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NEW METALLOCHROMIC INDICATORS FOR COMPLEXOMETRIC TITRATION OF COPPER (II) IN PRESENCE OF INTERFERING SPECIES

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ABSTRACT: Six hydroxytriazenes has been introduced as a metallochromic indicator in the complexometric titration with EDTA and applied successfully to the titration of copper (II). A direct method of titration of Cu (II) with EDTA using these hydroxytriazenes has been proposed. The change of colour of the solution at the end point during titration was from yellow to green at a pH range of 5.5-6.0, 5.05-5.50, 5.0-5.5 for reagent number (i, ii, iii, vi), iv, v respectively. The titration can be performed at the temperature range of 25-60℃. The minimum concentration of copper (II) which can be titrated is as follows: 1.0 x 10^{-3} M Cu (II), 3.0 x 10^{-3}M Cu (II), 5.0 x 10^{-3}M Cu (II) when using reagent number (i, iii, v, vi), iv, ii respectively as metallochromic indicators. Interference studies of a number of cations and anions have been studied. Cl^-, Br^-, CH_3COO^-, PO_4^{3-}, SO_4^{2-},$C_2O_4^{2-}$, $S_2O_3^{2-}$, NO_3^-, SO_3^-, S^{2-}, HPO_4^{2-}, F^-, NO_3^-, WO_4^{2-},$Mo_2O_{24}^{6-}$, I^-, NH_4^+, Na^+, K^+, Ba^{2+}, Hg^{2+}, Mg^{2+}, Ca^{2+} can be tolerated on the titration of Cu (II) up to tenfold excess. However, {U(vi), Sn(II), Zr(iv), Co(II), Zn(II), Ni(II)}, {Pb(II), Th(IV)}, {Mn(II), Cd(II)} interfered at equivalent amounts, fivefold excess, tenfold excess respectively.

Keywords: Complexometric, Metallochromic indicator, Hydroxytriazenes, Copper determination, Interference species, Chelators, Elemental**.**

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1.INTRODUCTION

Copper is widely used in industrial, domestic and high technology applications because of its properties such as: high ductility, thermal and electrical conductivity, malleability, resistance to

corrosion [1]. The low position of copper in reactivity series makes it not to corrode and also to be used in decoration, jewelry, statutes and parts of the building to make them attractive for many years [2]. Combination of copper with other metals such as zinc, tin, nickel forms alloys with desirable characteristics which can be utilized for highly specialized applications depending on their composition. The use of copper-nickel alloy to the hulls of ships is due to the fact that it does not corrode in sea water and is capable of reducing adhesion of marine life, thus reducing drag and increasing efficiency. Copper being a non-magnetic and non-sparking is utilized in special tools and military applications. The research has shown that copper has antibacterial, antifungal and antiviral activities [3]. [4] revealed that human corona virus 229E dies within hours on a copper surface. They attributed this to copper ion being released and also generation of reactive oxygen species (ROS) which are responsible in inactivation of HUCOV-229E on copper and copper alloy surfaces. The inactivation resulted to fragmentation of the viral genome making the process of inactivation being irreversible. [5] further demonstrated that the novel corona virus which is purely responsible for the COVID-19 pandemic cannot survive on copper surfaces. Copper also plays major roles in human, animals and plants. The functions of copper in the human body includes making red blood cells, maintaining immune system and nerve cells, helping the body to form collagen absorption of iron and energy production. It can also help to keep certain conditions at bay like anemia and osteoporosis. However, ingesting high levels of soluble copper salts can lead to hepatic cirrhosis, hemolytic anemia and degeneration of basal ganglia [6]. These excessive levels can be brought about through diet, food supplements and water [7, 8]. Sea food (oysters, lobster, squid, mussels), organ meats, nut and the chocolate are the major sources of copper [9]. Hence the need to determine the levels of copper in the diet, supplements and water using simple, cheap, sensitive and rapid method to avoid excessive copper in the body. The use of pesticides, fungicides, industrial effluent and waste water irrigation in developing countries contributes in pollution of agricultural soils by copper which lowers the growth and yield of foods resulting to food safety threats and food insecurity [10]. The high level of copper is cytotoxic because it acts as the catalyst for the reaction responsible for generating reactive oxygen species (ROS) which results to increase in oxidative stress in plants. The disruption of secondary structure of thiol bonds within proteins will take place whenever there is a free copper ion. The leaf chlorosis and growth inhibition will be experienced whenever excessive amount of copper is used [11]. This implies that the information pertaining the residual effect of micronutrient fertilization which can result to reduced crops to be made available using cost-effect rapid and interference free method. [12] Showed that 3-hydroxy-3-phenyl-1-m-chlorophenyltriazene can be used as metallochromic indicator for the determination of copper with EDTA as a titrant in the presence of certain cations and anions. Furthermore, [13] employed a direct method to determine copper employing 3-hydroxy-3-phenyl-1-p-chlorophenyltriazene, 3-hydroxy-3-phenyl-1-m-chlorophenyltriazene and 3-hydroxy-3-methyl-1-p-sulphonamidophenyltriazene as

