great activity against different types of bacteria. They have been used in several of industrial synthetics of, e.g., herbicides, corrosion inhibitors, dyes, polymers, washing and bleaching compositions, catalysis, lubricants, and optical sensitizers and are widely utilized as azo dyes, PH-indicators, pigments, food additives, initiators for some radical reaction, therapeutic agents, etc. The coordination science of AACs has been extensively studied throughout the last decades on account of potential combination of important functional properties with a diversity of molecular structure of their complexes.

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Global Journal of Science Frontier Research: B
Chemistry
Volume 19 Issue 1 Version 1.0 Year 2019
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals
Online ISSN: 2249-4626 & Print ISSN: 0975-5896

Proximate and Mineral Analysis of Watermelon Sold at North Bank Market, Makurdi, Nigeria

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Abstract- The study was carried out on proximate and mineral analysis of water melon sold at North bank market Makurdi. Water melon samples were purchased at North bank in January, 2018. The samples were sliced using a knife to separate the seeds, pulp and rind. These samples were dried and milled into powder and were analyzed for proximate and mineral content using standard methods and atomic absorption spectrophotometer (AAS). The results for the proximate analysis revealed the presence of moisture content ranging from 3.50 % to 10.90 %, ash content (2.80 % to 6.50 %), fibre content (5.80% to 15.30%), crude fat (0.40 % to 13.10 %), crude protein (3.20 % to 19.20 %) and carbohydrate (46.10 % to 75.50 %). The result obtained from the mineral analysis revealed the presence of selected element, with Calcium having the highest mean value (25.69 mg/100g), followed by Magnesium (3.60 mg/100g), Iron (0.22 mg/100g) and Chromium (0.11 mg/100g), there were no traces of Lead and Cadmium. The data (result) of the study showed that the proximate and mineral parameter with the exception Pb and Cd were present in the bark, pulp and seeds although their concentration in the rind, seed and pulp vary significantly and also fall below the WHO recommended standard for minerals element.

Keywords: *watermelon, proximate, minerals, AAS.*

GJSFR-B Classification: *FOR Code: 030699*

PROXIMATEANDMINERALANALYSISOFWATERMELONSOLDATNORTHBANKMARKETMAKURDINIGERIA

Strictly as per the compliance and regulations of:

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Gav, B. L [α], Anyanwu, S. N., Oloruntoba, S. O [σ] & Tor. P [ρ]

Abstract- The study was carried out on proximate and mineral analysis of water melon sold at North bank market Makurdi. Water melon samples were purchased at North bank in January, 2018. The samples were sliced using a knife to separate the seeds, pulp and rind. These samples were dried and milled into powder and were analyzed for proximate and mineral content using standard methods and atomic absorption spectrophotometer (AAS). The results for the proximate analysis revealed the presence of moisture content ranging from 3.50 % to 10.90 %, ash content (2.80 % to 6.50 %), fibre content (5.80% to 15.30%), crude fat (0.40 % to 13.10 %), crude protein (3.20 % to 19.20 %) and carbohydrate (46.10 % to 75.50 %). The result obtained from the mineral analysis revealed the presence of selected element, with Calcium having the highest mean value (25.69 mg/100g), followed by Magnesium (3.60 mg/100g), Iron (0.22 mg/100g) and Chromium (0.11 mg/100g), there were no traces of Lead and Cadmium. The data (result) of the study showed that the proximate and mineral parameter with the exception Pb and Cd were present in the bark, pulp and seeds although their concentration in the rind, seed and pulp vary significantly and also fall below the WHO recommended standard for minerals element. This research generally reveals that these three parts of watermelon should be consumed while eaten it since they have high nutritional values.

Keywords: watermelon, proximate, minerals, AAS.

I. Introduction

Humans possess great capacity to adapt physiologically to different types of foods. In spite of this, nutrition science has demonstrated that there are certain foods that cannot be eliminated, such as fruits and fresh vegetables. Fruits offer the most rapid methods of providing adequate supplies of vitamins, minerals and fibres to people living in the tropics. Most fruits and vegetables have low energy density and are recommended for weight management (Rolls & Ello Martins *et al.,* 2004). The optimal diet for everyone as recommended by the world health and food/agricultural organization is a low-fat, and fibre diet rich in complex carbohydrate characterized by a frequent consumption of fruits and vegetables at least 400g daily as well as whole-grains, cereals and legumes at least 30g daily (WHO/FAO, 2003).Watermelon (*Citrullus lanatus)* is a scrambling and trailing vine in the flowering plant family *cucurbitaceae.* The species originated in Southern Africa, with evidences of its cultivation in Ancient Egypt. It is grown mostly in tropical and sub-tropical areas worldwide for its large edible fruit, which is a special kind of berry with a hard rind and no internal division, botanically called a *pepo.* Watermelon consists mostly of water (91 %) and carbs (7.5 %). It contains almost no protein (0.6 g) or fat (0.15 g), and is very low in calorie (30 kcal). The carbs are mostly simple sugars such as glucose, fructose and sucrose. Watermelon also contains small amount of fibers about 0.4 grams per 100 grams. However, it is considered high in fermentable short chain carbohydrate referred to as FODMAPs. FODMAPs cause unpleasant digestive symptoms in individuals who cannot digest them, such as those with irritable bowel syndrome.

Watermelon is the richest known dietary source of amino acid *Citrulline.* The highest amount is found in the white rind that surrounds the flesh (Tarazona– Diaz *et al.,* 2011). In the body, *Citrulline* is transformed into the essential amino acid Arginine. Both *Citrulline* and Arginine plays an important role in the synthesis of nitric oxide (NO), which helps to lower blood pressure by dilating and relaxing our blood vessels (Rimando *et al.,* 2005). Arginine is also important for many organs such as the lungs, kidneys, liver and the immune and reproductive systems. It has been shown to facilitate the healing of wounds (Ikeda *et al.,* 2000). Studies have shown that watermelon juice is able to increase blood levels of both *Citrulline* and Arginine considerably (Wu *et al.,* 2000). Despite being one of the best dietary sources of *Citrulline* one would have to consume about 5 pounds (2.3 kg) of watermelon to meet the recommended daily intake for Arginine.

Watermelon is a good source of vitamin C and also a decent source of several other vitamins and minerals. Vitamin C an antioxidant that is essential for skin health and immune function. Potassium (K) a mineral that is important for blood pressure control and heart health.VitaminB_5 also known as Pantothenic acid which to some extent is almost present in all food. The aim of this study is to carry out the proximate and mineral analysis of watermelon fruits sold in North Bank Market.

II. Materials and Methods

a) Study Area

According to Benue State Ministry for Lands and Survey, 2011, Makurdi lies between coordinate of 704350 North and 803210 East within central Nigeria

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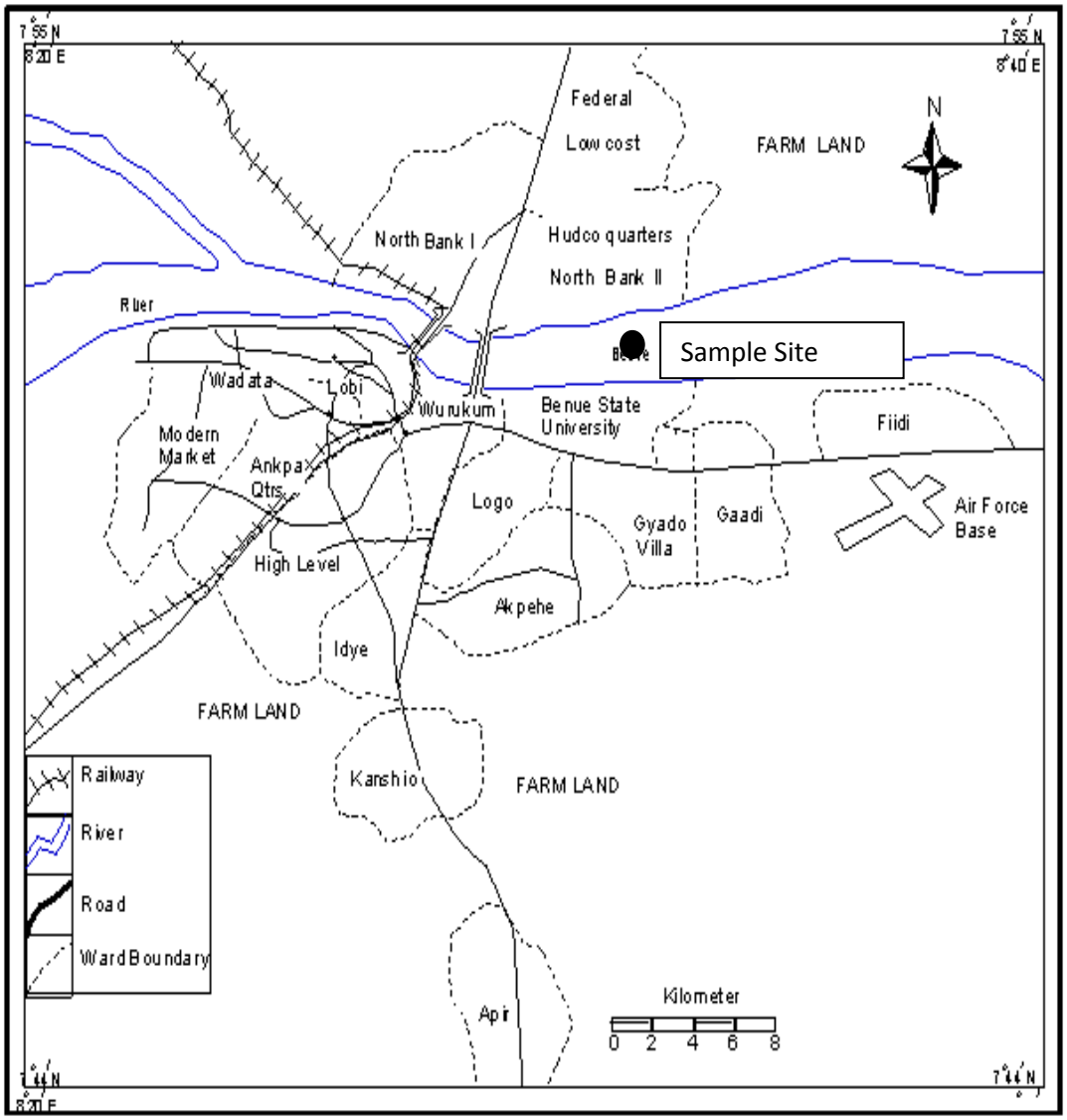
with an estimated population of five hundred thousand, seven hundred and ninety-seven persons (500,797) base on "The world Gazette" (2007). The network of rivers, gutters, standing pool of water, streams blocked gutters which enhanced disease emergence from time to time drains the area. Makurdi, the state capital was established in the early twenties and gained prominence in 1927 when it became the headquarters of the Benue province. Being a river port, it attracted the establishment of trading depots by companies such as United Africa Company of Nigeria and John Holt Plc. Its commercial status was further enhanced when the Railway Bridge was completed and opened in 1932. In 1976, the town became the capital of Benue State and today doubles as the headquarters of Makurdi Local Government Area. The town is divided by the River Benue into the North and South Banks, which are connected by two bridges. The sampling site is indicated in Figure 1.



