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DETERMINATION OF ELEMENTS IN NIGERIAN APHRODISIAC HERBS BY THERMAL NAA

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Abstract

In this paper fresh medicinal plants that are popularly believed to have aphrodisiac properties namely Tacca involucrate, Imperata cylindrical, Cyperus esculentum, Momordica charantia, Garcinia kola, and Fadogia agrestis were collected in the form of stem bark, nuts, fruits, seeds, and roots. Instrumental Neutron Activation Analysis (INAA) was applied for multi-elemental determination of six aphrodisiac herbs used as treatment of Sexual dysfunction. Concentrations of twenty two elements Al, As, Ba, Br, Ca, Co, Cr, Cs, Dy, Eu, Fe, Hf, K, La, Lu, Mg, Mn, Na, Rb, Sb, Sc, Sm, Ta, Th, Ti, U, V, Yb, and Zn have been determined by short and long irradiation with a thermal neutron flux of $5*10^{11} ncm^{-2}s^{-1}$, and the spectra analyzed using WINSPAN 2004 software package. Several recent researches indicate that impaired sexual function which is now a common problem may be improved by Iron and Zinc supplements. Our result shows that

Fadogia agrestis (black aphrodisiac) recorded high Zn (123 ± 11 μgg^{-1}) which was more than the amount (95 μgg^{-1}) in Jaipal sex tonic, (92.02 and 84.24 μgg^{-1}) in the popular aphrodisiac Panax gingseng and Radix codonopsis respectively. The level of K in Imperata cylindrica, and Tacca involucrata, was 19,170 μgg^{-1} and 19,340 μgg^{-1} respectively. These values are about three times that of Aphrodisiac powder. The present study can be used to some extent to assess the safety of aphrodisiac herbs in the final formulation. Quality control and quality assurance of the method was tested by analyzing two Standard Reference Materials (IAEA-359 and SRM-1573) simultaneously with the samples.

Key words: *Aphrodisiac, Sexual dysfunction, Thermal NAA*

1. INTRODUCTION

Traditional herbs have brought about a revolutionary breakthrough in the management of sexual inadequacies and are known worldwide as an instant treatment (Adimoelja et al., 2000). The increasing incidence of male sexual dysfunction is necessitating more and rapid search for plants with aphrodisiac potentials. This is very important because of the side effects associated with other treatment options and the readily availability of medicinal plants and world is fast turning to the use of medicinal plants for managing sexual dysfunctions. Sexual dysfunction is more prevalent in males than in females, (Guay et al., 2003). It has been discovered that men between 17 and 96 years old could suffer sexual dysfunction as a result of psychological or physical health problems (Joe 1990).

Aphrodisiacs act as catalysts that stimulate sexually, thus enhance potency for reproduction. The Aphrodisiacs are used to address the problem of repeated inability to achieve normal sexual intercourse (male impotence) which in addition may contribute to infertility (Yakubu et al., 2003). Before the advent of fast and accurate multi-elemental analytic techniques, studies on medicinal plants were mostly concentrated on their organic contents, including essential oils, glycosides, vitamins, alkaloids saponin and other active components (Yakubu et al., 2005; 2006). It is now well established that many trace elements play a vital role in curing diseases (Lamari *et. al*., 2008) and enhancing sexual desire, (Obi *et. al.,* 2006) as well as general well-being (Barry et al., 1996; Funtua *et. al.*, 2003).

The effect and influences of these elements on administration of medicinal plant has received relatively little attention (Funtua *et. al.*, 2003). The studies of elemental contents of herbal medicine have been documented in literature (Serfor et al., 2001; Funtua *et. al., 2003;* Obi *et. al.,* 2006; Muh'd et al., 2007; Lamari *et. al., 2008;* Rajeev et al., 2010; Mohammed *et al*., 2011). However, the present study is apparently the first multi elemental analysis of Nigerian aphrodisiac herbs (Mohammed *et al*., 2011). Some of the mineral elements that are reported to address the issue of impaired sexual function which is now a serious medical and social symptom that occurs in 10-52% of men and 25-63% of women (Porst, 2004), include Mn (Guenther and Konieczynski, 2003), Zinc (Serfor *et. al.,* 2001; Chaturvedi et al. 2004), and Iron (Shils et al., 1988). In this study one of the sensitive analytical techniques INAA was applied to determine the elemental content in commonly used aphrodisiac herbs in Nigeria.

