ORIGINAL ARTICLE

PATTERN OF SODIUM THIOPENTAL-INDUCED SLEEP IN WISTAR RATS IN DOSES OF ARTESUNATE AND ETHANOLIC EXTRACT OF *GARCINIA KOLA*

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Background: There are several reports of the medicinal applications of *Garcinia kola* and the efficacy of various other uses of this plant. The fact remains that emphasis is rapidly shifting from synthetic drugs to the use of plant materials for medicinal formulations. This study was undertaken to compare the patterns of thiopental-induced sleep in separately administered doses of *Garcinia kola* extract and artesunate in Wistar rats. **Methods:** The onset and duration of the induced sleep were computed in 40 mg/Kg dose of thiopental sodium in Wistar rats, after pretreatments in separate doses of *Garcinia kola* extract and artesunate. **Results:** Opposing influences or negative effects on the induced sleep was observed in doses of *Garcinia kola* extract. A clear facilitating influences or promoting effects on the induced sleep was observed in doses of artesunate. **Conclusion:** It is believed that the dopaminergic neural mechanism may have accounted for the opposing influences of *Garcinia kola* on the thiopental-induced sleep observed in this study. The present result was also suggestive that the facilitating influences of artesunate on the induced sleep was mediated through GABA neuronal mechanism in the central processes of sleep production.

Keywords: Artesunate, *Garcinia kola*, Facilitatory influences, Induced sleep, Wistar rat

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INTRODUCTION

It is well known that most plants are the natural sources of compounds or active ingredients that are often employed in the formulation of important drugs. The fact remains that medicines derived from plants have made significant contributions to human health over the years.[1] With the shifting of attention from synthetic drugs to natural plant products, the use of plant extracts for the treatment of ailments and enhance organs systems for the improvement of general body performance is now on the increase.[2]

Garcinia kola, is a plant that is indigenous to sub-Saharan, Central and West Africa, and has been extensively used in Africa for herbal medicinal formulations.[3,4] Thus, the plant has over the years been used in the treatment of various diseases, including liver disorders and diarrhoea[5–7], diabetes, bronchitis and throat infections[8,9]. The plant has also found use traditionally as a natural antimicrobial agent[10], and it has been reported to possess hepatoprotective and aphrodisiac properties.[11] Other notable bioactivity properties attributed to *Garcinia kola* includes bronchodilatory effects[8] as well as its known effectiveness in the treatment of dermatological disorders associated with melanin pigmentation[12]. The known active constituents of *Garcinia kola* include the dimeric flavonoid molecules fused with biflavonoids, xanthones and benzophenones.[13]

Artesunate is a semi synthetic derivative of artemisinin, the active compound extracted from the Chinese herb *Artemisia annua*.[14] It is highly effective against multi-drug resistant strains of *Plasmodium falciparum* hence its increasingly wide usage for the treatment and management of malaria.[15] The drug is well tolerated at therapeutic doses; therefore a lot of people, pregnant women inclusive, take the drug.

Sleep is a behavioural state and is found in all animals, including birds, fish, reptiles, foetuses etc. Sleep can actually be defined as the naturally recurring state of relatively suspended sensory and motor activity in the animal, characterised by total or partial unconsciousness and nearly complete inactivity of voluntary muscles.

Sodium thiopental is a short acting barbiturate and it has been noted that it acts on $GABA_A$ receptors (inhibitory channels) in the brain and spinal cord[16] to induce sleep. This duration of action was found to be modified by various compounds which act on the central nervous system, the duration of action of sodium thiopental was shown to be lengthened by central nervous system depressant and shortened by central nervous system stimulant. The present study aimed to compare the pattern of sodium thiopental-induced sleep in varying doses of artesunate and extracts of *Garcinia kola* in Wistar rats.