metallochromic with EDTA as a titrant. The work by these authors gave hopes of obtaining metallochromic indicator belonging to hydroxytriazenes which can be used in environmental samples containing a large number of interfering species without involving in separation techniques. Moreover, hydroxytriazenes is easy to synthesize and the yield is always high [14]. Hence the need for continued searching.

2. MATERIALS AND METHODS

Equipment/Apparatus, reagents and chemical

Ultra-pure water equipment (model milli-Q equipped with Q-POD), pH meter (HI22 II pH/or Meter), HANNA instruments, Elemental analyzer Perkins Elmers model 2400 CHN/O Analyzer, melting point apparatus (model Kruess M 500), ultra-pure water, copper (II) sulphate pentahydrate (99.999%), disodium ethylenediamine tetra acetic acid (99.9%), ethyl alcohol (95% V/V), perchloric acid (99.9985%), Acetone (99.9%), anhydrous sodium acetate (99.9%), Ethyl alcohol (95% V/V), Tris(hydroxymethylaminomethane)– Buffer (99.5%), murexide (ammonium purpurate) (99.9%).

Synthesis of hydroxytriazenes

The three steps which were followed during the synthesis of hydroxytriazenes include; reduction of Nitro compounds, diazotization of amino compound, and coupling [15, 16].

Detection of Hydroxytriazenes Using Spot Tests

∝– Naphthylamine Test

∝– Naphthylamine solution in acetic acid was added drop wise to acetic acid containing small amount of hydroxytriazene. An immediate development of red or reddish-brown or brown colour which intensified on gentle warming of the test solution confirms the presence of hydroxytriazenes.

N, N–Dimethylaniline Test

A few drops of N, N–dimethylaniline and 0.5 ml of concentrated hydrochloric acid was added to a pinch of hydroxytriazenes followed by heating to boiling. An immediate development of red, reddish brown or brown colour indicated the compounds prepared were hydroxytriazenes [17].

Picric Test

4 to 5 drops of saturated solution of Picric acid in acetone was added to a pinch of Hydroxytriazene followed by heating on a water bath set at 58℃. An immediate development of yellow, orange or reddish colour demonstrated that the compound synthesized were Hydroxytriazenes.

Sulphuric acid Test

1.0 ml of concentrated Sulphuric acid was added to a pinch of hydroxytriazene and then heated slowly for a few seconds. The immediate appearance of a brown or reddish brown colour revealed that the prepared compounds were Hydroxytriazenes. The Hydroxytriazenes synthesized gave positive tests.

Purification of Hydroxytriazenes by crystallization

The choice of the solvent used in crystallization was based on the solubility. The activated charcoal was also used to improve crystallization by adsorbing coloured impurities at their surface areas. The crude hydroxytriazene was placed in a beaker, ethanol (For reg no. i to vi, ix) while acetone (For reg no. viii) was added gradually with constant stirring while heating the mixture on a water bath. The heating continued to nearly boiling, a beaker containing solution of hydroxytriazenes was removed from the water bath and then ultra-pure water was added drop wise until the solution became just cloudy. After this, the solution was again taken to water bath for heating. The heating continued until the solution became clear again. Thereafter, the activated charcoal was added immediately with constant stirring. The hot solution was then filtered through a fluted filter paper and the hot filtrate was collected in a dry beaker. The residue on a Buchner funnel was washed using a hot solvent which was used during crystallization. During the process of filtration, the funnel was kept hot in order to avoid crystallization on its stem. The filtrate was left to cool gradually at a room temperature until some crystals were seen starting to form. Immediately the beaker containing solution of hydroxytriazene was taken into the freezer for further cooling for 12 hours. Then, the filtration under vacuum followed while employing a Buchner funnel. In some cases, crystallization had to be induced by microscopic scratches in the glass surface which provided sharp edges hence assisting in the start of crystal growth [18]. The physical characterization and Elemental analysis were done in order to establish the purity and the composition of each of the hydroxytriazene synthesized.