2. EXPERIMENTAL

Six different medicinal plants that are adjudged to have aphrodisiac properties were collected in the form of stem bark, nuts, fruits, seeds, and roots from Dambo, Dakace Shika, and Sabon-Gari market in Zaria Northern Nigeria. The sampling sites were shaded and depicted in fig 1. After collection, each of the samples was put into a light polyethylene bag which was again put into a larger and stronger polyethylene bag. Surface contaminants of the plant samples were removed by washing with deionized water twice and then with deionized double distilled water to remove superficial dust. The leaves were first air dried and then dried at 80^{o}C in an oven to constant weight.

The samples were powdered in agate mortar and passed through 125 μm-mesh sieve. Two sub samples of the homogenized analyte samples were prepared from each sample giving a total of 12 subsamples for this work. The samples have weight between the range of 0.2422 mg and 0.2897 *mg* and each placed in a high density polyethylene vial and weighted using a Mettler Toledo balance model CH-8606 (manufactured by Mettler-Toledo AG Greifensee, Switzerland) and the vial heat sealed. Two analyte samples of certified reference material IAEA-359 (Cabbage) and SMR 1573 (Tomato Leaves) prepared and packed in a similar manner was used as quality control standard to validate the analytical technique. The irradiation was achieved using a neutron flux of 5x 10^{11} n cm^{-2} s^{-1} of Nigerian Research Reactor-1 (NIRR-1).

Table 1 List of the aphrodisiac herbs and the parts used for aphrodisiac treatment

No	Botanical Name	Family Name	Plant part used	Sample source
1	*Tacca involucrate*	*Taccaceae*	Roots	Dambo
2	*Imperata cylindrical*	*Poaceae*	Rhizome (root)	Dakace
3	*Cyperus esculentum*	*Cyperaceae*	Nuts	Sabon Gari market

4	*Momordica charantia*	*Cucurbitaceae*	Roots and fruits	Dambo
5	*Garcinia kola*	*Guttiferae*	Seeds	Sabon Gari market
6	*Fadogia agrestis*	*Rubiaceae*	Stem	Shika

