



RESEARCH ARTICLE

MICROWAVE RADIATION EXPOSURES AFFECT HEMATOLOGICAL PARAMETERS AND
ANTIOXIDANTS MODIFY THE EFFECTS IN RATS

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ABSTRACT

Purpose: The immediate effects of whole body exposure to 2.450 GHz microwave (MW) radiation on hematological parameters.

Materials and Methods: 140 adult Wistar rats were grouped into 7 of 20 rats each for the 7 hematological parameters studied. 5 from each group served as control, irradiated with vitamins C and E and without administration of vitamin. After exposures to 2.45 GHz MW, Hb, RBC, WBC, PCV, Platelet, Neutrophil and Lymphocyte counts were determined.

Results: Hb reduced from 16.5 to 10.0 g/dl immediately after exposure, and the normal value was recovered 8 weeks after. Vitamins C and E cushioned the effect and recovery of normal values attained within 2 weeks. RBC reduced from 50.0 to 41.0 x 10⁴ cells/mm³ immediately after exposure, and the normal value was recovered 2 weeks after. Vitamins C and E cushioned the effect and recovery of normal values within 24 hours. WBC reduced from 6880.0 to 4000.0 x 10⁶ cells/mm³ immediately after exposure, and the normal value was recovered a week after. Vitamins C and E cushioned the effect and there was no reduction at all in the values, rather there was an increase in each case. PCV reduced from 49.0% to 30.0% immediately after exposure, and the normal value was recovered 8 weeks after. Vitamins C and E cushioned the effect and recovery of normal values within 2 weeks. Platelet counts reduced from 300.0 x 10⁹/l to 210.0 x 10⁹/l immediately after exposure, and did not recover the normal value within the study period. Vitamins C and E cushioned the effect and recovery of normal values within a week. Neutrophil and Lymphocyte counts were not significantly affected.

Conclusion: Exposures to MW radiation affect the peripheral blood parameters that may have negative health impacts. Administration of vitamins C and E may cushion the potential deleterious health impacts.

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INTRODUCTION

The application of non-ionizing electromagnetic radiation in industry and medicine has increased greatly over the last few decades. It is well established that ionizing radiation at high intensities produces somatic and deterministic damages in biological tissues. This property is employed in Radiotherapy to destroy unwanted tumor growths. Much less is known however of the deleterious effects of non-ionizing radiations of which microwave (MW) is greatly used today. MW radiation is widely applied in modern technologies, especially in telecommunication industries, radar, weather, security, industrial heating and domestic appliances. Its modern medical applications include diathermy, MW tumor ablation, and hyperthermia in cancer management (Baillie 1988, McCrindle 2000). Studies by previous investigators showed that partial or whole body exposures to MW cause a variety of changes in biological systems, including the immune and hematopoietic systems. The radiation enhances or suppresses functions, depending on exposure conditions, animal species and the

radiation frequency (Novoselova 1999, Schwam 1983, Rao *et al.*, 1983, Pichard and Rusanbaum 1978). The heat associated with MW interactions has been reported to cause perturbations in biochemical reactions, increase reaction rates, current flow and the integrity of cell membranes (Pichard and Rusanbaum 1978). It has also been reported to promote the production of free radicals (Aweda 2003, Aiken and Dix 1981), cause hemolysis (Aweda *et al.*, 2004) and produce DNA single strand breaks (Aweda *et al.*, 2010, Lai *et al.*, 1995, Lai 1996) and circulatory failure due to oxidative stress (Kaln *et al.*, 2000). This paper presents, as part of a series of our studies on radiation safety and protection of MW at most commonly used frequency. This is to determine the effects of exposures on hematological parameters, using animal model. The results of this study will be essential in the assessment of the health impacts of environmental, professional, occupational and public exposures to the fast growing MW technologies. The knowledge and the awareness of the eventual health impacts would assist in developing protective devices and strategies of minimizing the hazards. Ascorbic acid is a water-soluble vitamin C that plays essential metabolic roles in-vivo (Sauberlich 1990). It is a good scavenger of