Preliminary investigation

The suitability of the nine Hydroxytriazenes as metallochromic indicator for copper (II) determination was investigated by using direct titration. The procedure involved taking a 10 ml solution of 1.0 x 10^{-2}M Cu^{2+} titrating it against equimolar solution of EDTA in pH range of 5.0-6.0 at room temperature employing 1.0% tris-buffer solution and 1.0% per chloric acid solution for pH adjustment.

Effect of pH Variation on Copper (II) Determination

The research has shown that, metallochromic indicators do respond to the change in the pM value of the metal ion and also on the pH of the solution. This implies that, for any new metallochromic indicator, it is necessary to establish the optimum pH range where the colour change at the end point is very sharp and most perceptible. This was done by titrating 10 ml of 1.0 x 10^{-2}M Cu^{2+} solution with 1.0 x 10^{-2}M EDTA solution at various pH utilizing 1.0% tris-buffer and 1.0% perchloric acid solutions for adjusting the pH.

Effect of Temperature Variation on Copper (II) Determination

A series of experiments were carried out in a wide temperature range to establish the optimum temperature range at which the colour change at the end point of titration was sharp and perceptible

with hydroxytriazenes. The results indicated that the titration can be carried out in the temperature range of 25 - 60℃. Hence, subsequent titrations were carried out at room temperature (25℃). This was important in that temperature can attribute to change of pH as well as dissociation of copper complex and this can have effect on the results [19]. The procedure involved titrating 10 ml of 1.0 x 10^{-2}M Cu^{2+} with equimolar EDTA solution under optimum conditions of solvent and pH while using Hydroxytriazenes as metallochromic indicator. The specific temperatures were maintained with the help of water bath.

Effect of Concentration Variation on Copper Determination

This was studied in order to determine the minimum concentration of copper (II) solution which can be determined under optimum conditions when using each of Hydroxytriazene as metallochromic indicator. This was done by titrating 1.0 x 10^{-3}M to 1.0 x 10^{-2}M solution of copper (II) against equimolar solution of EDTA using Hydroxytriazenes as indicator.

Effect of Diverse Ions in Copper (II) Determination

The utility of this method was established by studying the effect of various diverse ions at equivalent amount, fivefold excess and tenfold excess. Using each of the hydroxytriazenes as metallochromic indicator under optimum conditions. The results obtained are summarized below.

Procedure of Titration

10 ml of Copper (II) solution was diluted to 30 ml using ethanol-water mixture (reg no. i to vii, ix) or acetone-water (reg no viii) due to their solubility. 1.0% tris-buffer and 1.0% perchloric acid solution were used for adjustment of pH. 5 to 6 drops of Hydroxytriazenes were added resulting to the development of yellow colour. The titration was carried slowly at room temperature with equimolar solution of EDTA. Towards the end, one or two drops of indicator were further added to enhance the perceptibility of the endpoint. The point where colour sharply changed to green was recorded as the end point.

3. RESULTS AND DISCUSSION

The results of physical properties and elemental analysis of Hydroxytriazenes are given in table 1.

Table 1: Physical Properties and elemental Analysis

S/No	Physical Properties			Elementary Analysis					
	Colour and shape of Crystals	Crystalized from	M.P ^{0}C	%C		H%		N%	
				Th	Exp	Th	Exp	Th	Exp
(i)	Light-yellow shining needles	Ethanol	180	47.39	47.22	3.67	3.69	12.76	12.32
(ii)	White-yellow shining Plates	Ethanol	59	21.66	21.47	1.55	1.48	10.83	11.07
(iii)	White-yellow needles	Ethanol	136	62.85	62.47	4.83	5.04	18.34	18.50
(iv)	Brownish-yellow shining needles	Ethanol	118	64.16	63.86	5.38	5.18	17.28	17.62
(v)	Orange-yellow needles	Ethanol	122	64.16	63.81	5.38	5.13	17.28	17.63
(vi)	Orange-yellow needles	Ethanol	124	64.16	63.90	5.38	5.18	17.28	17.59
(vii)	Light-yellow shining needles	Ethanol	112	65.33	64.96	5.88	5.77	16.34	16.67
(viii)	Light-yellow shining needles	Acetone	70	59.64	59.34	4.62	4.39	16.06	16.27
(ix)	Light-yellow shining needles	Ethanol	178	61.96	61.64	4.83	4.69	15.49	15.16