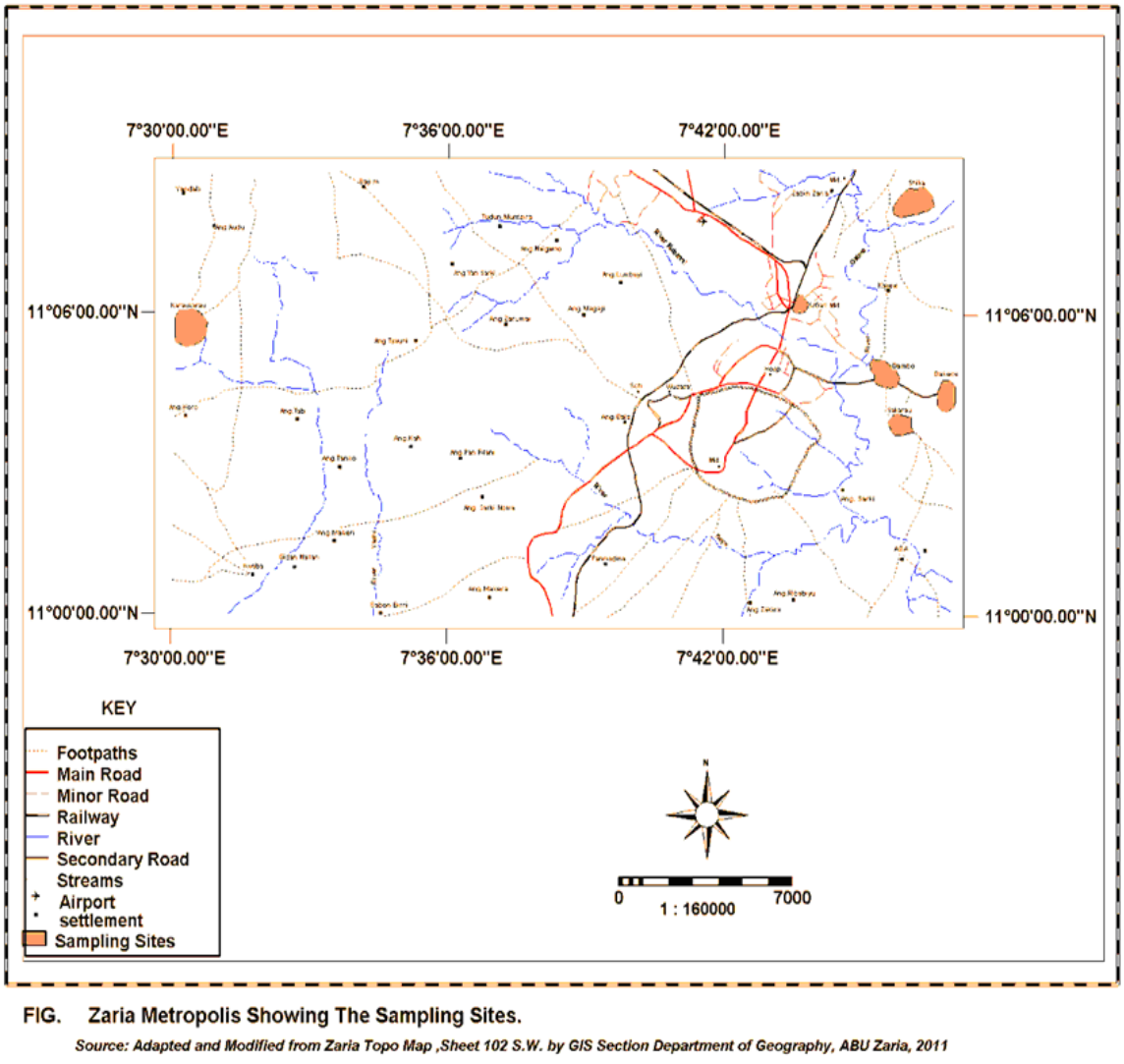


Fig 1. Map of Zaria metropolis showing the sampling sites for Aphrodisiac Plants

2.1 Irradiation Counting and Measurement

The INAA with NIRR-1 facilities utilize two analytical procedures. The first involves a short irradiation regime followed by gamma ray measurements after appropriate decay periods. This procedure is suitable for quantifying the following elements: Al, Br, Ca, Cd, Cl, Dy, I, K, Mn, Na, and Ti, V. The second procedure involves a long irradiation regime of approximately 6–7 h and is suitable

for the following elements: As, Au, Ba, Ce, Co, Cr, Cs, Eu, Fe, Ga, Gd, Hf, Ho, La, Lu, Nd, Rb, Sb, Sc, Se, Sm, Sr, Ta, Tb, Th, U, Yb, W, and Zn. The irradiation and counting procedures adopted for NIRR-1 were adequately described earlier by Jonah et al. (2006). The data processing was done by using the gamma ray spectrum analysis software WINSPAN version 2004 (Liyu, 2004), a software developed at CIAE, Beijing, China. On the basis of the well-known activation equation, the software requires that calibration factors be pre-determined by a multi-element standard reference material for elements of interest using adopted irradiation and counting regimes.

Basically, the relative standardization NAA method uses (Eq. 2.0) to obtain the concentration of the element of interest with high presicion of up to parts per million (ppm).

$$\frac{C_{std}}{C_{sample}} = \frac{\mathrm{W_{std}}}{\mathrm{W_{sample}}} \qquad (2.0)$$

C_{std} and C_{sample} are the count rates of standard and sample respectively. W_{std} is the mass of the element in the standard. W_{sample} which is the mass in the sample can then be calculated if the other three parameters are known.

