1	Original Research Article
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3	Response of Okra to Incorporating Humic acid-derived Active Molecules into NPK
4	Fertilizer and Lime on Coastal Plain Sand in Calabar, Nigeria
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7	ABSTRACT
8	This study evaluated the response of okra to NPK fertilizer with humid acid and lime at the
9	University of Calabar, Teaching and Research Farm between October, 2015 and January,
10	2016 cropping season. The experiment was laid out in a randomized complete block design
11	(RCBD) with four replications. The experiment consisted of NPK with humic acid and liming
12	materials as treatments. The test crop was okra. The result of initial analysis of the chemical
13	properties of the soil before the experiment showed that the soil was acidic and was low in
14	exchangeable bases, organic matter (1.13%), total nitrogen (0.14%) and ECEC but high in
15	available phosphorus (26.63mg/kg) and base saturation. After application of treatment the
16	result showed that 6.2g of lime and 4.6g of NPK with humic acid increase the soil pH from
17	5.6 units in the control to 5.8 in NPK with Humic Acid, 6.0 in NPK with Humic Acid and
18	lime and 6.4 in lime. Organic carbon content, total nitrogen and available phosphorus also
19	increases in like manner. The results for the growth parameters showed that okra height,
20	number of leaves, and pod yield were significantly ($p \le 0.05$) different from the control. Based
21	on the result of this study further research is recommended in other locations to fully
22	ascertain the effects of this treatment using different combination.
23	
24	Keywords: Okra, Coastal plain sand, Humid acid, Lime, fertilizer
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27 28	1. INTRODUCTION
28 29	Okra (<i>Abelmoschus esculentus (L) Moench</i>), also known as ladies' fingers in many English-
30	speaking countries is a flowering plant that belongs to the family mallow, and it is popularly
31	grown for its immature fruits and young leaves throughout the tropical and sub-tropical parts

- of the world. In Nigeria and other African countries, okra is often grown in gardens as
 mixed crop alongside other arable crops [1]. Okra occupies about 1.5 million hectares of the
- 34 arable land in Nigeria alone [2] while the world production of okra as fresh fruit is estimated
- 35 at 1.7 million t/year [3].
- 36

Although okra has great potential both as domestic and commercial crop in Nigeria, but its yield on peasant farmer's fields is often very low, yielding about 2.10 t ha⁻¹ in Nigeria, which is far less compared to other countries like India (10.12 t ha⁻¹) and world average (7.65 t ha⁻¹) [4]. However, inadequate knowledge of the management of the soils, improved 41 varieties and some of the necessary cultural practices such as optimum plant density, and

42 nutrient requirements might have contributed to the low okra yield obtained by these farmers

- 43 <mark>[5]</mark>.
- 44

Soils of the rain forest zone of Nigeria are acidic in nature, classified as Ultisols and Oxisols 45 which are inherently low in pH, organic matter, nitrogen, potassium and other nutrients [6,7]. 46 47 These soils are prevalent in areas experiencing high annual rainfall (above 1500mm) and usually have problems associated with aluminum toxicity, low nutrient status, nutrient 48 49 imbalance and multiple nutrient deficiencies [8] and therefore require fertilization and liming for good crop growth and yield. Nitrogen is the main limiting nutrient for most crops on these 50 51 soils because of heavy leaching as a result of high rainfall couple with the sandy nature of the 52 soils [9].

53

54 Liming is a soil amendment management strategy of applying substances to manage or raise the pH of the soil to a favorable level. The overall effects of lime on soils include increased 55 pH, Ca, Mg and P availability, neutralization of toxic concentrations of Al and Mn, increase 56 57 in pH dependent CEC and improved nutrient uptake by plants [10,11]. According to 58 Oluwatoyinbo et al. [12], lime reduced the amount of P fertilizer required for optimum 59 growth and fruit yield of okra and obtained the highest yield at 0.5t/ha lime rate. There is 60 need for agricultural practices which are cheap and affordable for the local farmers, although 61 accurate rates of these local liming materials are yet to be established for optimum yield [13]. 62 Okra is often sensitive to slightly acidic soil in the tropics and hence was used as a test crop in this experiment. 63