(Source: Benue State Ministry for Lands and Survey, 2011).

Fig.1: Map of Makurdi Local Government Area showing the study area.

b) Collection of Samples

Three watermelon samples of average weight of 20 kg and diameter of 28 – 30 cm were purchased from North bank market in Makurdi metropolis, Benue State, Nigeria. All samples were collected in January, 2018.

c) Sample preparation/Digestion

The watermelon fruits were bought from North bank market, Makurdi, Benue State. The fruits were sliced/ chopped into small sizes using kitchen knife. The seeds were removed from the pulp before separating the red pulp from the rind. The seeds were washed, allowed to drain and placed on a foil. The pulps were chopped into shreds, allowed to drain and placed in another lined with foil. The rinds were chopped into tiny cubes and placed in a separate tray line with foil. The whole set up were afterward dried under Sun for 5 – 10 days ensure total dryness prior analysis.

5 g of each sample was weighed into 150 cm^3 conical flask and digested using 150cm^3 nitric acid, 2cm^3 perchloricacid and was placed on a hot plate for 3 hours. On cooling the digest was filtered into 100cm^3 volumetric flask and made up to the mark with distilled water. The digested sample were stored in a safe place prior to the AAS analysis.

d) Proximate Analysis of Sample

Proximate analysis of the sample for moisture content, ash content, crude lipid, crude fibre and crude protein were carried out in triplicate using the methods as described in AOAC(2000) while carbohydrate was determined by difference as shown below; %Carbohydrate = 100- (%moisture + %crude protein + %crude fat + %ether extract + %ash). All the proximate values were reported in g/100g sample. All chemicals used were of Analar grade.

e) Trace Metal Determination

2.0 cm^3 of the samples were each weighed and digested with concentrated HNO_3. After complete digestion, the volume was made up with deionized water in a volumetric flask. The samples were analyzed for trace metal using a computer controlled Atomic Absorption Spectrophotometer (AAS 696 Model).

f) Statistical Analysis

The results obtained were subjected to statistical evaluation. Data obtained from the parameters were evaluated using mean, standard deviation and coefficient of variation. All determinations were in triplicates.

III. Result

Table 1: Proximate composition of watermelon part

S/N	Parameter	Bark (%)	Pulp (%)	Seeds (%)	Mean($\bar{X}$)	SD	Coefficient of variation(%)
1	Moisture	8.702	10.903	3.500	7.7016	3.104	40.30
2	Ash content	6.501	4.202	2.801	4.5013	1.525	33.90
3	Crude fat	2.400	0.404	13.103	5.3023	5.576	105.20
4	Crude protein	4.202	3.200	19.201	8.868	7.318	82.50
5	Crude fibre	10.500	5.801	15.300	10.534	3.878	36.80
6	Carbohydrate	67.804	75.502	46.100	63.135	12.449	19.70

Table 2: Metal concentrations of watermelon part samples in mg/100g

S/N	Metal	Bark (%)	Pulp (%)	Seeds (%)	Mean($\bar{X}$)	SD	Coefficient of variation(%)	WHO Standard
1	Mg	4.36	3.61	2.83	3.60	0.62	17.30	50.00
2	Ca	29.25	25.14	22.67	25.69	2.71	10.60	75.00
3	Fe	0.28	0.22	0.16	0.22	0.05	23.70	0.30
4	Pb	ND	ND	ND	ND	ND	ND	0.01
5	Cd	ND	ND	ND	ND	ND	ND	0.003
6	Cr	0.17	0.10	0.05	0.11	0.05	45.10	0.05

ND = Not Detected.

IV. Discussion

a) Proximate analysis

The proximate composition of *Citrullus lanatus* bark, pulp and seeds are shown in Table 1. The result showed that the moisture content of the seed (3.50%) was significantly lower than that of the pulp, the bark (8.70%) was also significantly lower than the pulp (10.90%) but higher than the seeds. The moisture content of either the rind (bark) or the seed flour is lower than that of processed and unprocessed *Dioscorea dumentorium* (Egbuonu *et al*., 2014). In particular, the moisture content of the rind (8.70%) is higher than the value (5.08%) reported by Fila *et al.* (2013) where as that of the seed flour is lower than that reported by Ogunlade *et al.* (2011) for *Afzelia Africana* (9.49%) and *Pachira glabara* (9.13%). The lower moisture content of seed sample suggests higher dry matter yield (Bamigboye *et al., 2010*). The lower moisture could enhance storage stability (Ejikeme *et al.,* 2010*;* Bamigboye *et al., 2010;* Nzewi and Egbuonu, 2011) of the seed flour compared to that of the rind and pulp.

The result of the ash content indicates that *Citrullus lanatus* bark (6.50%) however was significantly higher than the seed (2.80%) and pulp (4.20%). The ash content of the rind agrees with that of Jack bean (6.51%) reported by Olalekan and Bosede (2010). The values were lower than the 7.45% of *cucurbita spp* reported by Aruah *et al.* (2011). The proportion of ash content is a reflection of the mineral content present in the food materials of the mineral contents presents in the food materials of the mineral contents presents in the food materials (Omotoso, 2006).

The result of the crude fat content of *Citrullus lanatus* in Table 1 showed that the seed (13.10%) was significantly higher than the pulp (0.40%) and the bark (2.40%). The mean value (5.30%) was higher compared with 1.6% for *Cucurbita spp* seeds reported by Aruah *et al.* (*2011*). However the values were lower than the 52.13% of *Cucurbita maxima* seed reported by Amoo *et al.* (2004). Dietary fats function in the increase of palatability of food by absorbing and retaining flavor (*Anita et al.,* 2006). Fats are also vital in the structural and biological functioning of cells and help in the transport of nutritionally essential fat soluble vitamins (Omotoso *et al.,* 2006)

The crude protein content result indicates that the bark (4.20%) is slightly higher than the pulp (3.20%) but significantly lower when compared with the seed (19.20%). The value of the seed was to be higher than the value (14.42 %) of *C. maxima* while that of the fruit pulp was of higher value to 0.2 – 2.7% reported for C. *maxima* by karaye *et al.*(2013). The level of protein in these indicates that they can contribute to the daily protein requirements for human which is based at 23-56g as stipulated by NRC.

The result of the crude fibre content revealed that the fibre content of the seed (15.30%) was higher than the pulp (5.80%) and the rind and seed flour is higher than the value (1.90%) reported by Fila *et al.* (2013). Fibre enhances the proper digestive function thereby preventing constipation and hemorrhoids (Erhirhie and Ekene, 2013).

The carbohydrate content of the Bark (67.80%) was significantly higher when compared with the seed (46.10%) but lower than the pulp (75.50%). These values were higher compared with the value of 6.39% reported for *Arachis hypogeal* by Loukou *et al.* (2007) and while the mean value was at an approximate range compared to 66.64% reported for *C. maxima* by Adebayo *et al.* (2013). The carbohydrate content obtained from these samples can be used to rank *Citrullus lanatus* as carbohydrate rich fruits due to the relatively high carbohydrate content of the fruit.

b) Metal analysis

The result for the mineral composition of the bark, pulp and seed are shown in Table 2. The most abundant mineral found in the samples is Calcium with a mean concentration of 25.69 mg/100g. The concentration was higher in the bark (29.25 mg/100g) compared to the pulp (25.14 mg/100g) and seed (22.67 mg/ 100g). These values were lower as compared to 294.74 ppm for *C. maxima* reported by Amoo *et al.* (2004). Calcium is a constituent of the bones and helps the body to contract correctly, blood to clot and nerves to convey messages. When Calcium supply to the body becomes insufficient the body on its own extracts the needed Calcium from the bones. If the body continues to tear down more Calcium than it replaces over a period of years, the bones will become weak and easily break.