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reactive oxygen species (Padayatty and Levine 2002) and it helps to recycle α -tocopherol in-vivo. Vitamin E is a major membrane-bound antioxidant present in the lipid core of cell membrane and lipoprotein where it protects the polyunsaturated fatty, free radical-mediated peroxidation. It is also essential along with cholesterol, for the structural stability of the membrane. The physiological parameters in this study are due to oxidative processes, the effects of administering these anti-oxidants to MW-exposed rats were studied to determine their eventual modifying role.

MATERIALS AND METHODS

Calibration of Microwave Source

The MW generator model ER660E, Serial No MX704CCR from Toshiba UK Ltd available in the Department of Radiation Biology and Radiotherapy, College of Medicine of the University of Lagos was used for the study. The MW detector was the non-interacting thermistor RS 141, which has a resistance of 4.7 k Ω at 25 °C. The thermistor was pre-calibrated in a 12 cm x 6 cm x 4 cm size water phantom with the aid of a digital readout and a mercury-in-glass thermometer as reference. Details of the calibration procedures have been described elsewhere (Aweda *et al.*, 2003)

Determination of Specific Absorption Rate (SAR)

Determination of the SAR was done by inserting the thermistor probe into the rectum of the animal during exposure following an earlier methods of Guy *et al.*, (1984) with slight modifications to adapt to local requirements. The irradiation chamber surfaces were lagged with water to minimize the reflective properties which may increase the heating rate (Bren 1996). The generator was operated at room conditions of 25 \pm 2 °C and 56 \pm 4 % relative humidity. Exposures were total body with the animal at placed 12 cm from the MW antenna. The SAR values were obtained using the methods that have been described elsewhere (Aweda *et al.*, 2003).

Animal and Sample Preparations

140 Sprague Dawley rats of both sexes, 6-8 weeks old weighing 0.090 - 0.130 g, obtained from the Laboratory Animal Centre of the College of Medicine, University of Lagos were used. The care of the rats was in conformity with the International, National and Institutional guidelines for care and use of laboratory animals in Biomedical Research according to the Canadian Council of Animal Care (1985). The rats were fed with standard rat *chow* obtained from Pfizer Nig. Ltd, Ikeja, Lagos, Nigeria, and they had free access to drinking water. The animals were firstly acclimatized to the laboratory conditions for 5 days and maintained at the standard conditions of 12 h/12 h of dark/light cycle. The animals were grouped into 7, each group comprising of 20 rats. 5 of the 20 in each group served as control (without exposure to MW), while another 5 were exposed to MW radiation. Another group of 5 were administered with vitamin C before irradiation while the last group of 5 was administered with vitamin E.

Animal irradiation

The rats were irradiated from the open side of the rectangular horn-antenna, each placed in a plastic cage measuring 5 x 10 x 25 cm. The field intensity of the animal location was

determined with the aid of the thermistor connected to a readout meter developed by the research team. The field distribution in these regions was bell-shaped with a central maximum value of 6 mW.cm⁻² and a power density of 10 mWcm⁻², 5 cm from the centre. The rats were allowed free movements during exposures. The local variation of incident power density was measured in the presence and absence of the animal, this variation did not exceed 3 %, thus the total variation of power was between 5.5 and 6.0 mW.cm⁻².