It is worthy of note that nutrient management is the main factor that is accountable for 64 65 sustainable soil fertility. Over the years the use of inorganic fertilizer on crops increased yield as well as economic return on investment have been documented [7, 8, 12]. Humic acid is the 66 67 active constituent of organic humus, which can play a very important role in soil conditioning 68 and plant growth [14]. However, recent researches have explored the study area under 69 investigations in the context of mineral and organic fertilizers effect on crop growth and 70 nutrient uptake [15, 16, 17, 18], but no study was found specific to incorporating humic acid-71 derived active molecules into NPK fertilizer to ascertain its effect in crops in Calabar 72 environment, which is an important food producing area in Cross River State. However, this 73 study seeks to examine the response of okra to incorporating humic acid-derived active molecules into NPK fertilizer and lime on coastal plain sand in Calabar, Nigeria. 74

75 76

77 2. MATERIALS AND METHODS

78 2.1 Description of the Study Area

The study was conducted between October, 2015 and January, 2016 in poly pots laid out in 79 the open field at Crop Science Teaching and Research farm of the University of Calabar. 80 University of Calabar lies between latitude $05^{0}32^{1}$ North and longitude $07^{0}15^{1}$ and $9^{0}28^{1}$ East 81 [18]. The climate of the study area was characterized by high relative humidity and rainfall. 82 83 Rainfall is usually heavy occurring between the months of March to October with short dry spell in August. The mean total annual rainfall exceed 3000 mm per annum and the relative 84 humidity of 85% with the mean annual minimum and maximum temperature of 22.93 ^oC and 85 26.51 ^oC [19]. The rainfall spreads between April and October and characterized by two peak 86 87 periods and a short break in the month of August known as 'August break'.

88

89 **2.2 Soil sampling and Sample preparation**

The experimental site has been previously cultivated for yam, cassava, water leaf, fluted pumpkin, okra, maize etc. The experimental site was cleared, winnowed and the surface debris was removed before randomly collecting the soil samples. The soil samples were collected at the depth of 0-30cm as a composite of four (4) auger points from the experimental site. The samples were thoroughly mixed together and 1.5kg of the soil sample weighed into each pot of length 17cm and diameter 16.5cm.

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97 2.3 Experimental Design, Treatment, Planting and Planting Materials

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99 The experimental was laid out in a randomized complete block design (RCBD) with four 100 replications. The experiment consisted of NPK+ humic acid and liming materials as 101 treatment. The test crop okra and liming materials were gotten from Calabar local market, 102 while NPK with humic acid was obtained from commercial fertilizer plant in Jinan City, 103 China. The composition of lime use is calcium carbonate (CaCO₃) and composition of NPK 104 with humic acid; N₂ (10%), P₂O₅ (5%), K₂O (25%), micro-nutrients, that is, Baron, Zinc, 105 Manganese, Iron, Molybdenum were all 0.1% and humic acid (3%).

106 2.4 Data Collection

Data was collected on growth parameters include plant height, number of leaves, leaf lengthand leaf width. The number of leaves was determined by counting each plant once in two

109 weeks. The tools that were used for growth parameter data was meter rule. Biomass data

110 include stem girth and yield was considered at the end of the experiment.

111

112 **2.5 Laboratory Analysis**

113 In the laboratory, the soil sample collected was air dried, sieved using 2mm sieve, stored and 114 analyzed for physical and chemical properties using standard procedures. Particle size 115 distribution was determined by the hydrometer method using sodium hexametaphosphate 116 (calgon) as the dispersant [20]. The percentages of sand, silt and clay were determined. The 117 pH (in water) was determined in soil water ratio of 1:2.5 using a glass electrode pH meter standardized with buffer solutions 4.0 and 6.85 [20]. Organic carbon was determined by the 118 119 Walkley and Black wet oxidation method described by Udo et al. [20]. Total nitrogen was 120 determined by Macro-Kjeldahl Distilation Method [21] using Sodium Sulphate (NaSO₄) and 121 Copper Sulphate ($CuSO_4$) as a catalyst. Available phosphorus was extracted using the Bray-1 122 method [22] and determined calorimetrically with a spectrophotometer. Exchangeable bases 123 (Ca, Mg, K and Na) were extracted in 1 N NH₄OAc at pH 7. Potassium and sodium were 124 determined with a flame photometer while calcium and magnesium were determined by the 125 EDTA titration method [23]. Exchangeable acidity was by titration method using 1 N Kcl 126 extract [24]. Cation Exchange Capacity (CEC) was by summation of exchangeable bases 127 while Effective Cation Exchange Capacity (ECEC) was a summation of exchangeable bases 128 (Ca, Mg, K and Na) and exchangeable acidity. Percent base saturation was obtained by 129 dividing the total exchangeable bases (Ca, Mg, K and Na) by the effective cation exchange 130 capacity.

131 **2.6 Data Analyses**

132

133 The data was statistically analyzed using Analysis of Variance (ANOVA). Means were

compared using F-LSD test at 0.05 level of probability [25].