Magnesium is the next abundant mineral element found in the samples with a mean value of 3.60 mg/100 g. The concentration was observed to be higher in the bark (4.36 mg/100 g). The least concentration occurs in the seed (2.83 mg/100 g) which is less than the value for the pulp (3.61 mg/100 g). Magnesium is beneficial to blood pressure and helps prevent sudden heart attack, cardiac arrest and stroke. Like Calcium, Magnesium is a very important component of bone and contribute to it structural development. While Calcium stimulates the muscles, Magnesium relaxes the muscles the daily value for Magnesium is 400 mg for Adult and Children aged 4 and older. Magnesium deficiency results in uncontrolled twisting of muscles leading to convulsion which may eventually lead to death and it is common in people with chronic alcoholism. The calcium and Magnesium content of the three parts fall within the WHO recommended range.

The concentration of Iron was found to have a mean value of (0.22 mg/100g). The concentrations for the respective parts are bark (0.28 mg/100 g), pulp (0.22

mg/100 g) and seeds (0.16 mg/100g). These values were lower than the 13.66 mg/100g reported by Mohammed (2004). Iron deficiency is a major problem in women diet in the developing world, particularly among pregnant women and especially in Africa (Bamigboye *et al., 2010).* This implies that these samples serve as blood building food and should be use for human and animal feeds formulations.

The mean value of Chromium in the samples is 0.11 mg/100g with the value of the bark, pulp and seeds as 0.17, 0.10 and 0.05 mg/ 100g respectively. Chromium is an essential nutrient that potentiates insulin action and thus influences carbohydrate, lipid and protein metabolism. Chromium has a biochemical function that affects the ability of the insulin receptor to interact with Insulin.

There was no trace of Lead and cadmium in the samples. This is highly important because Lead is classified as a category 2B carcinogen by the International Agency for Research on Cancer (IARC/WHO, 1993). The nervous system of infant and children are particularly sensitive to lead toxicity. Adult exposed occupationally or accidentally to excessive high levels exhibit peripheral neuropathology and chronic nephropathy (WHO, 2003). However, the critical or most sensitive effect for adults in the general population may be the development of hypertension. Individuals with severe Cadmium may have renal calculi and exhibit excessive urinary loss of Calcium, with chronic exposure calcium may eventually decline to become less than normal. These metals Fe, Pb, Cd and Cr fall within the World Health Organization (WHO) limit for mineral element in *Citrullus lantus.*

V. Conclusion

The results obtained from this study have shown that there is a significant difference in the nutritional and mineral contents of the pulp, seeds and rind of *Citrullus lanatus* sold at north bank market Makurdi, On comparing the nutritional content, the result revealed that the moisture contents ranges from 3.5 % - 10.90 %, ash content (2.80% - 6.50%), fibre content (5.80% - 15.30 %), crude fat (0.40 % - 13.10 %), crude protein (3.20 % - 19.20 %) and carbohydrate (46.10 % - 75.50 %). Variations were also observed in the mineral constituent of the pulp, seeds and rind (bark) of *Citrullus lanatus*, Mg (2.83 – 4.36 mg/100g), Ca (22.67 – 29.25 mg/100g), Fe (0.16 – 0.28 mg/100g) and Cr (0.05 – 0.17 mg/100g). There were no trace of Lead (Pb) and Cadmium (Cd) which are highly toxic to the body. The result for the metal analysis indicate that all the tested metal fall below the world health standard (WHO).

The analytical information available from the research revealed that the three (3) portion of *Citrullus lanatus*(pulp, seeds and bark are very essential and nutritive to the human body and should all be consumed when eaten it without discarding any part.

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Global Journal of Science Frontier Research: B
Chemistry
Volume 19 Issue 1 Version 1.0 Year 2019
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals
Online ISSN: 2249-4626 & Print ISSN: 0975-5896

Nutraceutical Potentials of *Spilanthes Filicualis*

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GJSFR-B Classification: *FOR Code: 060199*

Strictly as per the compliance and regulations of:

Nutraceutical Potentials of *Spilanthes Filicualis*

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I. Introduction

Africa is a continent with plants of economic and medicinal importance capable of meeting the nutrient and health needs of the populace (Josiah and Bartholomew, 2015). The current emphasis on healthy living based on antioxidant intake and the implication of oxidative stress molecules / free radicals on some diseased conditions has generated renewed interest in screening for plants with high antioxidant properties (Bouayed *et al.,* 2008). The identification and quantification of bioactive components that contribute to free radical scavenging activity are essential in the discovery of new drugs (Farombi, 2003). The basic functional units of plants are phytochemicals, which are the bioactive ingredients present in plants (Srinath and Laksmi, 2014). They include alkaloids, saponins, tannins, terpenoids, polyphenols, etc. These compounds are believed to be responsible for the medicinal properties attributed to plants, which is as result of their ability to inhibit the reactions of ROS, neutralizing free radicals by donating one of their electrons and blocking nitrosamine formation, stimulate the immune system and maintain cell membrane integrity (Sen *et al.*, 2010; Saha and Tamaraka, 2011). This study focused on exploring the nutritional compositions of leaves of *Spilanthes filicualis* and the *in-vitro* antioxidants properties of its ethanol leaf extract.*Spilanthes filicaulis* is a common plant grown in Brazil, Africa, and South America. It belongs to the family Asteraceae. The extract of the plant has been employed in the therapeutic cure of different ailments (Atawodi *et al.*, 2014). The flower heads of *Spilanthes filicaulis* are used to prevent scurvy and aid digestion. Experiments on rats suggest that cold water extract of *Spilanthes filicaulis* acts as a loop diuretic (Ratnasooriya *et al.*, 2004). Jahan *et al.* (2013) demonstrated that the ethanol extract of *Spilanthes filicaulis* showed antimicrobial activity against *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus pyogenes, Enterococcus faecalis*, *Escherichia coli, Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Proteus mirabilis*, *Salmonella typhi,* and *Shigella dysenteriae. S. filicaulis* contains Spilanthol which shows activity against Plasmodium falciparum (Gasquet *et al.*, 1993). Aqueous leaf extracts of *S. filicaulis* have been reported as potent antidote for poison (Atawodi *et al.*, 2014)

Plate. 1: Spilanthes filicaulis leaves

Local names of this plant in Nigeria are: Hausa = parpehi, Igbo = osana or ósē ànì, Bayelsa (Kolokuma) = kírí ẹ̀bẹ̀dẹ̀, Common name = Toothache plant.

II. Materials and Methods

a) Materials

i. *Collection of plant material and identification*

Spilanthes filicualis leaves were harvested from a swampy field site in Emonu– Orogun, Delta State, Nigeria in April, 2017. The Department of Botany, Delta

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State University, Abraka– Nigeria identified and authenticated the plant under study.

b) Methods

i. *Preparation of plant extract*

Fresh leaves of *Spilanthes filicualis* were washed with distilled water, and air dried for two weeks and then reduced to coarse powder using a manual grinder. Then, 100g of coarsely powdered leaves was extracted with 400ml of 80% ethanol using cold maceration for 24hours. The extract was filtered through cheesecloth with a fine pore, and the filtrate filtered for the second time using Whatman No. 1 filter paper. The resulting extract concentrated at 50°C in a rotary evaporator for 2hr, and then transferred to a water bath maintained at 40°C and evaporated to dryness to yield a dark green mass. The obtained extract was put in a glass container and stored at 4°C until when required for use.

ii. *Proximate analysis*

Moisture, ash, crude fiber, crude fat, crude protein, and total carbohydrate contents were determined using standard analytical procedure (AOAC, 1990).

iii. *Determination of energy content of the leaves*

The energy content was calculated by multiplying the mean values of crude protein, crude fat and total carbohydrate by the Atwater factors of four (4), nine (9), four (4) respectively, taking the sum of the products and expressing the result in Kcal per 100g sample as reported by Onyeike and Acheru (2002).

iv. *Determination of antioxidant vitamins*

Vitamin A and C were estimated using Kirk and Sawyer (1991), while Vitamin E was determined by Futter – Mayer colorimetric method.

v. *Determination of minerals*

The Varian AA240 Atomic Absorption Spectrophotometer was used in the mineral analysis by the method of American Public Health Association (1995).

