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Study on Effect of Lycopene Rich Tomato Puree Supplementation on Wellbeing/Healthiness in Hyperlipidemic Wistar Rats

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ABSTRACT

Background: The red-colored tomatoes are characterised as a mine of lycopene to manufacture food grade ingredient and projected its placement in a specialty class where nutrient with pharmaceutical value justifies dietary status. Tomato lycopene has justified its status as a potential antioxidant compensating with nutraceutical importance and provided an option for endorsing specialty nutrition. The diversity epidemiological experiments recorded noteworthy modulations in lipid profile and projected risk reduction potential of lycopene against non-communicable diseases. The progressive research efforts to monitor haematological parameters to justify efficacy of tomato lycopene against adverse effects on wellbeing of human body is required for precision governance. The present research work aims to assess the potential of lycopene rich tomato puree in monitoring the haematological parameters in hyperlipidemic Wistar rats.

Methods: Wistar rats (Healthy control, high fat diet fed, HFD-Stain treated and HFD-tomato puree treated) were examined for physiological, haematological and blood biochemical parameters on 0, 14 and 28th day of experiment to assess effect of tomato puree administration on wellbeing of rats against healthy, hyperlipidemic and standard drug status.

Result: The increased haemoglobin and total erythrocyte count on 14th and 28th day of experiment justifies the erythropoiesis stimulatory effect of tomato puree lycopene. Moreover, increased total leukocyte count in high fat diet-tomato puree fed rats on 14th and 28th day validates the positive effect of tomato puree lycopene to monitor healthiness in Wistar rats. The regulatory mechanism based on haematological specialty features that monitors the risk of physiological disorders and diseases outlined through animal study experimental data provided a logistic option to confirm the efficacy of tomato puree lycopene to protect wellbeing/healthiness of live entity (Wistar rats).

Key words: Haematology, Haemoglobin, Lycopene, Tomato puree, Total erythrocyte count, Total leukocyte count, wellbeing.

INTRODUCTION

Phyto-bioactive compound rich source recognition of fruits and vegetables, based on variety components (polyphenols, carotenoids, phytonutrients and vitamins) and relative functional efficacy is projected as an option due admissible for processing technology to justify food ingredient integrity focusing on essential supplementary nutraceutical nature (Maya and Dubey, 2016 and Lauricella *et al*., 2017). The red tint tomatoes characterised as a rich commercial source of lycopene (Davis *et al*., 2003; Fadupin *et al*., 2012) for production of food grade ingredient under innovation and creativity. Moreover, significant dietary lycopene provision from tomato (around 85%) against rest of fruits and vegetables (15%) has featured tomato as viable source for scientific research (Levy and Sharoni, 2004; Chauhan *et al*., 2011). Lycopene, an extremely unsaturated, hydrocarbon chain carotenoid characterized as lipophilic molecule (Aghel *et al*., 2011; Basuny, 2012) projects its placement in a specialty class where nutrient with pharmaceutical value justifies dietary status. As a dietetic complement lycopene has already justified its potency as an antioxidant compound to compensate with nutraceutical importance and provided an option for endorsing specialty nutrition.

Epidemiological consistence revival of risk reduction potential against non-communicable diseases resting on regular consumption of tomato lycopene also supported its nutraceutical value status (Agarwal and Rao, 2000; Singh and Goyal, 2008; Kelkel *et al*., 2011; Kumar *et al*., 2017). Its cutting-edge administration through diet monitoring for diversity lipid profile projected noteworthy rise in HDL cholesterol and sudden drop in total cholesterol, serum triglycerides and LDL cholesterol (Kilany *et al*., 2020). The findings of the experiment are coiling around risk reduction potential of CVD and provided an option to locate the related health claim. The progressive research efforts coiling around cutting-edge monitoring of haematological parameters to justify mutual compatibility against adverse effects on wellbeing of human body is required for precision governance.