135

136 3. RESULTS AND DISCUSSION

- 137
- 138 **3.1** *Physical Properties*

140 **3.1.1** Particle size Analysis

The soil was high in sand (73.0%, 74.0% and 76.0%) for NPKHA, control and NPKHA + Lime and Lime, silt (14%, 15%, 16%, and 18%) for Lime, NPKHA + Lime, control and NPKHA and the clay (9%, 10%, and 11%) for NPKHA, control and Lime, and NPKHA + Lime. The soil still maintain sandy loam as its textural classes and such soil lack the absorption capacity for basic plant nutrients and water [26].

- 146
- 147 **3.2 Chemical Properties**
- 148

149 **3.2.1 Soil pH**

- 150 The pH increases with treatment, for instance control (5.6), NPKHA (5.8), NPKHA + Lime
- 151 (6.0), and lime (6.4). The increased soil pH was due to addition of lime.Liming material has
- the ability to raise soil pH from slightly acidic to near neutral [12].
- 153 3.2.2 Organic Carbon
- 154 The organic carbon content increase with treatments, for instance control (1.12%), NPKHA +
- Lime (1.54%), Lime (1.56%) and NPKHA (1.70%) which is below the critical level of 2%
- 156 for soils of the humid tropical region [27,28]. This low value obtained for the result might be
- 157 due to acidic nature of the humic acid fertilizer and non-availability of organic materials in
- the treatments. However, the value can be increase by including organic sources as part of the
 treatments
- 160 **3.2.3 Total nitrogen**
- 161 The value of total nitrogen reduces from control (0.07%), NPKHA + Lime and Lime (0.05%)
- and NPKHA (0.03%) which was lower than the critical level of 1.5% given by Aduayi *et al.*
- 163 [27]. Meanwhile nitrogen is one of the main nutrients needed by these crops. The decrease in
- 164 nitrogen might be attributed to leaching as a result of high rainfall that characterized the area
- 165 [9] alongside with the sandy nature of the studied soil. It has become one of the most limiting
- 166 nutrients in the soil.
- 167

168 **3.2.4 Available phosphorus**

- 169 The value of available phosphorus increases with the treatments Lime (18.00 mg/kg), control
- 170 (26.61 mg/kg), NPKHA (34.75 mg/kg) and NPKHA + Lime (40.13 mg/kg) as compared to
- the critical level of 8 mg/kg given by Aduayi *et al.* [27]. There was also a significant increase
- in available phosphorus, except on the sole application of lime which yielded the least

available phosphorus. The increased P level could be attributed to increased soil pH. It has

been reported that overall effects of lime on soils include increased pH level and availability

- 175 of nutrient including P availability [10].
- 176

177 3.2.5 Exchangeable Calcium, Magnessium, Potassium and Sodium

178 The value of calcium present in the soil increases with treatment Lime (4.5 cmol/kg), 179 NPKHA + Lime (3.9 cmol/kg), NPKHA (3.6 cmol/kg) and control (2.4 cmol/kg). The value of magnesium present increase with treatment Lime (2.7 cmol/kg), NPKHA + Lime (2.01 180 181 cmol/kg), NPKHA (1.6 cmol/kg) and control (0.6 cmol/kg). There was significant increase in the exchangeable Ca⁺⁺ in the treatment with Lime as well as the combination of Lime and 182 NPK with humic acid yielding the highest exchangeable Ca^{++} . The value of potassium in the 183 soil slightly increases with treatment Lime (1.08 cmol/kg), NPKHA + Lime (0.11 cmol/kg), 184 185 control (0.09 cmol/kg), NPKHA (0.08 cmol/kg). There was no significant increase in 186 magnesium and sodium. The values of sodium present in the soil increases with treatment 187 NPKHA and NPKHA + Lime (1.2 cmol/kg), Lime (0.91 cmol/kg) and Control (0.07) cmol/kg). The increase in level of most exchangeable bases maybe attributed to increased soil 188 189 pH [10].

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191 **3.2.6 Exchangeable acidity**

The values of Al^{+++} reduce in treatment with lime was 0.01 (cmol/kg) and NPKHA + Lime 0.09 (cmol/kg). The exchangeable acidity was low, this influencing the biochemical behavior of the soils. The values of H⁺ increased with treatment Lime 0.40 (cmol/kg), NPKHA + Lime (0.90 cmol/kg) and NPKHA (1.0 cmol/kg) and control (2.3 cmol/kg).