III. Phytochemical Analysis

Preliminary phytochemical screening of ethanol leaf extract of *Spilanthes filicaulis* was carried out using standard methods as described by Borokini and Omotayo (2012), and Njoku and Obi (2009) to screen for the presence of various chemical constituents, while the quantitative phytochemical estimation was done using the Varian Gas Chromatograph (HRGC, DB-5MS, England).

a) Determination of antioxidant activity and free radical scavenging potentials

i. *DPPH radical scavenging assay*

The determination of free radical scavenging activity was by the1,1- diphenyl- 2- picrylhydrazyl (DPPH) radical assay (Manzocco *et al.*, 1998). A0. 2 ml of different concentrations of ethanol leaf extract of *Spilanthes filicualis* (2. 0- 10 mg/ml) was added to 2 ml of DPPH solution (0. 3 mM). After 30 min of incubation in the dark, the absorbance was measured at 517 nm. The percentage inhibition of the DPPH radical scavenging was calculated using the equation below:

$$\% \text{ inhibition of DPPH radical} = ([A_0 - A_1]/ A_0) \times 100$$

Where A_0 is the absorbance of the control (blank, without extract) and A_1 is the absorbance in the presence of the extract.

ii. *Nitric oxide (NO) free radical scavenging activity*

The method of Marcocci *et al.,* 1994 was used for Nitric oxide assay. Two millilitres of 10 mM sodium nitroprusside dissolved in 0. 5 ml of 10mM phosphate buffer saline (pH 7. 4) was mixed with 0. 5 ml of different concentrations of ethanol leaf extract of *Spilanthes filicualis* (2.0- 10mg/ml). The mixture was then incubated at 25°C. After 150 min of incubation, 0.5 ml of the incubated solution was withdrawn and mixed with 0.5 ml of Griess reagent [(1.0 ml sulfanilic acid reagent (0. 33% in 20% glacial acetic acid at room temperature for 5 min with 1 ml of naphthylethylenediamine dichloride (0. 1% w/v)]. The mixture was then incubated at room temperature for 30 min, and its absorbance was measured at 546 nm against blank. The percentage inhibition of the nitric oxide radical scavenging was calculated using the equation below:

$$\% \text{ inhibition of NO radical} = ([A_0 - A_1]/ A_0) \times 100$$

Where A_0 is the absorbance of the control (blank, without extract) and A_1 is the absorbance in the presence of the extract.

iii. *Reducing power assay (RP)*

Reducing power was assayed using the method of Oyaizu, (1986). A 2.5ml of 0.2 M phosphate buffer (pH 6.6) and 2.5 ml of $K_3Fe(CN)_6$ (1% w/v) were added to 1.0 ml of different concentrations of ethanol leaf extract of *Spilanthes filicualis* (2.0 - 10 mg/ml). The resulting mixture is incubated at 50°C for 20 min, followed by the addition of 2.5 ml of Trichloroacetic acid (10% w/v). The mixture was centrifuged at 3000 rpm for 10 min to collect the upper layer of the solution (2.5 ml), mixed with distilled water (2.5 ml) and 0.5 mL of $FeCl_3$ (0.1%, w/v).The absorbance was then measured at 700 nm against blank sample (that contained distilled water and sodium phosphate buffer).

iv. *Metal chelation assay*

A 150µl of freshly prepared 2 mM $FeSO_4.7H_2O$ was added to a reaction mixture containing 168 µl of 0.1 M Tris-HCl (pH 7.4), 218 µl saline and100 µl of different concentrations of ethanol leaf extract of *Spilanthes filicualis* (2.0 - 10 mg/ml). The reaction mixture was incubated at 37°C for 5 min, before the addition of 13 µl of 0.25% 1,10 -Phenanthroline (w/v). The absorbance

was subsequently measured at 510spectrophotometer against the blank (Puntel *et al.*, 2005).

The percentage inhibition of the Metal chelating radical scavenging was calculated using the equation below: % inhibition of Metal chelating radical = ($[A_0 - A_1]/ A_0$) x 100

Where A_0 is the absorbance of the control (blank, without extract) and A_1 is the absorbance in the presence of the extract.

v. *Inhibition of lipid peroxidation in egg homogenate*.

Ten percent egg homogenate of 0.5 ml volume and different concentrations of ethanol leaf extract of *Spilanthes filicualis* 0.1 ml (2.0 - 10 mg/ml) were mixed in a test tube, and their final volume was made to 1.0 ml by addition of distilled water. Finally 0.05 ml 0.07M FeSO4 was added to the above mixture and incubated at 37°C for 30 min to induce lipid peroxidation. After that, 1.5 ml of 20 % acetic acid and 1.5 ml of 0.8 % TBA (prepared in 1.1% sodium dodecyl sulphate) and 0.05 ml 20 % TCA was added, vortexed and heated in boiling water bath for 60 min. After cooling, 5.0 ml normal butanol was added to each tube and centrifuged at 3000 rpm for 10 min. The absorbance of the organic upper layer was measured at 532 nm (Roberto *et al.*, 2000).

% inhibition of Lipid peroxidation = ([1– E]/C) x 100

Where C = absorbance of fully oxidized control and E = absorbance in the presence of extract.

vi. *Statistical analysis*

All data were subjected to statistical analysis. Values were reported as Mean ± Standard deviation while one way ANOVA was used to test for differences. The results were considered significant at p-values of less than 0.05 ($p<0.05$).

IV. Results

The results of the proximate composition of the leaves of *Spilanthes filicualis* are in Table 1. Total carbohydrate (64.9%), crude protein (6.30%), crude fat (2.00%), Fibre (4.50%), Moisture (17.8%) and Ash (4.55%). The energy content of the leaves of *Spilanthes filicualis* was 303Kcal/100g sample.

Table 1: Proximate composition of *Spilanthes filicualis* leaves

Parameters	Composition (%)
Total carbohydrate	64.9 ± 0.89
Crude protein	6.30 ± 0.30
Crude fat	2.00 ± 0.10
Fibre	4.50 ± 0.26
Moisture	17.8 ± 0.14
Ash	4.55 ± 0.07
Energy content (Kcal/100g sample)	303

Values are means ± standard deviations of triplicate determinations.

Results from the evaluation of the antioxidant vitamin composition are in Table 2. Antioxidant vitamins of A, C and E, were determined with the highest value of these vitamins being vitamin A (239mg/100g).

Table 2: Antioxidant vitamins composition of *Spilanthes filicualis* leaves.

Parameters	Composition (mg/100g)
Vitamin A	239 ± 2.90
Vitamin C	7.03 ± 0.17
Vitamin E	0.23 ± 0.02

Values are means ± standard deviations of triplicate determinations.

Mineral composition of *Spilanthes filicualis* leaves is presented in Table 3. Calcium (20.9 mg/kg) recorded the highest concentration, followed by Magnesium (19.8 mg/kg) and the lowest was Cobalt (0.13 mg/kg).

Table 3: Mineral concentration of *Spilanthes filicualis* leaves

Mineral	Concentration (mg/kg)
Chromium	0.81 ± 0.10
Magnesium	19.8 ± 0.71
Cobalt	0.13 ± 0.02
Iron	17.5 ± 0.37
Copper	0.87 ± 0.12
Manganese	2.06 ± 0.03
Zinc	18.5 ± 0.33
Selenium	0.59 ± 0.09
Calcium	20.9 ± 0.06
Sodium	14.2 ± 0.19
Potassium	8.33 ± 0.19

Values are means ± standard deviations of triplicate determinations.

Phytochemical screening of ethanol leaf extract of *Spilanthes filicaulis* is in Table 4. Bioactive components of saponins, tannins, flavonoids, Alkaloid, and phenol were found to be present.

The results of the quantitative phytochemical analysis of leaves of *Spilanthes filicualis* are presented in Table 5. Bioactive components of the flavonoids family were obtained. Highest concentrations were rutin (27.6 μg/ml) followed by kaempferol (26.0 μg/ml), and the lowest was anthocyanin (0.69 μg/ml).

Table 4: Qualitative phytochemical screening of ethanol leaf extract of *Spilanthes filicualis*

Phytochemi	Concentrations
Saponin	+
Tannin	+
Terpene	–
Flavonoid	+
Phlobatannin	–
Alkaloid	+
Glycoside	–

Key: + Present, - Absent

Table 5: Quantitative phytochemical analysis of Spilanthes filicualis leaves

Phytochemicals	Concentration (µg/ml)
Anthocyanin	0.69
Tannin	7.77
Rutin	27.6
Phenol	7.08
Epicatechin	3.59
Lunamarine	8.77
Saponin	21.3
Sapogenin	20.0
Phytate	0.91
Kaempferol	26.0
Catechin	20.2

Results of the i*n-vitro* antioxidant and free radical scavenging activities of ethanol leaf extract of *Spilanthes filicualis* are embodied in Table 6. It revealed that plant activity against DPPH radical, reducing power and lipid peroxidation (LPO) was significantly increased with increasing concentrations ($p<0.05$), while metal chelating activity recorded a statistical decrease with increasing dose (24.36 $\pm$ 1.41 to -32.31 $\pm$ 3.60 % inhibition) ($p<0.05$). Nitric oxide scavenging activity recorded a statistical increase from 2mg/ml to 6mg/ml (1.87 $\pm$ 0.20 to12.68 $\pm$ 2.1 % inhibition) and conversely statistical decrease in from 8mg/ml to 10mg/ml (-6.64 $\pm$ 0.62 to -29.80 $\pm$ 2.15 % inhibition) ($p<0.05$).

Table 6: In-vitro antioxidant activity of ethanol leaf extract of *Spilanthes filicualis*

Conc.(mg/ml	% Inhibition				700nm
	DPPH	NO	MC	LPO	RP
2	31.03 $\pm$ 1.44^{a}	1.87 $\pm$ 0.20^{a}	24.36 $\pm$ 1.41^{a}	42.32 $\pm$ 2.73^{a}	0.37 $\pm$ 0.04^{a}
4	46.15 $\pm$ 1.87^{b}	12.26 $\pm$ 0.73^{b}	17.84 $\pm$ 1.47^{b}	61.20 $\pm$ 1.59^{b}	0.54 $\pm$ 0.16^{b}
6	52.87 $\pm$ 1.67^{c}	12.68 $\pm$ 2.17^{b}	7.26 $\pm$ 0.30^{c}	63.22 $\pm$ 3.27^{b}	0.61 $\pm$ 0.05^{b}
8	57.54 $\pm$ 1.23^{d}	-6.64 $\pm$ 0.62^{c}	2.76 $\pm$ 0.19^{d}	68.67 $\pm$ 0.74^{c}	0.67 $\pm$ 0.03^{c}
10	61.81 $\pm$ 1.86^{e}	-29.80 $\pm$ 2.15^{d}	-32.31 $\pm$ 3.60^{e}	85.35 $\pm$ 1.44^{d}	0.74 $\pm$ 0.02^{c}

Values are means $\pm$ standard deviations of triplicate determinations. Values not sharing common superscript on the same column differ significantly ($p < 0.05$).