Biochemical and haematological assay

Blood samples for the laboratory analysis were obtained from the rats by cutting the tales or by means of a sharp capillary tube inserted into the sclera via the conjunctiva under anaesthetized condition using ether. The hemoglobin (Hb), RBC, WBC, PCV, Platelet, Neutrophil and Lymphocyte counts were determined. PCV was determined using the Wintrobe Hematocrit method. The tube was filled with heparinized blood, making sure that no air bubble was trapped. The filled test tube was then spun for 30 min at 3000 G in a centrifuge. The volume of packed cell based on the total volume of the wintrobe capillary by the cells before being spun multiplied by 100 gave the hematocrit as follows:

$$\frac{\text{Packed cells}}{\text{Blood volume}} \times 100 = \text{Hematocrit.}$$

The Hb was obtained using the methods described by Eiler (1967). The antioxidants, vitamins C (ascorbic acid) and E (α -tocopherol) also from Pfizer Nig Ltd, Ikeja, Nigeria were administered intra-peritoneally at a dosage of 1 mg/kg body weight 30 min prior to exposures.

RESULTS AND DISCUSSION

Hb value reduced from the control mean value of about 16.2 g/dl to 10.0 g/dl immediately after irradiation and maintain this value till after 2 days when the value slightly increased to 10.0 g/dl. It then increased to 13.0 g/dl after 4 days and the control value was attained only at the 8th week (Figure 1). Hb value for the rats administered with vitamin C reduced from the control mean value to 11.2 g/dl immediately after irradiation and then increased to 12.0 g/dl after a day. This value increased more rapidly than in rats without vitamin C to attain 14.0 g/dl, 16.3 g/dl, 17.1 g/dl and 17.5 g/dl after a week, 2 weeks, 4 weeks and 8 weeks respectively. The effect of vitamin E was similar but a bit more than that of vitamin C. The value immediately after irradiation was 12.6 g/dl, 14.0 g/dl after 1 day and 16.0 g/dl, 17.0 g/dl, 17.5 g/dl and 17.5 g/dl after a week, 2 weeks, 4 weeks and 8 weeks respectively. RBC value reduced from the control mean value of about 50.0 x 10⁴ cells/mm³ to 41.0 x 10⁴ cells/mm³ immediately after irradiation. The value then increased to 45.0 x 10⁴ cells/mm³ after 1 day and then gently increased to 49.0 x 10⁴ cells/mm³, 51.0 x 10⁴ cells/mm³, 52.0 x 10⁴ cells/mm³ and 53.0 x 10⁴ cells/mm³ after 1, 2, 4 and 8 weeks respectively (Figure 2). The RBC value for the rats administered with vitamin C reduced from 53.0 x 10⁴ cells/mm³ to 48.0 x 10⁴ cells/mm³ immediately after irradiation and then increased to 52.0 x 10⁴ cells/mm³ after a day. This value increased more rapidly than in rats without vitamin C to stabilize at about 54.0 x 10⁴

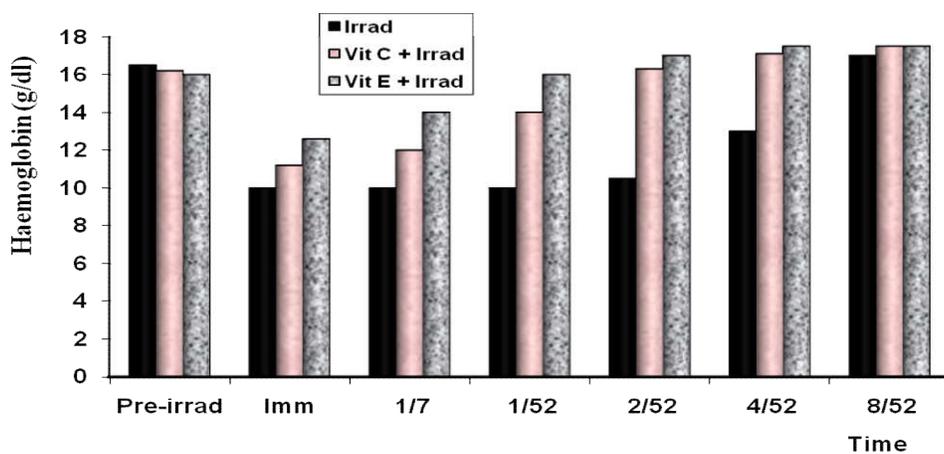


Figure 1: Variation of haemoglobin level over a period of 8 weeks after MW exposure and administration of vitamins C and E