3. RESULTS AND DISCUSSION

The results of INAA analysis of aphrodisiac herbs used in this work is displayed in Table 2. The presence of the elements in different plants was confirmed by the gamma energy of the corresponding isotope formed. This shows the unique advantage of INAA technique in terms of its multi-element capability and ability to analyze major, minor and trace elements in a single analysis. The variation in elemental concentration is mainly attributed to the differences in botanical structure, as well as the mineral composition of the soil in which the plants are cultivated. Other factors responsible for a variation in elemental content are preferential absorbability of the plant, use of fertilizers, irrigation water and climatological conditions (Rajurkar and Pardeshi, 1997). The calcium content in various medicinal plants analyzed varies from 571-11090 mg/kg with the highest concentration from *Fadogia agrestis* and the lowest concentration from *Cyperus esculentum,* this extremely high Ca content may be used to overcome Ca deficiency. Moreover, earlier research on human has shown that optimal intakes of Ca can reduce individual risk factors for health problems such as cardiovascular disease (Mertz 1982; Sanchez *et al*., 1998), a study by Greger (1989) shows that a high calcium intake may inhibit the intestinal absorption of iron, zinc, and other essential minerals in humans.

Zinc is among the nutritionally important element (Paul *et al*., 2007). It is also known to prevent impaired spermatogenesis (Chaturvedi *et al*., 2004), which may be part of reasons why herbal plants containing high concentration of Zn are used in treatment of infertility. On the other hand Zinc deficiency is characterized by recurrent infections, lack of immunity, growth retardation poor appetite, skin changes, and mental lethargy (FAO/WHO, 1974). Estimated safe and adequate daily intake for Zn is between 10,000 and 20,000μg/day (N.R.C, 1989). However, prolonged consumption of large doses can result in some health complications such as fatigue, dizziness and neutropenia bleeding, boils, wounds, insect bites, and skin disease (Hess *et al*., 2002). All the aphrodisiac herbs analyzed except *Garcinia kola* contain Zn within the concentration range of 18-123 µg/g. The level of Zn in *Garcinia kola* was found to be below the detection limit which indicates that Zn is not active ingredient for the aphrodisiac property of this herb.

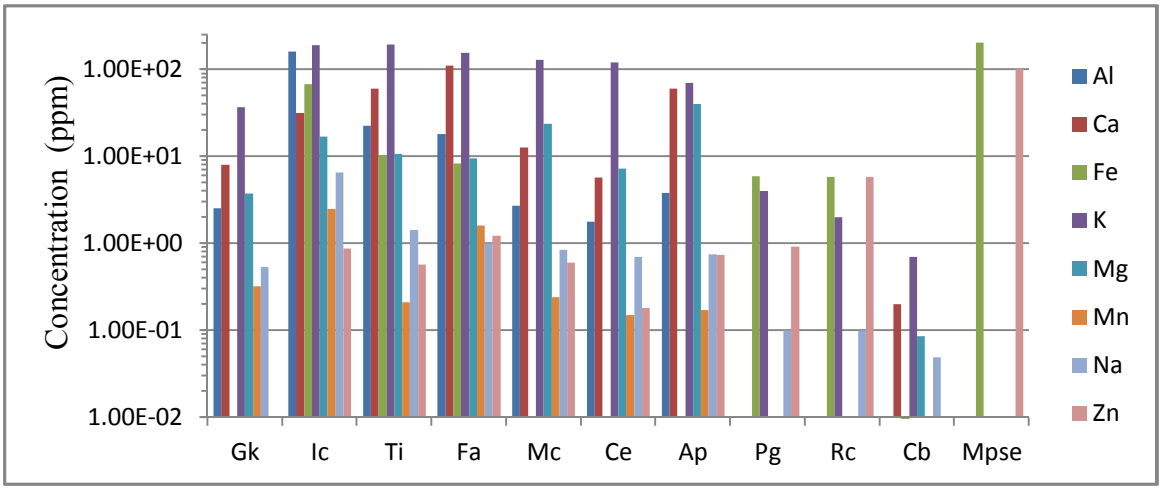
Moreover, Cobalt is regarded as an essential element and form part of vitamin B_{12} required for red-blood synthesis. Cobalt also aid in fixing of nitrogen in the root tubercles of plants. The Co content of *Garcinia kola, Momordica charantia* and *Cyperus esculentum* was below the detection limit, the highest observed was 3.4 mg/kg in *Imperata cylindrical.* It should however be noted that there is a

wide margin of safety between toxic concentration and nutritional required level of Co (Awofolu *et al*., 2005). However, adverse chronic effects of Co ingestion at concentration of $2mgl^{-1}$ may occur (DWAF, 1996). *Imperata cylindrical* contained the highest level of Al (16270±135), whereas *Cyperus esculentum* contain the lowest level (179 ± 15.4). There is little information on effects of high level consumption of Al in healthy individuals (Funtua *et al*., 2003).