196

3.2.7 Effective Cation Exchange Capacity

The value of ECEC was low in control as established for productive soil [28]. The values of lime and NPKHA + Lime were also. This can be attributed to the low content of exchangeable cation.

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202 3.2.8 Base Saturation

The base saturation (Ca, Mg, K and Na) increases with treatment indicating that the supply of nutrients in the soil can support growth to an extent [11]. This show that the exchange complex of the soil particles is occupied by reserved acidity. There was a significant increase in the base Saturation (BS) with soils treated with sole applications of Lime giving the

- highest BS of 95.73%. The implication of this lime can reduce acidity and increase theexchangeable cations thereby making the soil fertile.

Physico-chemical properties	Quantity
Sand (%)	74.0
Silt (%)	16.0
Clay (%)	10.0
pH (H ₂ O)	5.6
Org. Carbon (%)	1.13
Total nitrogen (%)	0.14
Av. P (mg/kg)	26.63
Exchangeable cations (cmol/kg)	
Ca^{2+}	2.4
Mg^{2+}	0
K^+	0.09
Na ⁺	0.07
Exchangeable acidity (cmol/kg)	
Al^{3+}	1.32
H^{+}	2.36
ECEC	6.64
B.S (%)	44.58

Table 2: Physico-chemical after application of treatment

properties	Control	NPKHA	NPKHA+Lime	Lime
Sand (%)	74	73	74	76
Silt (%)	16	18	15	14
Clay (%)	10	9	11	10
pН	5.6	5.8	6.0	6.4
Organic Carbon (%)	1.12	1.70	1.52	1.56
Total Nitrogen (%)	0.07	0.03	0.05	0.05
Av. P	26.61	34.75	40.13	18.00
Exchangeable cation				
(cmol/kg)				
Ca ⁺⁺	2.4	3.6	3.9	4.5
Mg^{++}	0.6	1.6	2.01	2.7
K^+	0.09	0.08	0.11	1.08

Na ⁺	0.07	1.2	1.2	0.91
Exchangeable acidity				
(cmol/kg)				
Al^{+++}	1.32	4.4	0.09	0.01
H^+	2.34	1.0	0.90	0.40
ECEC	6.82	11.88	9.64	9.60
B.S (%)	46.33	54.55	74.90	95.73

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220 **3.3 Growth Parameter**

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222 3.3.1 Plant Height and Number of Leaves

In table 3, the response of okra to NPK with humic acid plus lime (NPKHA + Lime) in height 223 at four sampling periods (3WAP, 5WAP, 7WAP and 9WAP) during the growing seasons 224 225 showed that there was no significant increase in height (p>0.05) at 3WAP and 9WAP. However, at 5WAP and 7WAP, there was significant increase in plant height. A close 226 227 examination of the data reveals that control and lime pots grew taller than NPKHA + Lime 228 and NPKHA pots. This is as a result of inability of the plant roots to access the available 229 nutrients in the soils at different rate of addition of treatments. This result is not in accordance 230 with the findings of El-Meskser et al. [29] who reported that application of humic acid was 231 associated with a significant increase in plant height of crops. The difference in this result 232 could be attributed to sandy nature of the soil which tends to leach any applied nutrient. The 233 response of okra to NPKHA and lime on number of leaves were not significant (p>0.05) in 3WAP, 5WAP and 7WAP. At 9WAP, the test crop was significant (p>0.05). This is in 234 235 agreement with the findings of Oluwatovinbo *et al* [12], who observed significant increases 236 in plant height, number of leaves and fruits of okra with the application of all rates of lime up 237 to the 1000kg/ha rate used. This is not surprising as Oluwatovinbo *et al.* [12], had earlier reported that lime can increase optimum growth and fruit yield of okra and obtained the 238 239 highest yield at 0.5t/ha lime rate.

240

241 **3.3.2 Leaf length and Leaf width**

In table 4, the response of okra to NPK with humic acid plus lime (NPKHA+Lime) in leaf length and leaf width at four sampling periods (3WAP, 5WAP, 7WAP and 9WAP) during the

growing seasons showed that there was no significant difference ($p \le 0.05$) in both parameters.

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246 **3.3.3 Leaf area index**

247 In table 5, the response of okra to NPK with humic acid plus lime (NPKHA +Lime) in leaf area index at four sampling period (3WAP, 5WAP, 7WAP and 9WAP) during the growing 248 249 seasons showed that there was no significant difference at ($p \le 0.05$) in the leaf area index. 250 This is a pointer to the fact that changes in numbers of leaves are bound to alter the overall 251 performance of the plant as the leaf serves as the photosynthethic organ of the plant [30]. 252 Also the unfertilized plants had lower leaf area due to less number of leaves resulting from premature leaf fall and early vine senescence [31]. The higher leaf area index associated with 253 254 the fertilized plants was probably due to higher number of leaves.