DPPH =1, 1-diphenyl -2-picryl hydrazyl, NO = Nitric oxide, MC = Melating Chelating, LPO = Lipid peroxidation, RP = Reducing power

V. Discussion

Proximate analysis of *Spilanthes filicualis* leaves reveals that the plant is rich in carbohydrate (64.9%). Carbohydrate is a source of fuel in living cells which is required for the production of energy and maintenance of general body function (Adesuyi *et al.*, 2012). Proteins play an essential role in information transmission in the body, serving as a neurotransmitter and genetic transmission of traits, tissue repair and general growth (Voet *et al.,* 2008). The presence of moisture content in food aids digestion. High values of moisture content result in short shelf life of food (Shukla *et al.*, 2015). Dietary fiber is essential in aiding digestion, removal of cholesterol, detoxification of carcinogens etc. (Dhingra *et al.,* 2012), while the level of ash content is an indication of the mineral concentration of the plant leaf. Thus with a crude protein content of 6.30%, a dietary fiber of 4.50%, an ash content of 4.55% and crude fat content of 2.00%, implies that if the leaves of *Spilanthes filicualis* supplemented with another nutrient-rich plant, it could be of great nutritional benefits.

Quantitative analysis of vitamin A, C and E are indicative of an enhanced free radical scavenging

capacity of the plant. Vitamin A composition of 239 mg/100 was the highest value recorded for selected antioxidant vitamins determined in this study. Vitamin A is not only responsible for neutralizing the effect of singlet oxygen but also contributes to the immunostimulatory properties of *S. filicaulis* and for better vision (Pham-Huy *et al*., 2008). The concentration of vitamin C is higher than that of vitamin E. Vitamin C potentially regenerates vitamin E and renews its potency. The presence of vitamin E in the leaves of this plant suggests its antioxidant activity which is responsible for stabilization of biomembrane structure. The vitamin constituents of *S.filicaulis* may establish, in part, the efficient regulation of reactive oxygen species and scavenging activity observed in the leaves extract investigated in addition to maintaining membrane fluidity and integrity (Niki *et al.,* 1995).

Minerals are essential component necessary for optimal functions of the body as they play an immense role in energy production, defense against disease, bone formation, blood coagulation, hormonal regulation, transportation of fluid, muscle contraction and nerve transmissions (Delvin, 2006). Zinc, copper, and manganese are cofactors of superoxide dismutase (SOD), and iron is a component of hemoglobin and also serves as a cofactor for catalase, Selenium is a component of the prosthetic group of glutathione peroxidase (GPx). Calcium plays an essential role in bone formation and regulation of vitamin D, while Potassium and sodium serve as the main cation of the internal and external cellular fluid respectively, and also aid intracellular membrane transport system (Na^+ - K^+ ATPase transporter). Cobalt is vital for DNA synthesis, cell division, and cell growth. Chromium is believed to have an implicating role in upholding the configuration of the RNA molecule (Delvin, 2006; Soetan *et al.,* 2010). Thus, *Spilanthe filicualis* leaves could be a useful supplementary source of mineral nutrients to humans.

Phytochemicals are the important component of plants, and they play a vital role that is beneficial to human health (Batta, 2016). Qualitative phytochemical screening of ethanol leaf extract of *Spilanthes filicualis* showed the presence of saponins, tannins, flavonoids, alkaloids, and phenol. Ndam *et al*. (2014) reported the presence of tannins and steroids, while Ilondu *et al*. (2014) reported the presence of the enlisted phytochemicals found in this study and also detected the presences of terpenes in leaf extracts of *Spilanthes filicualis*. This variation could be as a result of soil and climatic conditions. Quantitative phytochemical analysis showed that plant contained flavonoids class of phytochemicals of anthocyanins, catechins epicatechin and kaempferol; phenol, glycosides class of saponins and rutin, alkaloid, and steroid glycoside compound sapogenin. Phytate was found to be 0.91 $\pm$ 0.02 µg/ml (0.91ppm). Phytate is well known for decreasing the bioavailability of minerals such as zinc, iron, calcium, magnesium, manganese, and copper (Kumar *et al.,* 2010). Wreesman (2014) reported that the levels of phytate more than 1000ppm affects mineral absorption. Saponin has anti-inflammatory effects, hemolytic activity, and cholesterol binding properties. Flavonoids are known to have antimicrobial, anti-inflammatory and antioxidant properties (Tijjani *et al.,* 2013; Myha *et al.,* 2014; Batta, 2016). Phenols play an active role in free radical scavenging by acting as a quencher to free radicals reactions (Tijjani *et al.,* 2013).

The *in-vitro* antioxidant study revealed that ethanol leaf extract of *Spilanthes filicualis* possesses antioxidant activity in a dose-dependent manner against DPPH, reducing power and lipid peroxidation activity ($p<0.05$). The extract showed a high level of percentage inhibition against lipid peroxidation than DPPH. Antioxidant activities of plants relates to the presence of phytochemicals. DPPH assay is based on scavenging activities of free radicals which serve as good criteria for measuring scavenging activities of the plants, the levels of total phenols and flavonoids are determinant to DPPH scavenging activity of plants (Boni *et al.,* 2014). Antioxidants activity against reducing power are attributed to the presences of reductones. Reductones mechanism of action is by the disintegration of free radical chains (Boni *et al.,* 2014).

VI. Conclusion

The study showed that *Spilanthes filicualis* leaves are rich in carbohydrate as shown in the result of the proximate composition, the presences of bioactive component of phenol, flavonoids, alkaloids, tannins, and saponins were detected from the qualitative phytochemical screening. The quantitative phytochemical analysis showed that rutin had the highest concentration, while the lowest was anthocyanin. Calcium concentration recorded the highest value and cobalt was the lowest from the mineral composition determination, whiles the highest vitamin, was recorded for vitamin A. The *in-vitro* antioxidant activities of ethanol leaf extract of *Spilanthes filicualis* showed scavenging activities against DPPH, reducing power and lipid peroxidation. The leaves of S. filicaulis possesses both good nutrient and medicinal properties.

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FELLOWS

FELLOW OF ASSOCIATION OF RESEARCH SOCIETY IN SCIENCE (FARSS)

Global Journals Incorporate (USA) is accredited by Open Association of Research Society (OARS), U.S.A and in turn, awards "FARSS" title to individuals. The 'FARSS' title is accorded to a selected professional after the approval of the Editor-in-Chief/Editorial Board Members/Dean.

- The "FARSS" is a dignified title which is accorded to a person's name viz. Dr. John E. Hall, Ph.D., FARSS or William Walldroff, M.S., FARSS.

FARSS accrediting is an honor. It authenticates your research activities. After recognition as FARSB, you can add 'FARSS' title with your name as you use this recognition as additional suffix to your status. This will definitely enhance and add more value and repute to your name. You may use it on your professional Counseling Materials such as CV, Resume, and Visiting Card etc.

The following benefits can be availed by you only for next three years from the date of certification:

FARSS designated members are entitled to avail a 40% discount while publishing their research papers (of a single author) with Global Journals Incorporation (USA), if the same is accepted by Editorial Board/Peer Reviewers. If you are a main author or co-author in case of multiple authors, you will be entitled to avail discount of 10%.

Once FARSB title is accorded, the Fellow is authorized to organize a symposium/seminar/conference on behalf of Global Journal Incorporation (USA). The Fellow can also participate in conference/seminar/symposium organized by another institution as representative of Global Journal. In both the cases, it is mandatory for him to discuss with us and obtain our consent.

You may join as member of the Editorial Board of Global Journals Incorporation (USA) after successful completion of three years as Fellow and as Peer Reviewer. In addition, it is also desirable that you should organize seminar/symposium/conference at least once.

We shall provide you intimation regarding launching of e-version of journal of your stream time to time.This may be utilized in your library for the enrichment of knowledge of your students as well as it can also be helpful for the concerned faculty members.

The FARSS can go through standards of OARS. You can also play vital role if you have any suggestions so that proper amendment can take place to improve the same for the benefit of entire research community.

As FARSS, you will be given a renowned, secure and free professional email address with 100 GB of space e.g. johnhall@globaljournals.org. This will include Webmail, Spam Assassin, Email Forwarders,Auto-Responders, Email Delivery Route tracing, etc.

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The FARSS member can apply for grading and certification of standards of their educational and Institutional Degrees to Open Association of Research, Society U.S.A.
Once you are designated as FARSS, you may send us a scanned copy of all of your credentials. OARS will verify, grade and certify them. This will be based on your academic records, quality of research papers published by you, and some more criteria. After certification of all your credentials by OARS, they will be published on your Fellow Profile link on website https://associationofresearch.org which will be helpful to upgrade the dignity.