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Food processing technology commercialization flag ship 20th century middle era projecting on tomato products (tomato juice, puree, paste, ketchup *etc*.) was focusing on nutrition transitional goodness only. It has provided an option to proceed further out of nutrition box and promulgate health goodness based on its pharmaceutical importance. The lycopene base food formulations will act as a special purpose vehicle to stimulate innovative functional food development with specialty health claim. Lycopene in its sole form has already justified its importance as nutraceutical. The insufficiency of evidence on food product matrix base efficacy of lycopene in monitoring the haematological components under hyperlipidemic condition justify the need of said research work. Lycopene in the form of tomato puree matrix may alter the haematological parameters, that formed the aim of present study.

MATERIALS AND METHODS

Experimentation and analysis

The lycopene bioactive compound determined functionality of tomato puree was assessed by specially designed experimental setup of animals (Wistar rats) in collaboration with College of Veterinary and Animal Sciences, Parbhani (MAFSU, Nagpur), Maharashtra during 2020-2022. The Wistar rats (healthy control) were examined for physiological and haematological parameters against the hyperlipidemic group (high fat diet fed) and animals fed with standard drug (Atorvastatin) and lycopene rich tomato puree as per the Institutional Animal Ethical Committee approved experiment protocol (IAEC 58/19, Date 20/08/2019) obeying the guidelines of Committee for the purpose of control and supervision of experiments on animals.

Wistar rats other than the healthy control group were provided with high fat diet for about 28 days immediately after the completion of acclimatization period (20 days). Animals were constantly monitored for behavioural changes and eating habits during the induction. Body weight and growth of each animal was recorded throughout the experimental period. The healthy control and high fat diet fed rats were kept without treatment and defined doses (Table 1). The rats from group III and IV were treated with statin (Atorvastatin) as standard drug and tomato puree as lycopene treatment, respectively (Table 1). The blood collection of anesthetized rats was carried out from the inner canthus of eye on scheduled 0, 14th and 28th day of experiment and were used for separation of serum.

Physiological and behavioural examinations

The rats from all the four groups were closely monitored for physiological (body weight) and behavioural changes throughout the experimental period.

Hematological parameters

The blood samples of rats from all groups were analysed for haemoglobin by acid hematin method using Sahli's haemoglobinometer (Jain, 1986), total erythrocyte and total leukocytic count by haemocytometer (Sastry, 1989), differential leucocyte count by wright's staining (Weiss and Wardrop, 2010) on 0, 14th and 28th day of study.

Relative organ weight and histopathology

At the end of the experiment the organ of concern was examined for the gross observations and relative organ weight of the animals. Gross and histopathology architectural examination of the organs from sacrificed animals were undertaken on completion of the experiment.

RESULTS AND DISCUSSION

Health benefit alignment

The data showcasing physiological, haematological parameters of experimental rats in Fig 1 to 10 projected specialty features of lycopene as erythropoiesis stimulatory and organ health monitory based on administered tomato puree as a predominant resource.

Behavioural and physiological changes

The rats from all the groups were alert and healthy. They presented normal behaviour during entire tenure of experiment.

The data on overall body weight of treated (tomato puree fed) and control groups rats presented in Fig 1 highlighted noteworthy rise in body weights of high fat diet administered rats (Groups II, III and IV) on 14th and 28th day against 0 day. Moreover, laboratory diet rats (healthy control) also recorded substantial rise in body weight on

Table 1: Animal groups admissible for feeding and treatment.

Group	Treatment	Feeding and treatment
I	Healthy control	Fed on the normal laboratory pellet diet and *ad-libitum* water throughout the experimental tenure without any further treatment
II	High fat diet group	Fed on the high fat diet @ 10-15 g/rat/day throughout the experimental tenure with *ad-libitum* water
III	Statin treated group (Standard treatment)	Fed on high fat diet @ 10-15 g/rat/day and *ad-libitum* water throughout the experimental tenure followed by standard antihyperlipidemic treatment of Atorvastatin @ 0.5mg/kg *via* oral route dosing per day from 29th day of experiment. Vehicle used was distilled water.
IV	Tomato puree fed (Lycopene treatment)	Fed on high fat diet @ 10-15 g/animal/day and *ad-libitum* water throughout the experimental tenure followed by treatment of lycopene rich tomato puree @ 0.16ml per day per rat by oral route dosing in the vehicle of distilled water

14th and 28th day against 0 day. Relatively higher body weights of group II, III and IV rats against group I may be due to administration of high cholesterol diet to rats of said groups. The reliable remarks as body weight gain noted are supported by the reports presented by Bugajski *et al*. (2007).