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257 3.3.4 Biomass data and Pod Yield

The response of okra to NPK with humic acid plus lime (NPKHA + Lime) in Biomas data and pod yield is presented in Table 6. The result shows that there was no significant difference at ($p \le 0.05$) in stem girth and root. However, there was marginal increase in stem girth and roots with NPKHA + Lime (2.62 and 0.60). These results is not in harmony with the findings of Cimrin and Yilmaz [32] who stated that humic substances have been shown to stimulate shoot and growth and nutrient uptake of vegetable crops. The result obtained for this study maybe due to the sandy nature of the experimental soil.

265

The result shows that there was significant difference at ($p \le 0.05$) in pod yield. Crop yield is significantly increased by integrated use of lime and fertilizer. These results are good agreement with those obtained by Karakurt *et al.* [33] who found that humic acid enhanced nutrient uptake, plant growth, yield and quality in a number of plant species. Lime had significant effect on all the yield attributes name fresh pod weight (Table 6).

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Table 3: Response of NPKHA and Lime on Plant Height and Number of Leaves

Treatment	Plant He	ight			Number	of Leaves		
	3WAP	5WAP	7WAP	9WAP	3WAP	5WAP	7WAP	9WAP
Control	7.17	11.87	14.2	16.23	4.67	4.83	5.5	7.67
NPKHA	4.35	6.05	6.88	10.52	3.16	4.33	5.33	5.5
NPKHA + Lime	4.28	4.9	9.02	14.93	3.16	4.00	5.17	5.83
Lime	5.28	9.12	11.97	15.27	3.83	4.5	4.67	5.67
F-LSD (0.05)	NS	**	**	NS	NS	NS	NS	**

274	WAP = Week after planting; NPKHA (NPK with Humic Acid) and NPKHA+Lime (NPK
275	with Humic Acid and Lime)
276	Note: NS means not significant * means Significant at 5 %
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284 Table 4: Response of NPKHA and Lime on Leaf Length and Leaf Width

Treatment	Leaf Leng	gth			Leaf Wi	dth		
	3WAP	5WAP	7WAP	9WAP	3WAP	5WAP	7WAP	9WAP
Control	3.26	4.67	5.1	5.82	3.6	5.43	6.38	7.52
NPKHA	2.18	3.52	2.93	4.82	2.2	3.58	4.22	6.08
NPKHA + Lime	2.6	4.53	6.22	6.95	2.71	5.03	7.52	8.85
Lime	3.08	3.93	4.63	5.22	3.22	4.9	4.52	6.52
F-LSD (0.05)	NS	NS	NS	NS	NS	NS	NS	NS

285 WAP = Week after planting; NPKHA (NPK with Humic Acid) and NPKHA+Lime (NPK

286 with Humic Acid and Lime)

287 Note: NS means not significant * means Significant at 5 %

288

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Table 5: Response of NPKHA and Lime on Leaf Area Index

3WAP	5WAP	7WAP	9WAP	
0.04	0.03	0.04	0.05	
0.06	0.04	0.02	0.06	
0.02	0.03	0.06	0.18	
0.03	0.02	0.04	0.13	
	0.04 0.06 0.02	0.04 0.03 0.06 0.04 0.02 0.03	0.04 0.03 0.04 0.06 0.04 0.02 0.02 0.03 0.06	0.04 0.03 0.04 0.05 0.06 0.04 0.02 0.06 0.02 0.03 0.06 0.18

Image: NPKHA+Lime (NPK with Humic Acid and Lime) Note: NS means not significant * means Significant at 5 % Table 6: Response of NPKHA and lime on biomas data Treatment Stem girth Root Pod yield Control 1.33 NPKHA + Lime 2.62 NPKHA + Lime 2.62 Lime 2.6 NPKHA + Lime 2.62 NPKHA + Lime 2.64 NPKHA + Lime 1.027 MAP = Week after planting; NPKHA (NPK with Humic Acid) and NPKHA+Lime (NPK with Humic Acid and Lime) Note: NS means not significant * means Significant at 5 % CONCLUSION The results from this study showed that for optimum performance of okra on of derived soil, the application of NPK with humic acid (HA) and lime respectively as this gave the highest pod yield. It also revealed the potency of NPK with humic										
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- 311 ascertain the performance of this fertilizer in okra and other arable crops.
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