MATERIAL AND METHODS

Adult Wistar rats of both sexes and of comparable age and weight were used for this study. The animals were allowed to acclimatize in the new environment for about two weeks before the commencement of the study. They were allowed free access to standard feeds and water throughout the experiments. The rats were arranged into six groups by the duration of the administration and treatment doses of artesunate and extract of *Garcinia*

kola. Group A and B animals that served as control, received 40 mg/Kg of thiopental sodium in acute dose. Group C were pre-treated with acute doses of ethanolic extract of *Garcinia kola* prior to administration of acute doses of thiopental sodium. Group D animals received chronic doses of *Garcinia kola* for a period of 2 weeks prior to acute dose of thiopental sodium. Groups E and F were similarly treated as the C and D animals with doses of artesunate solution and thiopental sodium, respectively.

Fruits of *Garcinia kola* were chopped into small pieces and air dried at room temperature and milled into powder. The powdered *Garcinia kola* was stored at room temperature until used. Weighed samples of the *Garcinia kola* were soaked in appropriate quantities of ethanol solvent and left to stand for about 24 hours. This was then filtered to separate the residue. The filtrate was then evaporated with crucial water bath into paste and stored in airtight sample containers in a refrigerator until used. Known quantity of artesunate was dissolved in physiological saline and the mixture was left to stand for about one hour before filtration. Various doses used were 10, 20, 40, 80 and 160 mg/Kg for both *Garcinia kola* and artesunate, respectively, and these were administered intraperitoneally. The choice and doses of *Garcinia kola* and artesunate used in this study were based on observations from previous study. The rats were kept in transparent cage for easy observation of the onset and duration of the induced sleep in all cases using a stopwatch.

RESULTS

The results in Table-1 revealed significant increase ($p \leq 0.05$, $p \leq 0.01$) in the time of the onset of the induced sleep in increasing acute doses of *Garcinia kola* extract from 40–320 mg/Kg. There was no definite pattern of alterations in the duration of the induced sleep at the different doses of the *Garcinia kola* extract, except the slight decrease ($p \leq 0.05$) seen at 320 mg/Kg dose.

Table-1: Onset and duration of sodium thiopental-induced sleep in Wistar rats in acute doses of artesunate and ethanolic extract of *Garcinia kola* (Min, Mean±SEM)

Dose	GARCINIA COLA		ARTESUNATE	
(mg/Kg)	Onset	Duration	Onset	Duration
Control	6.10±0.18	85.24±3.14	6.10±0.18	85.24±3.14
10	7.08±0.12	84.40±3.19	6.20±0.24	85.56±1.20
20	7.24±0.25	83.06±5.20	5.16±0.13	86.96±1.50
40	8.06±0.11*	80.56±4.75	4.30±0.51*	90.84±3.21
80	10.08±0.14*	82.10±2.65	3.68±0.33*	92.06±3.40
160	14.16±0.16**	70.08±1.50	2.70±0.35**	97.94±4.10*
320	14.04±0.10**	55.04±6.53*	2.56±0.22**	98.04±4.50*

*$p \leq 0.05$, **$p \leq 0.01$

Pattern of the onset time of the induced sleep which showed significant decrease ($p \leq 0.05$, $p \leq 0.01$) in increased doses of artesunate (40–320 mg/Kg), with corresponding increases ($p \leq 0.05$, $p \leq 0.01$) in the duration of the induced sleep. It shows that the animals go into the sleep quicker, and they stay longer in the induced sleep in doses of artesunate (Figure-1).

The patterns of the induced sleep in chronic doses of *Garcinia kola* extract appeared confirmatory to the suggestion of possible opposing influence on sleep. This was explicit in the observed increases in the onset time of induced sleep ($p \leq 0.05$, $p \leq 0.01$), with the decreases in duration of the induced sleep (Figure-2).