The major medical conditions associated with increased levels of Al are chronic or end stage renal disease with or without dialysis (Cornelis *et al*., 1996). It was reported that the presence of Fe in the body is associated with transfer of oxygen from lungs to tissue cells and with hemoglobin in the blood cells (Sigel, 1978). Fe deficiency mainly caused by poor dietary intake, excessive menstrual-flow or multiple births constitutes major nutritional deficiency in humans (Reddy et al; 1987) and usually results in anemia. Fe content in the various samples analyzed in this work ranges from 833 mg/kg in *Fadogia agrestis* to 6846 mg/kg in *Imperata cylindrica*. Therefore intake of some of these plants can readily compensate for Fe deficiency. The intake of these plants should be administered with caution though because excessive intake of Fe above the WHO recommended daily intake level of 10-30 mg/day to avoid chronic overload of Fe in the body.

The result for the concentrations of Mn and Na, and Zn in *Cyperus esculentum, Imperata cylindrical,* respectively agreed with the values obtained for aphrodisiac powder by (Seifor *et. al*., 2001). Our result shows that the elements of Al, Ca, K, Na,Zn, Mn, Fe,Ti, and Mg were found to be most enriched in the samples analyzed. *Fadogia agrestis* (black aphrodisiac) (Yakubu *et al*., 2007) recorded high Zn ($123 \pm 11 \mu gg^{-1}$) which is more than the amount ($95.0 \mu gg^{-1}$) in Jaipal sex tonic as reported by Shils *et al* (1988) as well as 74.2 ± 5.2 in aphrodisiac powder by Serfor *et al* (2001), 4.08 ± 0.01mg/100g in aphrodisiac *Chlorophytum borivilianum* by Rajeev *et al* (2010), as well as in *Panax gingseng* a remarkable known aphrodisiac prescription in the traditonal Chinese (Chen, 1997; and Paola, 2001) recorded $92.02 \mu gg^{-1}$ of Zn as reported by (Hamzah *et al*., 2004). In addition the Zn concentration in *Imperata cylindrica* agrees closely with that of *Panax gingseng* and *Radix codonopsis.* However, it is very small compared to $10{,}250 \mu gg^{-1}$ in Nigerian ready-to use herbal product called Man Power sexual energy as reported by Obi *et al* (2006). This indicates that Zn is one of the active ingredients in the Man Power herbal product. Also, the level of Fe in Man power sexual energy (Nigerian herbal remedies that is used in the treatment of low sperm count), was 20, 500 μgg^{-1} as reported by Obi *et al*, (2006). This value is three times the highest value of Fe recorded in this work.

It was reported that the stem or bark of a species of *Corynanthe pachycerus* (Agbovie *et al*., 2002), the bark of the species of *Pausinystalia yohimbe* (Fabricant *et al*., 2001), and the fruit or leaf of a species of *Oxyantllus unilocularis,* (Sugiyama *et al*., 1992) all contain aphrodisiac activity. This is an interesting similarity, with *Fadogia agrestis* which belong to the same family as the above mentioned plants. However, the active elements in the above mentioned plants might not be the same as in the *Fadogia agrestis,* probably, due to the differences in the soil type; availability of soil water; and pH value of the soil. Similarly the flower or fruit of *Trichosanthes dioica* (Wild snake-gourd), the seed of *Cucurbita pepo* (Pumpkin), and the fruit of *Benincasa hispida* (Ash gourd) which belong to the same family with *momordica charantia* were reported by Joy (1998) to show aphrodisiac activity. Also, the leaf of momordica charantia was reported also by Sharma (1960) to contain aphrodisiac activity. Sugiyama, (1992) again reported that the bark of *Garcinia afzelii* Engl and the leaf of *Garcinia kola* Heckel contain aphrodisiac activity. The comparison between the concentration of Al, Ca, Fe, K, Mg, Mn, Na, and Zn in this work and those obtained by other workers is depicted in Figure 1



Gk= *Garcinia kola*, Ic = *Imperata cylindrica*, Ti = *Tacca involucrate*, Fa = *Fadogia agrestis*, Mc = *Momordica charantia*, Ce = *Cyperus esculentum*, Ap = Aphrodisiac powder (2001), Pg = *Panax gingseng* (2004), Rc = *Radix codonopsis* (2004), Cb = *Chlorophytum borivilianum* (2010), Mpse = Man power sexual energy (2006).