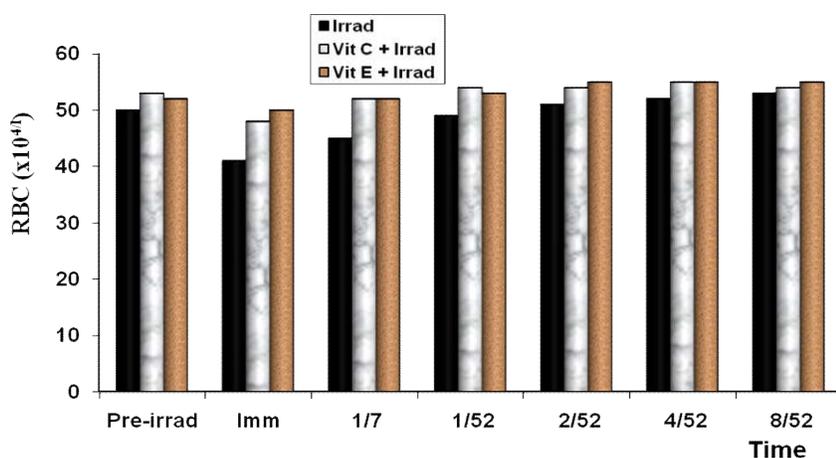


Figure 2: Variation of RBC counts over a period of 8 weeks after MW exposure and administration of vitamins C and E

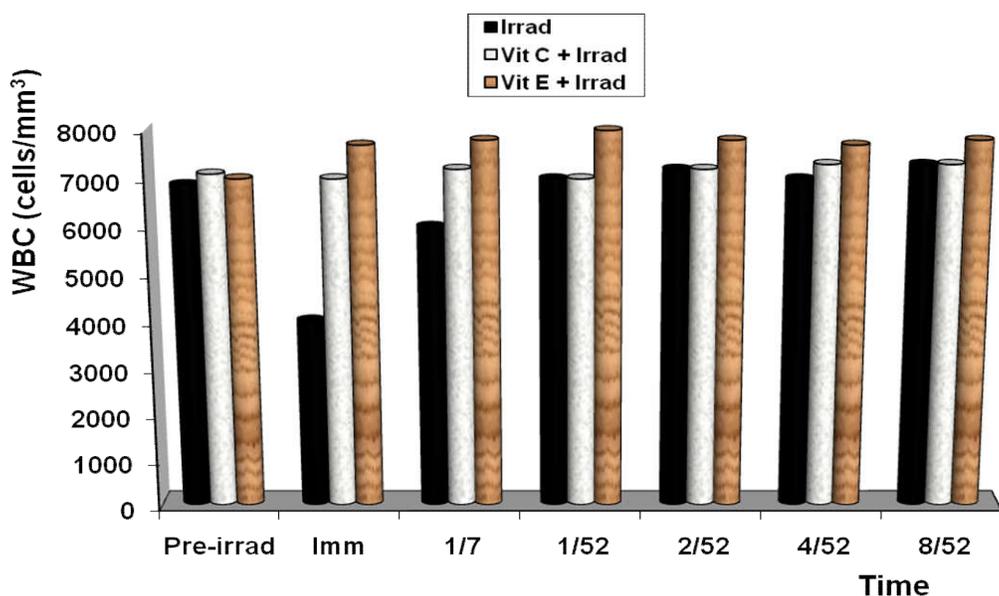


Figure 3: Variation of WBC counts over a period of 8 weeks after MW exposure and administration of vitamin C and E