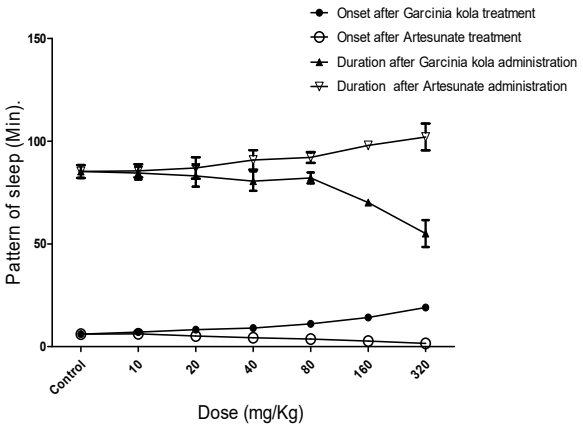


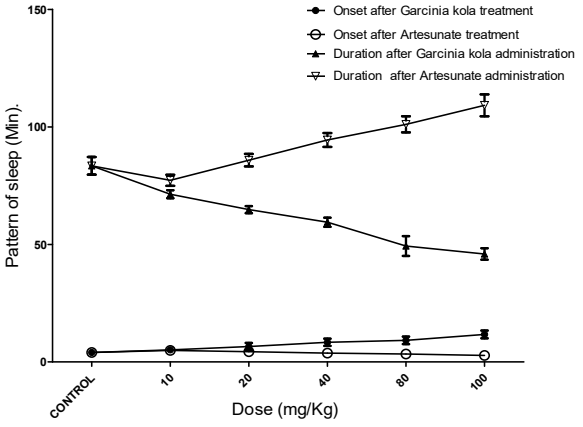
Figure-1: The pattern of sodium thiopental-induced sleep in acute doses of ethanolic extract of *Garcinia kola* and Artesunate in Wistar rats

Table-2: Onset and duration of sodium thiopental sleep in Wistar rats in chronic doses of ethanolic extract of *Garcinia kola* and Artesunate (Min, Mean±SEM)

Dose	GARCINIA COLA		ARTESUNATE	
(mg/Kg)	Onset	Duration	Onset	Duration
Control	4.36±0.14	83.43±3.76	4.36±0.14	83.43±3.76
20	5.13±0.25	71.34±1.70	4.15±0.18	77.24±2.37
40	6.56±0.50*	64.82±1.5*	4.06±0.16	88.25±2.37*
60	8.37±0.55**	59.43±1.95*	3.76±0.13	94.45±2.96*
80	9.19±0.95**	48.29±4.22**	3.36±0.10*	101.08±3.43**
100	11.72±0.61**	49.50±2.43**	2.76±0.11**	109.18±4.67**

*$p<0.05$, **$p<0.01$)

Figure-2: Pattern of sodium thiopental-induced sleep in chronic doses of ethanolic extract of *Garcinia kola* and Artesunate in Wistar rats



DISCUSSION

This study was designed to examine the patterns of sodium thiopental-induced sleep in separately administered doses of ethanolic extract of *Garcinia kola* and artesunate in Wistar rats. It was the aim of this study to compare possible modulating influences on thiopental-induced sleep in Wistar rat in separately administered doses of artesunate and *Garcinia kola* extract.

The results revealed significant increase in the time of the onset of the induced sleep in increasing acute doses of *Garcinia kola* extract from 40 to 320 mg/Kg. There was no definite pattern of alterations in the duration of the induced sleep at the different doses of the *Garcinia kola* extract, except the slight decrease seen at 320 mg/Kg dose. Thus, it was obvious that the animals were staying longer time before falling into sleep in doses of *Garcinia kola* extract, with the decrease in the duration of the sleep at 320 mg/Kg dose. These observations are seen as indicative of possible opposing effects of *Garcinia kola* on sleep production. It is therefore possible that *Garcinia kola* may have some clearly definable modulating influences on central mechanism of sleep. However, the present result appears to be at variance with the work of Braide[6] who reported a prolonged pentobarbital sleeping time in Wistar rats in doses of *Garcinia kola*.