Key:

(i)	3-Hydroxy-3-m-tolyl-p-sulphonato (Sodium salt) phenyltriazene	$C_{13}H_{12}N_3O_4SN_9$
(ii)	3-Hydroxy-3-methyl-1-(2,4,6-tribromophenyl) triazene	$C_7H_6N_3OBr_3$
(iii)	3-Hydroxy-3-phenyl-1-m-hydroxyphenyltriazene	$C_{12}H_{11}N_3O_2$
(iv)	3-Hydroxy-3-o-tolyl-1-m-Hydroxyphenyltriazene	$C_{13}H_{13}N_3O_2$
(v)	3-Hydroxy-3-m-tolyl-1-m-Hydroxyphenyltriazene	$C_{13}H_{13}N_3O_2$
(vi)	3-Hydroxy-3-p-tolyl-1-m-Hydroxyphenyltriazene	$C_{13}H_{13}N_3O_2$
(vii)	3-Hydroxy-3-m-tolyl-1-p-Methoxyphenyltriazene	$C_{14}H_{15}N_3O_2$
(viii)	3-Hydroxy-3-m-tolyl-1-o-Chlorophenyltriazene	$C_{13}H_{12}N_3OCl$
(ix)	3-Hydroxy-3-m-tolyl-1-o-Carboxyphenyltriazene	$C_{14}H_{13}N_3O_3$

The four-spot test (∝- Naphthylamine test, N.N-Dimethylaniline test, picric tyest and sulphuric acid test) gave positive results with each Hydroxytriazene synthesized. Reagent no(i) to (vii, ix) were crystalized from ethanol whereas reagent no (viii) from the acetone. The choice of the solvent was based on the solubility of these hydroxytriazenes. A close examination of table 1 revealed that the colour of reagent no (i), (vii, viii and ix) was light yellow shinning needles while for reagent no (ii), iii), (iv), (v) and (vi) the colour was white yellow shining plate, light yellow needles, brownish yellow shinning needles, orange-yellow needles and orange-yellow needles respectively. The process of crystallization was repeated until a constant, sharp and characteristic melting point with the range of about 0.5^0C was obtained. This was an indication that the compounds are now in pure form. The temperature at which individual hydroxytriazenes just started to melt was taken as the melting point. It was observed that as the crystallization was being repeated, the melting point kept on increasing with corresponding decrease in the melting point range. This was attributed to the decrease in the amount of soluble impurities in the synthesized hydroxytriazenes as the crystallization was being repeated [20, 21]. The melting point of the synthesized hydroxytriazenes ranged between 59^0C to 180^0C. The author attributed these differences in melting to different molecular shape and intermolecular forces between molecules of hydroxytriazenes like London dispersion forces, hydrogen bonding forces and other intermolecular interactions [22]. Examination of table 2 indicates that all hydroxytriazenes synthesized had absolute error for carbon, hydrogen, Nitrogen of less than 0,4% which showed that the compound do not need to be purified again and also indicated that the experimental and theoretical values agreed so well hence the intended compound had been prepared [23]. The different errors obtained during elemental analysis were associated with the properties of the analyzed Hydroxytriazenes and the character of their thermal decomposition [24].