Haematology-Haemoglobin

The data on haemoglobin (Hb) as a constituent element in RBC presented graphically in Fig 2, reflect on reduction in blood Hb concentration of healthy control rats on 14th and 28th day (13.31 to 13.87 g/dl) against 0 day (14.12 g/dl). Significant reduction in Hb level of rats from high fat diet group recorded on 14th day (12.50 g/dl) followed by marginal rise on 28th day (13.75 g/dl). However significant increase in Hb level of statin fed rats (15.06 and 15.25 g/dl) and tomato puree fed rats (17.18 and 17.43 g/dl) recorded on 14th and 28th day against 0 day. The increased Hb value as an effect of tomato puree lycopene (treatment group) and statin (standard drug) may be responsible for erythropoiesis as a stimulatory effect (Masajtis-Zagajewska and Nowicki, 2018; Eze *et al*., 2019).

Total erythrocyte count (TEC)

The data on TEC (million/cu mm) of rats showcased in Fig 3, projected significant reduction in group II (6.34 million/cu mm) and III (6.92 million/cu mm) rats against healthy control group (7.30 million/cu mm) on 14th day of the experiment. However, significant increase in TEC of rats from group IV (7.52 million/cu mm) recorded on 14th day followed by marginal decrease on 28th day (7.44 million/cu mm) may be associated with tomato puree treatment. The RBC analogous projection as erythrocyte count, as a specialty feature of physiological assessment, supported by both the groups of scientists (Ogundeji *et al*., 2013 and Eze *et al*., 2019) through stress and diabatic preventive action as a function of lycopene diet monitoring treatment of rats.

Total leukocyte count (TLC)

The TLC of rats from all the groups observed to be in the range of 9.19 to 10.20 thousand/cu mm (group I), 9.42 to 10.04 thousand/cu mm (group II), 9.16 to 10.38 thousand/cu mm (group III) and 8.66 to 10.15 thousand/cu mm (group IV) for experimental tenure of 28 days. The non-significant alterations in TLC of rats in group I (9.31 and 10.20 thousand/cu mm) and II (9.91 and 10.04 thousand /cu mm) recorded on 14th and 28th day of experiment against 0 day (Fig 4). The marginal increase in TLC of rats from group IV (9.61 and 10.15 thousand/cu mm) recorded on 14th and 28th day (Fig 4), may be associated with impact of lycopene. The relative TLC values of all the groups on 14th and 28th day were observed to be complementary to normal physiological limits. The similar type of changes in leukocyte count of lycopene supplemented Wistar rats and male Swiss mice are reported by Boeira *et al*. (2014), respectively.

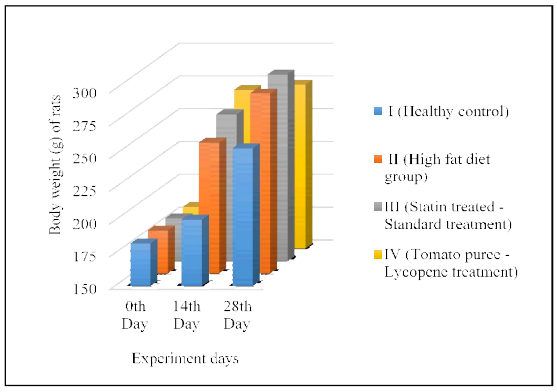

Fig 1: Body weight (g) changes in rats.

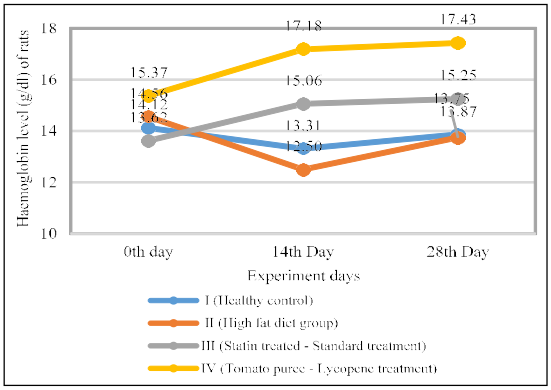

Fig 2: Haemoglobin (g/dl) changes in rats.