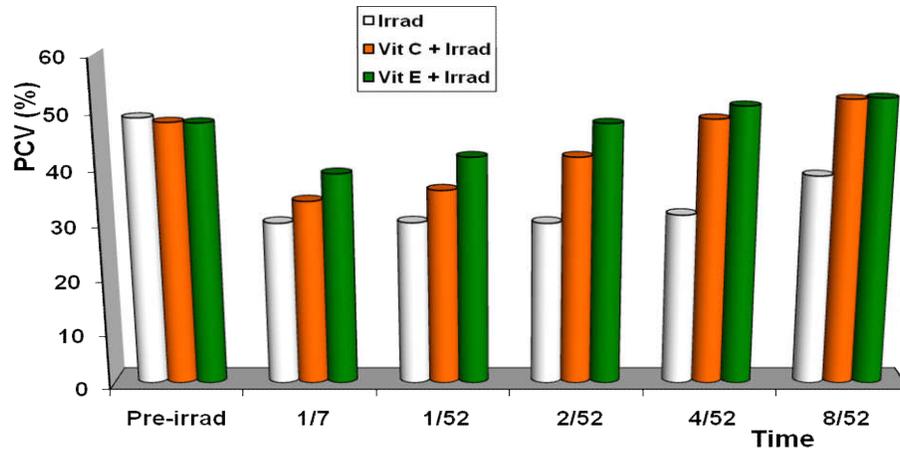


Figure 4: Variation of PCV over a period of 8 weeks after MW exposure and administration of vitamins C and E

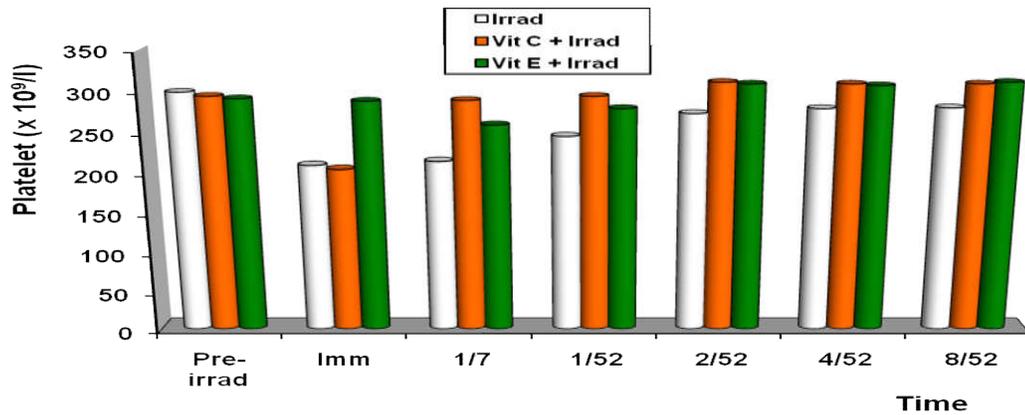


Figure 5: Variation of platelet counts over a period of 8 weeks after MW exposure and administration of vitamins C and E