Pattern of the onset time of the induced sleep which showed significant decrease ($p \leq 0.05$, $p \leq 0.01$) in increased doses of artesunate (40–320 mg/Kg), with corresponding increases ($p \leq 0.05$, $p \leq 0.01$) in the duration of the induced sleep. From this observation, it shows that the animals go into the sleep quicker, and they stay longer in the induced sleep in doses of artesunate. This result appears to clearly support the notion of possible sleep promoting properties of artesunate proposed in one earlier report.[17]

Chronic administrations in this study served for confirmatory tests employed to further examine the results in acute doses of the artesunate and *Garcinia kola* extract. The patterns of the induced sleep in chronic doses of *Garcinia kola* extract appeared confirmatory to the suggestion of possible opposing influence on sleep. This was explicit in the observed increases in the onset time of the induced sleep ($p \leq 0.05$, $p \leq 0.01$), with the decreases in the duration of the induced sleep. Thus, the observation in both the acute and chronic administrations in this study significantly demonstrated that *Garcinia kola* may have some clearly definable modulating properties of some negative influences on the central regulatory mechanism of sleep. On the other hand, the results of both acute and chronic administration of artesunate in this study were in support of the notion of possible facilitating influences of artesunate on some neural processes in the central regulatory mechanisms of sleep.

Most postulations on sleep and waking states focused on the biogenic amines as putative neurotransmitters in the central mechanism of sleep, and the brain levels of these biogenic amines have been determined in various brain regions in rat.[18] The principal catecholamines —epinephrine, norepinephrine and dopamine are all known to be present in the brain. There are also suggestions that dopamine play important role in wakefulness[19], and it has been demonstrated that compounds such as cocaine and amphetamine increase wakefulness and alertness by enhancing dopamine release[20]. Evidence is now accumulating from experimental animals that acetylcholine subserve the function of wakefulness and arousal and recent reports have shown that acetylcholine is released in high levels to maintain wakefulness.[21]

The notion of possible negative influences or opposing effects of *Garcinia kola* on central neural mechanism of sleep proposed in this study is consistent with the reported wakefulness function or alerting effects of acetylcholine and dopamine by earlier workers. It is therefore possible that the observed opposing influences of *Garcinia kola* on the induced sleep was mediated through dopaminergic mechanism or the acetylcholine transmitter system mechanism.

Considerable evidence suggests that serotonin is involved in the production of the initiating mechanism of sleep; the discharge of serotonin secreting neurons was linked with the onset of non-rapid-eye movement sleep, the first stage upon falling asleep. A number of workers have noted that sleep is initiated and maintained by the inhibitory mechanism of the neurotransmitter GABA or Galanin, through the inhibition of the arousal regions of the brain.[22,23]

The work of Seigel[24] demonstrated the specific GABA-receptor interaction mechanism in sleep production and it was hypothesised that $GABA_A$ receptors inhibit the neurons involved in wakefulness to promote sleep. It was reported that sodium thiopental can activate $GABA_A$ receptors even in the absence of gamma amino butyric acid thereby facilitating sleep production.[16] Although the neurotransmitter systems in sleep production is still a subject riddled with controversy as no specific roles or individual neurotransmitter system implicated in sleep production have been mapped out with certainty. Nevertheless, it is possible that the facilitating effect of artesunate on the induced sleep observed in this study was linked to the probability of the ability of artesunate to stimulate GABA receptors. It is suggested that the induced sleep promoting influences of artesunate which featured prominently in this study may have been mediated through the GABA transmitter neural mechanism in sleep production and this is a possibly tenable hypothesis. In the sleep model proposed by some workers, the ventrolateral preoptic nucleus (VLPO) of

the hypothalamus is considered to be the 'switch' factor in the initiation of sleep, and the area is said to use the inhibitory neurotransmitter Gamma Amino Butyric Acid (GABA) in the initiation of sleep by inhibiting the arousal regions of the brain.[25] The VLPO is also known to innervate and can inhibit the awake-promoting regions of the brain in the production of sleep.

CONCLUSIONS

GABA neural mechanism may account for the observed sleep promoting effects of artesunate. The dopaminergic neural activity may be the mechanism by which the observed opposing influences or the negative effects of *Garcinia kola* on induced sleep are initiated. It is suggested that more work be carried out on the normal sleep of rats to further elucidate the present observations of the effects doses of *Garcinia kola* and artesunate on thiopental-induced sleep in rats.

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