Fig. 1 Comparison between concentration of element for this work and the work done by others

Table 3 Elemental concentration (in μgg^{-1}) of reference materials used for quality control.

Elements	IAEA-359 (Cabbage)	
	This work	Certified values
Na	601±33.7	580±28
K	32300±230	32500±264
	SRM-1573 (Tomato Leaves)	
	This work	Certified values
Zn	61.81±19.8	62±6
Rb	16.72±4.23	16.5±0.1
Fe	982.3±235.8	690±25

Table 2 Elemental concentration (in μgg^{-1}) for Nigerian aphrodisiac herbs

ELEMENTS(	Garcinia kola	Imperata cylindrica	Tacca involucrate	Fadogia agrestis	Momodica charantia	Cyperus esculentum
Al	253 ±22	16270±135	2255 ± 183	1820±151	272 ± 24	179 ± 15.4
As	BDL	0.66 ± 0.10	BDL	BDL	BDL	BDL
Ba	BDL	245 ± 28	56 ± 11	60 ± 12	BDL	BDL
Br	0.63±0.1	11.8 ± 1.5	1.4 ± 0.2	3.7 ± 0.3	0.67 ± 0.05	2.13 ± 0.6
Ca	811±16	3149 ± 554	6020 ± 89	11090±16	1262 ± 228	571 ± 181
Co	BDL	3.4 ± 0.2	0.8 ± 0.2	1.0 ± 0.2	BDL	BDL
Cr	BDL	19 ± 3	BDL	BDL	BDL	BDL
Fe	BDL	6846 ± 281	1033 ± 89	833 ± 103	BDL	BDL
Hf	BDL 5	16.6 ± 1.4	1.1 ± 0.1	2.2 ± 0.2	BDL	0.8 ± 0.1
K	3671±30	19170 ±1380	19340 ± 1334	15690±109	12970±908	12030±217
La	0.19±0.02	14.1 ± 1.0	3.9 ± 0.3	5.4 ± 0.4	0.16 ± 0.02	0.19 ± 0.02
Mg	376 ±78	1699 ± 285	1081 ± 166	961 ± 166	2397 ± 175	733 ± 87
Mn	32 ± 1	248 ± 10	21 ± 1	162 ± 7	24 ± 1	15 ± 0.2
Na	53 ± 3	661 ± 33	144 ± 8	100 ± 6	84 ± 5	70 ± 2
Rb	10 ± 2	81 ± 6	23 ± 2	30 ± 4	11 ± 3	14 ± 2
Sc	0.05±0.01	2.24 ± 0.11	0.25 ± 0.02	0.35 ± 0.02	0.03 ± 0.01	0.04 ± 0.01
Ta	BDL	0.6 ± 0.1	BDL	BDL	BDL	BDL
Th	BDL	7.5 ± 1.0	2.0 ± 0.3	0.8 ± 0.1	BDL	0.6 ± 0.1
Ti	BDL	2053 ± 335	127 ± 29	282 ± 62	BDL	BDL
V	BDL	11.3 ± 1.5	1.6 ± 0.2	1.9 ± 0.3	0.3 ± 0.1	BDL
Yb	BDL	2.2 ± 0.3	0.26 ± 0.06	0.53±0.10	BDL	BDL
Zn	BDL	87 ± 7	57 ± 6	123 ± 11	60 ± 4	18 ± 5

CONCLUSIONS

This study has demonstrated that mineral elements play a vital role in addressing the problem of sexual dysfunction. It is hoped that this work will provide information that can help to standardize the use of aphrodisiac herbs, because, without the knowledge of the active ingredients, it is difficult to standardize a product and its dose, to guarantee efficacy and also to enhance quality control and quality assurance. It should be noted that QA/QC values obtained in Table 3 are for method validation only not elemental. Therefore, since our samples are purely biological in matrix IAEA-359 (Cabbage)

and SRM-1573 (Tomato Leaves) are used. The element concentrations in these standard materials are in good agreement with those obtained in our samples.

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