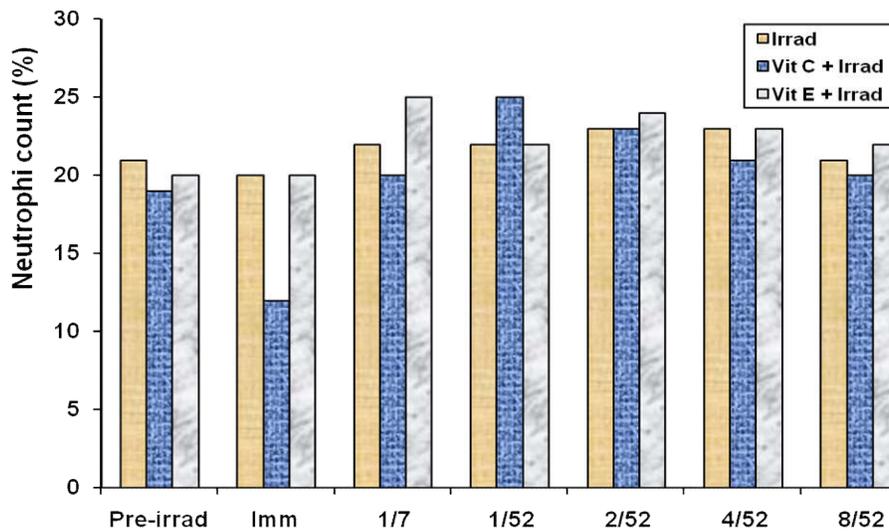


Figure 6: Variation of neutrophil counts over a period of 8 weeks after MW exposure and administration of vitamin C and E

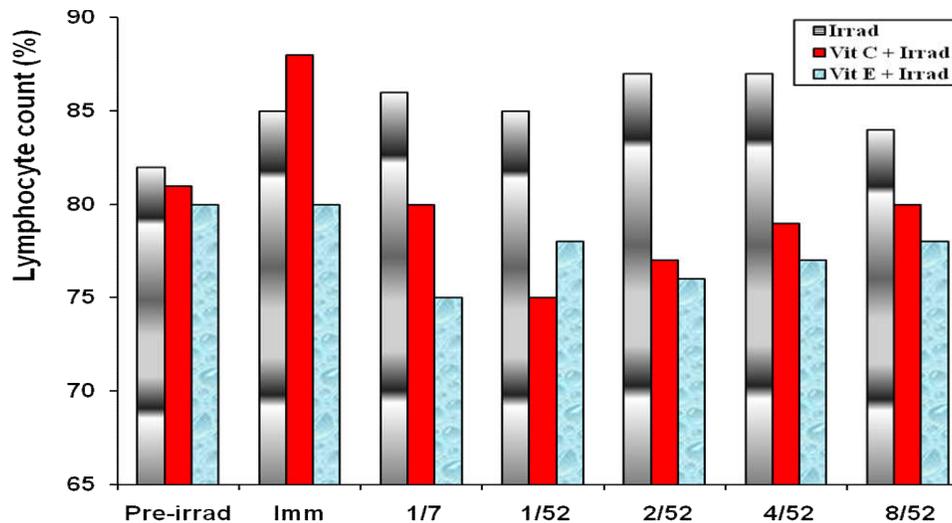


Figure 7: Variation of lymphocyte counts over a period of 8 weeks after MW exposure and administration of vitamins C and E

cells/mm³ after a week. The effect of vitamin E was also similar and a bit more pronounced than that of vitamin C. The value only slightly reduced from 52.0 x 10⁴ cells/mm³ immediately after irradiation. It thereafter rose to 52.0 x 10⁴ cells/mm³ and varied between 53.0 x 10⁴ cells/mm³ and 55.0 x 10⁴ cells/mm³ for the rest of the study period. WBC value reduced from the control mean value of about 6880.0 x 10⁶ cells/mm³ to 4000.0 x 10⁶ cells/mm³ immediately after irradiation.

The value then increased to 6000.0 x 10⁶ cells/mm³ after 1 day and then gently varied between 7000.0 x 10⁶ cells/mm³ and 7300.0 x 10⁶ cells/mm³ between the first and the 8th (Figure 3). The WBC value for the rats administered with vitamin C reduced slightly from 7100.0 x 10⁶ cells/mm³ to 7000.0 x 10⁶ cells/mm³ immediately after irradiation and then increased to 7200.0 x 10⁶ cells/mm³ after a day. The value became stabilized and only slightly varied between 7000.0 and 7300 x 10⁶ cells/mm³ for the rest of the study period. The effect of vitamin E was more pronounced than that of vitamin C as the value did not reduce but rapidly increased from 7000.0 x 10⁶ cells/mm³ to vary between 7700.0 x 10⁶ cells/mm³ and 8000.0 x 10⁶ cells/mm³ all through the study period. The PCV decreased significantly from the control value of 49.0% to 30.0% immediately after exposure. There was no significant increase in the value until after 4 weeks when it became 38.6% and it finally attained the value of 51.1% only after the 8th week. In the animals administered with vitamin C the value decreased immediately from 48.2% to 34.0%. It only gently increased with time from 36.0% after 1 day, to 42.0%, 48.8, 52.3% and 52.6% after 1, 2, 4 and 8 weeks respectively (Figure 4). Similar observation was noted with vitamin E but with greater effect; the rate of increase was higher and by the 2nd week, 51.1% value had been attained and thereafter, 52.5% till the end of the study period.