Table 2: Absolute and relative Error in %

S/No	Absolute Error%			Relative Error%		
	C	H	N	C	H	N
(i)	-0.17	+0.02	-0.28	-0.36	+0.54	-0.02
(ii)	-0.19	-0.07	+0.24	-0.88	-4.52	+2.22
(iii)	-0.37	+0.21	+0.18	-0.59	+4.32	+0.98
(iv)	-0.30	-0.20	+0.34	-0.47	-3.72	+1.97
(v)	-0.35	-0.25	+0.35	-0.55	-4.65	+2.03
(vi)	-0.26	-0.20	+0.31	-0.41	-3.72	+1.79
(vii)	-0.37	-0.11	-0.33	-0.57	-1.87	+2.02
(viii)	-0.30	-0.23	+0.21	-0.50	-4.98	+1.31
(ix)	-0.32	-0.14	-0.33	-0.52	-2.90	-2.13
	- Observed value is big + Observed value is small					

The preliminary investigation results are summarized in table 3.

Table 3: Preliminary Investigation

S/No	Colour change at the end	Volume of EDTA consumed (ml)	Remark
(i)	Yellow-green	10.0	Sharp end point
(ii)	Yellow-green	10.0	Sharp end point
(iii)	Yellow-green	10.0	Sharp end point
(iv)	Yellow-green	10.0	Very Sharp end point
(v)	Yellow-green	10.0	Very Sharp end point
(vi)	Yellow-green	10.0	Very Sharp end point
(vii)	Yellow ppt on addition EDTA	-	Not suitable
(viii)	Yellow ppt on addition EDTA	-	Not suitable
(ix	No change in greenish Yellow colour even in excess of EDTA	-	Not suitable

Conc. of Cu(II) Solution = 5.0×10^{-3}M

Conc. of EDTA Solution = 5.0×10^{-3}M

pH of titration using Hydroxytriazenes = 5.0 – 6.0

Volume of Cu(II) solution taken =10.0ml

Volume of Cu(II) obtained using murexide (pH range 10-11) taken =10.0ml

The results indicated that reagent no (i) to (vi) can be used as metallochromic indicator using EDTA as a titrant for copper (II) determination. The colour at the end point changed from yellow to green at pH range of 5.0 – 6.0. The results which were obtained by these six indicators were comparable with those of murexide indicator. The sharp end point was obtained with each of these six Hydroxytriazenes indicators. Reagent (vii) and (viii) gave yellow ppt when EDTA was added while for reagent (ix) no change in greenish yellow colour was observed, hence these three hydroxytriazenes were found not suitable. The results of the optimum pH range effect of temperature and concentration range in complexometric determination of Cu (II) using hydroxytriazenes as metallochromic indicator are shown in Table 4.

Table 4: Determination of 3.1773mg of Cu(II) at different concentration using Hydroxytriazenes as Indicator

S/No	Conc. of copper (II) ions		1.0×10^{-2}M	5.0×10^{-3}M	3.0×10^{-2}M	1.0×10^{-3}M
	Conc. of EDTA		1.0×10^{-2}M	5.0×10^{-3}M	3.0×10^{-2}M	1.0×10^{-3}M
	pH range of the titration	Temp. range of the titration	Titre Value in ml			
(i)	5.5-6.0	25-60	10.0	10.0	10.0	10.0
(ii)	5.5-6.0	25-60	10.0	10.0	9.8	-
(iii)	5.5-6.0	25-60	10.0	10.0	10.0	10.0
(iv)	5.05-5.50	25-60	10.0	10.0	10.0	9.7
(v)	5.0-5.5	25-60	10.0	10.0	10.0	10.0
(vi)	5.5-6.0	25-60	10.0	10.0	10.0	10.0

Titration volume of Copper (II) solution =10.0ml

Volume of EDTA consumed corresponding to each concentration of copper (II) using Murexide as indicator =10.0ml

The results from this table shows that the pH range of titration for reagent (i),(ii),(iii),(vi) was 5.5-6.0 whereas for reagent (iv), (v) were 5.05-5,50, 5.0-5.5 respectively. The results from Table 4 further revealed, the hydroxytriazenes in the present investigation gave accurate results in the temperature range of 25-60^{0}C. This shows that, they have a large temperature range which can be utilized for complexometric titration for copper determination compared with nitrosochromotropic acid indicator which has only the temperature range of 20^{0}C to 40 ^{0}C and also Nitrochromotropic acid indicator cannot be used for determination of copper in the presence of alkaline earth metal such as Ba^{2+} Mg^{2+} [19] whereas the six Hydroxytriazenes synthesized can be used. The results of the effect of concentration on complexometric determination of copper (II) using Hydroxytriazenes as metallochromic indicators are also given in table 4. The objective of this investigation was to establish the minimum concentration of Copper (II) which can be determined accurately using these new indicators. The results revealed that, using reagent (i), (iii), (v) and (vi) as metallochromic