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The FARSS is eligible to earn from sales proceeds of his/her researches/reference/review Books or literature, while publishing with Global Journals. The FARSS can decide whether he/she would like to publish his/her research in a closed manner. In this case, whenever readers purchase that individual research paper for reading, maximum 60% of its profit earned as royalty by Global Journals, will be credited to his/her bank account. The entire entitled amount will be credited to his/her bank account exceeding limit of minimum fixed balance. There is no minimum time limit for collection. The FARSS member can decide its price and we can help in making the right decision.

The FARSS member is eligible to join as a paid peer reviewer at Global Journals Incorporation (USA) and can get remuneration of 15% of author fees, taken from the author of a respective paper. After reviewing 5 or more papers you can request to transfer the amount to your bank account.

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The "MARSS" is a dignified ornament which is accorded to a person's name viz. Dr. John E. Hall, Ph.D., MARSS or William Walldroff, M.S., MARSS.

MARSS accrediting is an honor. It authenticates your research activities. After becoming MARSS, you can add 'MARSS' title with your name as you use this recognition as additional suffix to your status. This will definitely enhance and add more value and repute to your name. You may use it on your professional Counseling Materials such as CV, Resume, Visiting Card and Name Plate etc.

The following benefitscan be availed by you only for next three years from the date of certification.

MARSS designated members are entitled to avail a 25% discount while publishing their research papers (of a single author) in Global Journals Inc., if the same is accepted by our Editorial Board and Peer Reviewers. If you are a main author or co-author of a group of authors, you will get discount of 10%.

As MARSS, you will be given a renowned, secure and free professional email address with 30 GB of space e.g. johnhall@globaljournals.org. This will include Webmail, Spam Assassin, Email Forwarders,Auto-Responders, Email Delivery Route tracing, etc.

We shall provide you intimation regarding launching of e-version of journal of your stream time to time.This may be utilized in your library for the enrichment of knowledge of your students as well as it can also be helpful for the concerned faculty members.

The MARSS member can apply for approval, grading and certification of standards of their educational and Institutional Degrees to Open Association of Research, Society U.S.A.

Once you are designated as MARSS, you may send us a scanned copy of all of your credentials. OARS will verify, grade and certify them. This will be based on your academic records, quality of research papers published by you, and some more criteria.

It is mandatory to read all terms and conditions carefully.

AUXILIARY MEMBERSHIPS

Institutional Fellow of Global Journals Incorporation (USA)-OARS (USA)

Global Journals Incorporation (USA) is accredited by Open Association of Research Society, U.S.A (OARS) and in turn, affiliates research institutions as "Institutional Fellow of Open Association of Research Society" (IFOARS).

The "FARSC" is a dignified title which is accorded to a person's name viz. Dr. John E. Hall, Ph.D., FARSC or William Walldroff, M.S., FARSC.

The IFOARS institution is entitled to form a Board comprised of one Chairperson and three to five board members preferably from different streams. The Board will be recognized as "Institutional Board of Open Association of Research Society"-(IBOARS).

The Institute will be entitled to following benefits:

The IBOARS can initially review research papers of their institute and recommend them to publish with respective journal of Global Journals. It can also review the papers of other institutions after obtaining our consent. The second review will be done by peer reviewer of Global Journals Incorporation (USA) The Board is at liberty to appoint a peer reviewer with the approval of chairperson after consulting us.

The author fees of such paper may be waived off up to 40%.

The Global Journals Incorporation (USA) at its discretion can also refer double blind peer reviewed paper at their end to the board for the verification and to get recommendation for final stage of acceptance of publication.

The IBOARS can organize symposium/seminar/conference in their country on behalf of Global Journals Incorporation (USA)-OARS (USA). The terms and conditions can be discussed separately.

The Board can also play vital role by exploring and giving valuable suggestions regarding the Standards of "Open Association of Research Society, U.S.A (OARS)" so that proper amendment can take place for the benefit of entire research community. We shall provide details of particular standard only on receipt of request from the Board.

The board members can also join us as Individual Fellow with 40% discount on total fees applicable to Individual Fellow. They will be entitled to avail all the benefits as declared. Please visit Individual Fellow-sub menu of GlobalJournals.org to have more relevant details.

We shall provide you intimation regarding launching of e-version of journal of your stream time to time. This may be utilized in your library for the enrichment of knowledge of your students as well as it can also be helpful for the concerned faculty members.

After nomination of your institution as "Institutional Fellow" and constantly functioning successfully for one year, we can consider giving recognition to your institute to function as Regional/Zonal office on our behalf.
The board can also take up the additional allied activities for betterment after our consultation.

The following entitlements are applicable to individual Fellows:

Open Association of Research Society, U.S.A (OARS) By-laws states that an individual Fellow may use the designations as applicable, or the corresponding initials. The Credentials of individual Fellow and Associate designations signify that the individual has gained knowledge of the fundamental concepts. One is magnanimous and proficient in an expertise course covering the professional code of conduct, and follows recognized standards of practice.

Open Association of Research Society (US)/ Global Journals Incorporation (USA), as described in Corporate Statements, are educational, research publishing and professional membership organizations. Achieving our individual Fellow or Associate status is based mainly on meeting stated educational research requirements.

Disbursement of 40% Royalty earned through Global Journals : Researcher = 50%, Peer Reviewer = 37.50%, Institution = 12.50% E.g. Out of 40%, the 20% benefit should be passed on to researcher, 15 % benefit towards remuneration should be given to a reviewer and remaining 5% is to be retained by the institution.

We shall provide print version of 12 issues of any three journals [as per your requirement] out of our 38 journals worth $ 2376 USD.

Other:

The individual Fellow and Associate designations accredited by Open Association of Research Society (US) credentials signify guarantees following achievements:

- The professional accredited with Fellow honor, is entitled to various benefits viz. name, fame, honor, regular flow of income, secured bright future, social status etc.

- In addition to above, if one is single author, then entitled to 40% discount on publishing research paper and can get 10%discount if one is co-author or main author among group of authors.
- The Fellow can organize symposium/seminar/conference on behalf of Global Journals Incorporation (USA) and he/she can also attend the same organized by other institutes on behalf of Global Journals.
- The Fellow can become member of Editorial Board Member after completing 3yrs.
- The Fellow can earn 60% of sales proceeds from the sale of reference/review books/literature/publishing of research paper.
- Fellow can also join as paid peer reviewer and earn 15% remuneration of author charges and can also get an opportunity to join as member of the Editorial Board of Global Journals Incorporation (USA)
- • This individual has learned the basic methods of applying those concepts and techniques to common challenging situations. This individual has further demonstrated an in–depth understanding of the application of suitable techniques to a particular area of research practice.

Note :

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- In future, if the board feels the necessity to change any board member, the same can be done with the consent of the chairperson along with anyone board member without our approval.
- In case, the chairperson needs to be replaced then consent of 2/3rd board members are required and they are also required to jointly pass the resolution copy of which should be sent to us. In such case, it will be compulsory to obtain our approval before replacement.
- In case of "Difference of Opinion [if any]" among the Board members, our decision will be final and binding to everyone.

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Preferred Author Guidelines

We accept the manuscript submissions in any standard (generic) format.

We typeset manuscripts using advanced typesetting tools like Adobe In Design, CorelDraw, TeXnicCenter, and TeXStudio. We usually recommend authors submit their research using any standard format they are comfortable with, and let Global Journals do the rest.

Alternatively, you can download our basic template from https://globaljournals.org/Template.zip

Authors should submit their complete paper/article, including text illustrations, graphics, conclusions, artwork, and tables. Authors who are not able to submit manuscript using the form above can email the manuscript department at submit@globaljournals.org or get in touch with chiefeditor@globaljournals.org if they wish to send the abstract before submission.

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Authors must ensure the information provided during the submission of a paper is authentic. Please go through the following checklist before submitting:

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2. Authors must accept the privacy policy, terms, and conditions of Global Journals.
3. Ensure corresponding author's email address and postal address are accurate and reachable.
4. Manuscript to be submitted must include keywords, an abstract, a paper title, co-author(s') names and details (email address, name, phone number, and institution), figures and illustrations in vector format including appropriate captions, tables, including titles and footnotes, a conclusion, results, acknowledgments and references.
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7. Manuscript submitted *must not have been submitted or published elsewhere* and all authors must be aware of the submission.

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2. Drafting the paper and revising it critically regarding important academic content.
3. Final approval of the version of the paper to be published.

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The corresponding author should mention the name and complete details of all co-authors during submission and in manuscript. We support addition, rearrangement, manipulation, and deletions in authors list till the early view publication of the journal. We expect that corresponding author will notify all co-authors of submission. We follow COPE guidelines for changes in authorship.

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Unless specified in the notification, the Editorial Board's decision on publication of the paper is final and cannot be appealed before making the major change in the manuscript.

Acknowledgments

Contributors to the research other than authors credited should be mentioned in Acknowledgments. The source of funding for the research can be included. Suppliers of resources may be mentioned along with their addresses.

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Authors can submit papers and articles in an acceptable file format: MS Word (doc, docx), LaTeX (.tex, .zip or .rar including all of your files), Adobe PDF (.pdf), rich text format (.rtf), simple text document (.txt), Open Document Text (.odt), and Apple Pages (.pages). Our professional layout editors will format the entire paper according to our official guidelines. This is one of the highlights of publishing with Global Journals—authors should not be concerned about the formatting of their paper. Global Journals accepts articles and manuscripts in every major language, be it Spanish, Chinese, Japanese, Portuguese, Russian, French, German, Dutch, Italian, Greek, or any other national language, but the title, subtitle, and abstract should be in English. This will facilitate indexing and the pre-peer review process.