Platelet count reduced from the control value of 300.0 x 10⁹/l to 210.0 x 10⁹/l immediately after exposure. It then increased to 215.0 x 10⁹/l after a day and to 246.0 x 10⁹/l,

274.0 x 10⁹/l, 280.0 x 10⁹/l and 281.0 x 10⁹/l after 1, 2, 4 and 8 weeks respectively (Fig. 5). Variation in the values for the rats administered with vitamin C less; the value decreased from 295.0 x 10⁹/l to 205.0 x 10⁹/l immediately after irradiation and then increased to 290.0 x 10⁹/l after a day. The value thereafter varied between 295.0 x 10⁹/l and 312.0 x 10⁹/l for the rest of the study period. Similar trend was observed with vitamin E but with greater effect. The value decreased from 292.0 x 10⁹/l to 289.0 x 10⁹/l immediately after irradiation and then increased to 290.0 x 10⁹/l after a day. The value thereafter varied between 320.0 x 10⁹/l and 322.0 x 10⁹/l for the rest of the study period. Neutrophill value did not vary significantly as the effect of MW exposure merely reduced the value from the control of 21.0 % to 20.0 % immediately after (Figure 6). The value varied between 21 % and 23 % from 1 day through all the 8 week study period. Probably due to insignificant variation in the values of neutrophill, the effect of vitamins C and E were not pronounced. There was no significant difference in the effects of both vitamins; the values fluctuated between 20.0 % and 25.0 % in both cases all throughout the study period. Observations with lymphocyte counts are similar to those of neutrophill; MW radiation exposure apparently did not produce any significant effect on the status (Figure 7).

Consequently, administration of either of the vitamins did not produce significant effects and there was no noticeable difference between the effects of vitamins C and E. This study on hematological parameters Hb, RBC, WBC, PCV etc showed that MW exposures may have harmful effects on the peripheral blood cells as indicated by the fall in the values of the parameters. The antioxidant vitamins C and E administration however have a protective effect on the MW deleterious effects as it can be seen in the differences in the values between the pre-administration of the vitamins and control groups of animals exposed. The potential harmful action of the MW as observed in this study, is consistent with the findings of the previous reports by Yagi *et al.*, (1988) but not quite with those of Novoselova (1999). This can be understood from the fact that the later applied extremely low

MW intensities in their study as opposed to Yagi *et al.*, and in this study. However, the stimulatory effect of the antioxidant treatment agrees closely with the later findings. It was noted that the role of vitamin E was more significant than that of vitamin C. This can be understood from the fact that vitamin E is the most powerful antioxidant.

CONCLUSION

The results of this study demonstrate that exposures to MW radiation may affect the peripheral blood parameters. This may have negative health consequences especially among those that are professionally and occupationally exposure on regular basis. The fast growing MW technologies and diversification of industrial, domestic and medical applications make it indispensable for the generality of the unsuspecting public to know and be conscious of the potential health detriments of MW exposures. The results of the administration of vitamins C and E which cushion the undesirable effects suggest that, in addition to regular hematological examinations, diets fortified with effective antioxidants may be recommended for those who may not but be exposed to MW radiation during routine practice of their profession, discharge of occupation duties or other uses of MW generating devices.

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