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# Effects of crude kerosene on testosterone levels, aggression and toxicity in rat

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#### ABSTRACT

The use of crude kerosene as a dietary supplement in boarding schools has been a common practice in east Africa and other countries for many years, with the belief of it reducing the sex drive (libido) at the pubertal stage. There is however no scientific basis for this belief. The present study aimed at using a rat animal model to investigate the effects of crude kerosene on serum testosterone levels, aggression and its possible toxic effects. Fifteen male albino rats of approximately similar age and average weights were put into three groups of five animals each; the control group (placebo), low kerosene dose  $(10 \,\mu l/day)$ group and high kerosene dose (300 µl/day) group. ELISA was used to determine the serum testosterone levels. During treatment, changes in aggression were observed and noted. Liver toxicity was determined using enzyme assays, total protein and albumin while renal toxicity was monitored using serum creatinine levels. A full hemogram was conducted to determine hematological effects. Various tissue biopsies were obtained and examined using histopathological techniques for evidence of toxicity. Contrary to the common belief, our findings showed an overall increase of serum testosterone levels of up to 66% in the low dose and 75% in the high dose groups, with an increasing trend by the end of the study. The high dose group showed significantly increased levels of white blood cells (WBC) (p = 0.036), red blood cells (RBC) (p = 0.025), hematocrit (HCT) (p = 0.03), red cell distribution width (p = 0.028) and platelets (p = 0.017). The histological results of the stomach indicated chronic gastritis.

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#### 1. Introduction

Kerosene is a distillate of crude petroleum that contains aliphatic, aromatic and a variety of other branched saturated and unsaturated hydrocarbons [1]. The use of crude kerosene has been a common practice in east Africa and other countries for many years, with the belief of it reducing the sex drive (libido) at the pubertal stage. In the course of daily meals consumption students are exposed to doses of kerosene as a dietary supplement, usually without their consent.

Abbreviations: ALP, alkaline phosphatase; ALT, alanine transaminase; AST, aspartate transaminase; EDTA, ethylenediaminetetraacetate; ELISA, enzyme linked immunosorbent assay; HCT, hematocrit concentration; LFT, liver function tests; RBC, red blood cells; RDW, red cell distribution width; RDW, red cell distribution width; RFT, renal function tests; T, testosterone; WBC, white blood cell.

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The process of puberty results in the release of some specific hormones which are primarily responsible for the development of secondary sex characteristics and for the emergence of reproductive capabilities in boys [2]. During this stage an increase in testosterone causes an increase in the sex drive (libido), enlargement of the reproductive organs such as the penis and testes, the production of sperm, increase of muscle mass and lowering of the voice, increased frequency of erection, and the growth of facial, chest, nipple and pubic hair among boys [3]. The link between testosterone (T) levels and the sexual drive was demonstrated in a study done using adolescent boys with the findings indicating that the adolescent boys who had higher levels T levels also reported higher levels of sexual activity (i.e. coitus) [4-7]. From the studies by Brooks-Gun and Halpern [5,6] it can be inferred that hormones may enhance feelings of sexual arousal in adolescents but how they act on those feelings is very much determined by multiple internal and external variables.

From the study conducted by Olweus et al. [4,8] it was noted that adolescent boys with higher T levels were more likely to engage in aggressive behavior. Under conditions of threat or unfair treatment, [9] they were shown to be aggressive. They further showed a link between higher T level and a lower tolerance for frustration. Further to these, they also observed that when no provoking situation occurred, T levels did not predict aggression. Various animal studies conducted on mice demonstrated the link between aggressive behavior and increased T levels [10,11]. In a study on mice exposed to jet kerosene continuously for 90 days, there was an observed increased incidence in the fighting of the test group mice [12].

There is increasing trend regarding the percentage of teenagers reporting sexual initiation at younger ages [13]. This early sexual initiation (before age 16) is likely to involve sexual risk-taking and expose young people to unwanted sex, sexually transmitted infections, and teenage pregnancy. This may be attributed to exposure to a highly sexualized media environment that may represent a primary source of sexual socialization [14,15]. Middlechildhood problems, poor parenting and peer pressure have also been shown to contribute to early sexual behavior [16,17]. Due to the dire consequences of early sexual activity [18], there have been efforts toward finding effective remedies to tame teenage sexual hyperactivity. In many Kenyan boarding schools, especially high schools, one such remedy that has been used traditionally is crude kerosene. In a recent survey that we conducted using structured questionnaires at a Public University admitting students from all over the country, (data not shown) we found out that 68% female and 76% male first year, random respondents from 28 of 47 counties in Kenya, reported that at least one of their main meals (lunch or dinner) was supplemented with kerosene on daily basis during their high school years. Interestingly, over 60% of respondents in the above category gave why they thought kerosene was included in their diets as being to reduce their desire for sex. The remainder (40%) did not know why it was added.

Kerosene is readily available and at fairly low costs throughout the country. The primary use is for lighting and in cooking stoves. Whether or not kerosene supplementation is effective in reducing libido has not been scientifically tested. Further, the dietary use of kerosene in schools to tame sexual drive occurs with little or no care at all on its possible hazardous effects on the health status these students. Although some information is currently available on the effect of dietary kerosene supplementation in animals and/or humans [12,19], such studies have failed to provide comprehensive information on effects on T levels, link to aggression and body tissue toxicity.

The present study was designed to monitor the effects on serum T levels, hematological, biochemical and histopathological changes in rats exposed to crude kerosene as a dietary supplement at doses that are comparable to those commonly used in Kenyan boarding schools.

#### 2. Materials and methods

#### 2.1. Ethics statement

All the animal protocols and experiments were approved by the Institution animal care and use committee of the University of Eldoret (Protocol No. UOE/001/14).

#### 2.2. Animals

Male Wistar rats (*Rattus norvegicus*) of approximately the same age (six weeks old) corresponding to early adolescent boys [20] and similar body weights were obtained from the University of Eldoret animal facility. They were acclimatized and given free access to water and standard rodent chow diet (Unga Farmcare East Africa Limited, Nakuru, Kenya) for two weeks prior to initiation of the experimental diet. The rats were housed and maintained at ambient temperature of 25 °C under a photoperiod of 12 h of light and 12 h of darkness. The animals were assorted into three groups of five rats each with all groups having similar average serum testosterone levels.

#### 2.3. Sample size determination

The sample size was determined according to the formula by Charan et al. [21].

E = total number of animals – total number of groups (if the value of E is between 10 and 20 then it is considered as an adequate, for this study, E = 12).

#### 2.4. Animal treatment

Animal groups consisted of the control (placebo group – distilled water) low dose ( $10 \mu$ l kerosene) and high dose ( $300 \mu$ l kerosene). All animals were maintained on regular rodent chow diet throughout the study. Kerosene (National Oil Corporation, Eldoret, Kenya) was delivered orally on a daily basis. Blood samples from animals in all groups (control and treatment) were collected from the tail under local anesthesia at baseline, day 7 and day 14. Since T levels in young male rats have been shown to vary with time of the day [22,23], all blood collections were done between 12.00 noon and 1.00 pm at all time points. Animals were also observed for changes in behavior on daily basis during

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