The following is the official style and template developed for publication of a research paper. Authors are not required to follow this style during the submission of the paper. It is just for reference purposes.

Manuscript Style Instruction (Optional)

- Microsoft Word Document Setting Instructions.
- Font type of all text should be Swis721 Lt BT.
- Page size: 8.27" x 11'", left margin: 0.65, right margin: 0.65, bottom margin: 0.75.
- Paper title should be in one column of font size 24.
- Author name in font size of 11 in one column.
- Abstract: font size 9 with the word "Abstract" in bold italics.
- Main text: font size 10 with two justified columns.
- Two columns with equal column width of 3.38 and spacing of 0.2.
- First character must be three lines drop-capped.
- The paragraph before spacing of 1 pt and after of 0 pt.
- Line spacing of 1 pt.
- Large images must be in one column.
- The names of first main headings (Heading 1) must be in Roman font, capital letters, and font size of 10.
- The names of second main headings (Heading 2) must not include numbers and must be in italics with a font size of 10.

Structure and Format of Manuscript

The recommended size of an original research paper is under 15,000 words and review papers under 7,000 words. Research articles should be less than 10,000 words. Research papers are usually longer than review papers. Review papers are reports of significant research (typically less than 7,000 words, including tables, figures, and references)

A research paper must include:

a) A title which should be relevant to the theme of the paper.
b) A summary, known as an abstract (less than 150 words), containing the major results and conclusions.
c) Up to 10 keywords that precisely identify the paper's subject, purpose, and focus.
d) An introduction, giving fundamental background objectives.
e) Resources and techniques with sufficient complete experimental details (wherever possible by reference) to permit repetition, sources of information must be given, and numerical methods must be specified by reference.
f) Results which should be presented concisely by well-designed tables and figures.
g) Suitable statistical data should also be given.
h) All data must have been gathered with attention to numerical detail in the planning stage.

Design has been recognized to be essential to experiments for a considerable time, and the editor has decided that any paper that appears not to have adequate numerical treatments of the data will be returned unrefereed.

i) Discussion should cover implications and consequences and not just recapitulate the results; conclusions should also be summarized.
j) There should be brief acknowledgments.
k) There ought to be references in the conventional format. Global Journals recommends APA format.

Authors should carefully consider the preparation of papers to ensure that they communicate effectively. Papers are much more likely to be accepted if they are carefully designed and laid out, contain few or no errors, are summarizing, and follow instructions. They will also be published with much fewer delays than those that require much technical and editorial correction.

The Editorial Board reserves the right to make literary corrections and suggestions to improve brevity.

Format Structure

It is necessary that authors take care in submitting a manuscript that is written in simple language and adheres to published guidelines.

All manuscripts submitted to Global Journals should include:

Title

The title page must carry an informative title that reflects the content, a running title (less than 45 characters together with spaces), names of the authors and co-authors, and the place(s) where the work was carried out.

Author details

The full postal address of any related author(s) must be specified.

Abstract

The abstract is the foundation of the research paper. It should be clear and concise and must contain the objective of the paper and inferences drawn. It is advised to not include big mathematical equations or complicated jargon.

Many researchers searching for information online will use search engines such as Google, Yahoo or others. By optimizing your paper for search engines, you will amplify the chance of someone finding it. In turn, this will make it more likely to be viewed and cited in further works. Global Journals has compiled these guidelines to facilitate you to maximize the web-friendliness of the most public part of your paper.

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A major lynchpin of research work for the writing of research papers is the keyword search, which one will employ to find both library and internet resources. Up to eleven keywords or very brief phrases have to be given to help data retrieval, mining, and indexing.

One must be persistent and creative in using keywords. An effective keyword search requires a strategy: planning of a list of possible keywords and phrases to try.

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One should start brainstorming lists of potential keywords before even beginning searching. Think about the most important concepts related to research work. Ask, “What words would a source have to include to be truly valuable in a research paper?” Then consider synonyms for the important words.

It may take the discovery of only one important paper to steer in the right keyword direction because, in most databases, the keywords under which a research paper is abstracted are listed with the paper.

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Numerical methods used should be transparent and, where appropriate, supported by references.

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Authors must list all the abbreviations used in the paper at the end of the paper or in a separate table before using them.

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Authors are advised to submit any mathematical equation using either MathJax, KaTeX, or LaTeX, or in a very high-quality image.

Tables, Figures, and Figure Legends

Tables: Tables should be cautiously designed, uncrowned, and include only essential data. Each must have an Arabic number, e.g., Table 4, a self-explanatory caption, and be on a separate sheet. Authors must submit tables in an editable format and not as images. References to these tables (if any) must be mentioned accurately.

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Figures are supposed to be submitted as separate files. Always include a citation in the text for each figure using Arabic numbers, e.g., Fig. 4. Artwork must be submitted online in vector electronic form or by emailing it.

Preparation of Eletronic Figures for Publication

Although low-quality images are sufficient for review purposes, print publication requires high-quality images to prevent the final product being blurred or fuzzy. Submit (possibly by e-mail) EPS (line art) or TIFF (halftone/ photographs) files only. MS PowerPoint and Word Graphics are unsuitable for printed pictures. Avoid using pixel-oriented software. Scans (TIFF only) should have a resolution of at least 350 dpi (halftone) or 700 to 1100 dpi (line drawings). Please give the data for figures in black and white or submit a Color Work Agreement form. EPS files must be saved with fonts embedded (and with a TIFF preview, if possible).

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Tips for Writing a Good Quality Science Frontier Research Paper

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1. Choosing the topic: In most cases, the topic is selected by the interests of the author, but it can also be suggested by the guides. You can have several topics, and then judge which you are most comfortable with. This may be done by asking several questions of yourself, like "Will I be able to carry out a search in this area? Will I find all necessary resources to accomplish the search? Will I be able to find all information in this field area?" If the answer to this type of question is "yes," then you ought to choose that topic. In most cases, you may have to conduct surveys and visit several places. Also, you might have to do a lot of work to find all the rises and falls of the various data on that subject. Sometimes, detailed information plays a vital role, instead of short information. Evaluators are human: The first thing to remember is that evaluators are also human beings. They are not only meant for rejecting a paper. They are here to evaluate your paper. So present your best aspect.

2. Think like evaluators: If you are in confusion or getting demotivated because your paper may not be accepted by the evaluators, then think, and try to evaluate your paper like an evaluator. Try to understand what an evaluator wants in your research paper, and you will automatically have your answer. Make blueprints of paper: The outline is the plan or framework that will help you to arrange your thoughts. It will make your paper logical. But remember that all points of your outline must be related to the topic you have chosen.

3. Ask your guides: If you are having any difficulty with your research, then do not hesitate to share your difficulty with your guide (if you have one). They will surely help you out and resolve your doubts. If you can't clarify what exactly you require for your work, then ask your supervisor to help you with an alternative. He or she might also provide you with a list of essential readings.

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7. Revise what you wrote: When you write anything, always read it, summarize it, and then finalize it.

8. Make every effort: Make every effort to mention what you are going to write in your paper. That means always have a good start. Try to mention everything in the introduction—what is the need for a particular research paper. Polish your work with good writing skills and always give an evaluator what he wants. Make backups: When you are going to do any important thing like making a research paper, you should always have backup copies of it either on your computer or on paper. This protects you from losing any portion of your important data.

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12. Know what you know: Always try to know what you know by making objectives, otherwise you will be confused and unable to achieve your target.

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Verbs have to be in agreement with their subjects. In a research paper, do not start sentences with conjunctions or finish them with prepositions. When writing formally, it is advisable to never split an infinitive because someone will (wrongly) complain. Avoid clichés like a disease. Always shun irritating alliteration. Use language which is simple and straightforward. Put together a neat summary.

14. Arrangement of information: Each section of the main body should start with an opening sentence, and there should be a changeover at the end of the section. Give only valid and powerful arguments for your topic. You may also maintain your arguments with records.

15. Never start at the last minute: Always allow enough time for research work. Leaving everything to the last minute will degrade your paper and spoil your work.

16. Multitasking in research is not good: Doing several things at the same time is a bad habit in the case of research activity. Research is an area where everything has a particular time slot. Divide your research work into parts, and do a particular part in a particular time slot.

17. Never copy others' work: Never copy others' work and give it your name because if the evaluator has seen it anywhere, you will be in trouble. Take proper rest and food: No matter how many hours you spend on your research activity, if you are not taking care of your health, then all your efforts will have been in vain. For quality research, take proper rest and food.

18. Go to seminars: Attend seminars if the topic is relevant to your research area. Utilize all your resources.

19. Refresh your mind after intervals: Try to give your mind a rest by listening to soft music or sleeping in intervals. This will also improve your memory. Acquire colleagues: Always try to acquire colleagues. No matter how sharp you are, if you acquire colleagues, they can give you ideas which will be helpful to your research.

20. Think technically: Always think technically. If anything happens, search for its reasons, benefits, and demerits. Think and then print: When you go to print your paper, check that tables are not split, headings are not detached from their descriptions, and page sequence is maintained.