indicators, Cu (II) can be determined as low as 1.0 $\times 10^{-3}$M. The relative error of -2% was obtained when reagent (ii) was used as indicator during the titration of 3.0 $\times 10^{-3}$M. Cu^{2+} with equivalent amount of EDTA. A relative error of -3% was realized when reagent (iv) was used for determination of 1.0 $\times 10^{-3}$M Cu^{2+} using equivalent amount of EDTA. Hence, reagent (ii) and (iv) are less sensitive compared to reagent (i), (iii), (v) and (vi). The six hydroxytriazenes have advantages over Eriochrome black T due to the fact that, direct titration can be performed whereas Eriochrome black T will require back titration to be used. Furthermore, titration when using Eriochrome black T must be performed rapidly with small amount of copper resulting a great margin error. The high concentration of copper can be titrated using Eriochrome black T only when 70% organic solvent such as ethanol is used to retard the reaction between Cu-EDTA complex with Eriochrome black T [25].

Effect of diverse ions

The interference study was carried out in order to establish the effect of various cations and anions on the quantitative determination of an aliquot containing 3.1773mg of Cu (II). The following ions did not interfere with the determination of 3.1773mg even when they were present in tenfold excess: Cl^-, Br^-, CH_3COO^-, CO_3^{2-}, PO_4^{3-}, SO_4^{2-},$C_2O_4^{3-}$, $S_2O_3^{2-}$, NO_3^-, SO_3^{2-}, S^{2-}, HPO_4^{2-}, F^-, NO_3^-, WO_4^{2-},$MO_2O_{24}^{6-}$, I^-, NH_4^+, Na^+, K^+, Ba^{2+}, Hg^{2+}, Mg^{2+}, Ca^{2+}. However, U(IV), Mn(II), Pb(II), Sn(II), Th(Iv), Cd(II), Zr(IV), Co(II), Zn(II) and Ni(II) interfere with the titration of Cu(II) with positive relative error as given in Table 5 and Table 6 below.

Table 5: Relative error in the determination of 3.1773mg of Cu(II) in the presence of foreign ions at equivalent amount

Reagent	Interfering species					
	U(VI)	Sn(II)	Zr(IV)	Co(II)	Zn(II)	Ni(II)
(i)	+3	+20	+10	+6	+3	+7
(ii)	+30	+24	+21	+2	+9	+7
(iii)	+9	+3	+4	+9	+2	+9
(iv)	+2	+3	+2	+4	+9	+2
(v)	+10	+4	+2	+41	+27	+39
(vi)	+38	+11	+42	+52	+22	+50

Table 6: Relative error in the determination of 3.1773mg of Cu(II) in the presence of foreign ions at five and ten fold excess

Reagent	Interfering species			
	Fivefold excess		Ten-fold excess	
	Pb(II)	Th(IV)	Mn (II)	Cd(II)
(i)	+8	+5	+2	+2
(ii)	+3	+2	+8	+6
(iii)	+9	+8	+9	+3
(iv)	+2	+7	+8	+6
(v)	+8	+3	+4	+31
(vi)	+8	+2	+2	+21

The species which interfered at low level were not studied at high level

4. CONCLUSION

Six new metallochromic indicators have been introduced for the direct complexometric determination of Cu (II) using EDTA as a titrant. The results obtained by these six indicators are comparable to those of murexide indicator. The titration can be performed in the presence of a large number of cations and anions. However, [U(VI), Sn(II), Zr(IV), Co(II), Zn(II), Ni(II)] [Pb(II), Th(IV)], [Mn(II), Cd(II)] interferes at equivalent amount, five-fold excess and tenfold excess. The proposed method is simple, rapid and fairly selective and can be used to a wide range of applications.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Not applicable.

HUMAN AND ANIMAL RIGHTS

No Animals/Humans were used for studies that are base of this research.

CONSENT FOR PUBLICATION

Not applicable.

AVAILABILITY OF DATA AND MATERIALS

The author confirms that the data supporting the findings of this research are available within the article.

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CONFLICT OF INTEREST

The author has no conflict of interest.

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