21. Adding unnecessary information: Do not add unnecessary information like "I have used MS Excel to draw graphs." Irrelevant and inappropriate material is superfluous. Foreign terminology and phrases are not apropos. One should never take a broad view. Analogy is like feathers on a snake. Use words properly, regardless of how others use them. Remove quotations. Puns are for kids, not grunt readers. Never oversimplify: When adding material to your research paper, never go for oversimplification; this will definitely irritate the evaluator. Be specific. Never use rhythmic redundancies. Contractions shouldn't be used in a research paper. Comparisons are as terrible as clichés. Give up ampersands, abbreviations, and so on. Remove commas that are not necessary. Parenthetical words should be between brackets or commas. Understatement is always the best way to put forward earth-shaking thoughts. Give a detailed literary review.

22. Report concluded results: Use concluded results. From raw data, filter the results, and then conclude your studies based on measurements and observations taken. An appropriate number of decimal places should be used. Parenthetical remarks are prohibited here. Proofread carefully at the final stage. At the end, give an outline to your arguments. Spot perspectives of further study of the subject. Justify your conclusion at the bottom sufficiently, which will probably include examples.

23. Upon conclusion: Once you have concluded your research, the next most important step is to present your findings. Presentation is extremely important as it is the definite medium though which your research is going to be in print for the rest of the crowd. Care should be taken to categorize your thoughts well and present them in a logical and neat manner. A good quality research paper format is essential because it serves to highlight your research paper and bring to light all necessary aspects of your research.

Informal Guidelines of Research Paper Writing

Key points to remember:

- Submit all work in its final form.
- Write your paper in the form which is presented in the guidelines using the template.
- Please note the criteria peer reviewers will use for grading the final paper.

Final points:

One purpose of organizing a research paper is to let people interpret your efforts selectively. The journal requires the following sections, submitted in the order listed, with each section starting on a new page:

The introduction: This will be compiled from reference matter and reflect the design processes or outline of basis that directed you to make a study. As you carry out the process of study, the method and process section will be constructed like that. The results segment will show related statistics in nearly sequential order and direct reviewers to similar intellectual paths throughout the data that you gathered to carry out your study.

The discussion section:

This will provide understanding of the data and projections as to the implications of the results. The use of good quality references throughout the paper will give the effort trustworthiness by representing an alertness to 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 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 avoid:

- Insertion of a title at the foot of a page with subsequent text on the next page.
- Separating a table, chart, or figure—confine each to a single page.
- Submitting a manuscript with pages out of sequence.
- In every section of your document, use standard writing style, including articles ("a" and "the").
- Keep paying attention to the topic of the paper.
- Use paragraphs to split each significant point (excluding the abstract).
- Align the primary line of each section.
- Present your points in sound order.
- Use present tense to report well-accepted matters.
- Use past tense to describe specific results.
- Do not use familiar wording; don't address the reviewer directly. Don't use slang or superlatives.
- Avoid use of extra pictures—include only those figures essential to presenting results.

Title page:

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

Abstract: This summary should be two hundred words or less. It should clearly and briefly explain the key findings reported in the manuscript and must have precise statistics. It should not have acronyms or abbreviations. It should be logical in itself. Do not cite 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 approaches 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. Use comprehensive sentences, and do not sacrifice readability for brevity; you can maintain it succinctly by phrasing sentences so that they provide more than a 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 limit the initial two items to no more than one line each.

Reason for writing the article—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 for this; results of any numerical analysis should be reported. Significant conclusions or questions that emerge from the research.

Approach:

- Single section and succinct.
- An outline of the job done is always written in past tense.
- Concentrate on shortening results—limit background information to a verdict or two.
- Exact spelling, clarity 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 of comprehending and calculating the purpose of your study without having to refer to other works. The basis for the study should be offered. Give the most important references, but avoid making a comprehensive appraisal of the topic. Describe the problem visibly. If the problem is not acknowledged in a logical, reasonable way, the reviewer will give no attention to your results. Speak in common terms about techniques used to explain the problem, if needed, but do not present any particulars about the protocols here.

The following approach can create a valuable beginning:

- Explain the value (significance) of the study.
- Defend the model—why did you employ this particular system or method? What is its compensation? Remark upon its appropriateness from an abstract point of view as well as pointing out sensible reasons for using it.
- Present a justification. State your particular theory(-ies) or aim(s), and describe the logic that led you to choose them.
- Briefly explain the study's tentative purpose and how it meets 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 for every section. If you make the four points listed above, you will need at least four paragraphs. Present surrounding information only when it is necessary to support a situation. The reviewer does not desire to read everything you know about a topic. Shape the theory 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 soundly written procedures segment allows a capable scientist to replicate 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 to give the least amount of information that would permit another capable scientist to replicate 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 section, mention the specific item describing the way, but draw the basic principle while stating the situation. The purpose is to show 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:

Materials may be reported in part of a section or else they may be recognized along with your measures.

Methods:

- Report the method and not the 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—detail how procedures were completed, not how they were performed on a particular day.
- If well-known procedures were used, account for the procedure by name, possibly with a reference, and that's all.

Approach:

It is embarrassing to use vigorous voice when documenting methods without using first person, which would focus the reviewer's interest on the researcher rather than the job. As a result, when writing up the methods, most authors use third person passive voice.

Use standard style in this and 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 as 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. Use statistics and tables, if suitable, to present consequences most efficiently.

You must clearly differentiate material which would usually be incorporated in a study editorial from any unprocessed data or additional appendix matter that would not be available. In fact, such matters should not be submitted at all except if requested by the instructor.

Content:

- Sum up your conclusions in text and demonstrate them, if suitable, with figures and tables.
- In the manuscript, explain each of your consequences, and point the reader to remarks that are most appropriate.
- Present a background, such as by describing the question that was addressed by creation of an exacting study.
- Explain results of control experiments and give 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 manuscript.

What to stay away from:

- Do not discuss or infer your outcome, report surrounding information, or try to explain anything.
- Do not include raw data or intermediate calculations in a research manuscript.
- Do not present similar data more than once.
- A manuscript should complement any figures or tables, not duplicate information.
- Never confuse figures with tables—there is a difference.

Approach:

As always, use past tense when you submit 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 section.

Figures and tables:

If you put figures and tables at the end of some details, make certain that they are visibly distinguished from any attached appendix materials, such as raw facts. Whatever the position, each table must be titled, numbered one after the other, and include a heading. All figures and tables must be divided from the text.

Discussion:

The discussion is expected to be the trickiest segment to write. A lot of papers submitted to the journal are discarded based on problems with the discussion. There is no rule for how long an argument should be.

Position your understanding of the outcome visibly to lead the reviewer through your conclusions, and then finish the paper with a summing up of the implications of the study. The purpose here is to offer an understanding of your results and support all of your conclusions, using facts from your research and generally accepted information, if suitable. The implication of results should be fully described.

Infer your data in the conversation in suitable depth. This means that when you clarify an observable fact, you must explain mechanisms that may account for the observation. If your results vary from your prospect, make clear why that may have happened. If your results agree, then explain the theory that the proof supported. It is never suitable to just state that the data approved the prospect, and let it drop at that. Make a decision as to whether each premise is supported or 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 an experiment might be personalized to accomplish a new idea.
- Give details of all of your remarks as much as possible, focusing on mechanisms.
- Make a decision as to whether the tentative design sufficiently addressed the theory and whether or not it was correctly restricted. Try to present substitute explanations if they are sensible alternatives.
- One piece of 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 other available information. Present work done by specific persons (including you) in past tense.

Describe generally acknowledged facts and main beliefs in present tense.

The Administration Rules

Administration Rules to Be Strictly Followed before Submitting Your Research Paper to Global Journals Inc.

Please read the following rules and regulations carefully before submitting your research paper to Global Journals Inc. to avoid rejection.

Segment draft and final research paper: You have to strictly follow the template of a research paper, failing which your paper may get rejected. You are expected to write each part of the paper wholly on your own. The peer reviewers need to identify your own perspective of the concepts in your own terms. Please do not extract straight from any other source, and do not rephrase someone else's analysis. Do not allow anyone else to proofread your manuscript.

Written material: You may discuss this with your guides and key sources. Do not copy anyone else's paper, even if this is only imitation, otherwise it will be rejected on the grounds of plagiarism, which is illegal. Various methods to avoid plagiarism are strictly applied by us to every paper, and, if found guilty, you may be blacklisted, which could affect your career adversely. To guard yourself and others from possible illegal use, please do not permit anyone to use or even read your paper and file.

Criterion for Grading a Research Paper (Compilation) by Global Journals

Please note that following table is only a Grading of "Paper Compilation" and not on "Performed/Stated Research" whose grading solely depends on Individual Assigned Peer Reviewer and Editorial Board Member. These can be available only on request and after decision of Paper. This report will be the property of Global Journals.

Topics	Grades		
	A-B	**C-D**	**E-F**
Abstract	Clear and concise with appropriate content, Correct format. 200 words or below	Unclear summary and no specific data, Incorrect form Above 200 words	No specific data with ambiguous information Above 250 words
Introduction	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
Methods and Procedures	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
Result	Well organized, Clear and specific, Correct units with precision, correct data, well structuring of paragraph, no grammar and spelling mistake	Complete and embarrassed text, difficult to comprehend	Irregular format with wrong facts and figures
Discussion	Well organized, meaningful specification, sound conclusion, logical and concise explanation, highly structured paragraph reference cited	Wordy, unclear conclusion, spurious	Conclusion is not cited, unorganized, difficult to comprehend
References	Complete and correct format, well organized	Beside the point, Incomplete	Wrong format and structuring

INDEX

Global Journal of Science Frontier Research

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ISSN 9755896