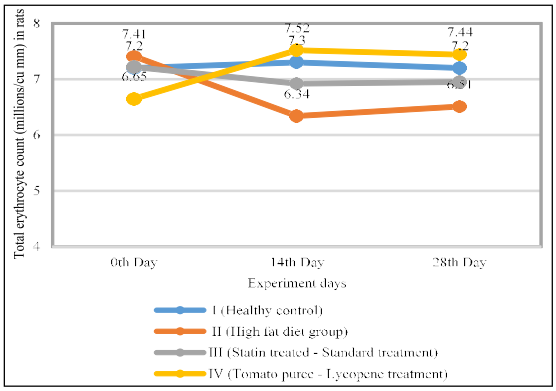

Fig 3: Total erythrocyte count (millions/cu mm) changes in rats.

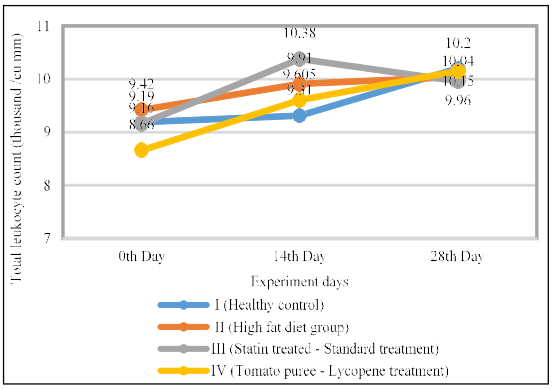

Fig 4: Total leukocyte count (thousand/cu mm) changes in rats.

Differential leucocyte count (DLC)

Neutrophil count

The higher neutrophil count of rats from groups II (38.50 and 38.25%), III (36.12 and 35.50%) and IV (36.13 and 35.37%) projected against healthy control group (group I-30.00 and 28.25%) on notified days (14th and 28th day) recorded comparatively higher drug statin and diet lycopene potential against positive control group with stability of high fat diet group (Fig 5). Moldovan *et al*. (2016) reported similar neutrophil count decreasing trend in diabetic Wistar rats as a diet supplementation effect of lycopene.

Lymphocyte count

The lymphocyte count of rats (Fig 6) from groups II (65.87%), III (64.50%) and IV (63.62%) on 28th day of treatment were observed to be at par against positive control group (61.25%). The lymphocyte count of rats from group IV (63.62%) though appeared to be higher than group I (61.25%) along with slightly lower but at par correlative with group II (65.87%) and III (64.50%) has underlined the effect of lycopene diet administration on monitoring the lymphocytes. The parallel trend of lymphocyte count transformation of diabetic Wistar rats supplemented with diet lycopene hypothesized by Moldovan *et al*. (2016) was appeared as data supportive research outcome.

Eosinophils count

The eosinophil count of rats from groups II (2.50%), III (3.62%) and IV (2.75%) on 28th day of treatment were observed to be at par against higher eosinophils in group I rats (4.12%). The non-significant increase in eosinophils count of rats from groups III (3.00 and 3.62%) and IV (1.75 and 2.75%) recorded on 14th and 28th day against their respective 0-day values projected the effect of lycopene and statin on monitoring the eosinophils (Fig 7). The similar type of alterations in eosinophil count of lycopene supplemented experimental animals (diabetic Wistar rats, high fat diet Sprauge Dawely rats and laying hens) were also reported by group of researchers (Moldovan *et al*., 2016, Kilany *et al*., 2020; Shevchenko *et al*., 2021).

Basophils count

The basophils count all the group rats was observed to be in the range of 1.37 to 4.25% (group I) 1.00 to 1.50% (group II), 1.25 to 1.75% (group III) and 1.50 to 2.00% (group IV) throughout the experiment tenure of 28 days (Fig 8). The non-significant alterations in basophil count of all the group rats (within and inter groups) recorded on notified experimental days (0, 14th and 28th day) projected non responsible effect of lycopene on high fat diet rats. The parallel trend of alterations in basophil count of lycopene supplemented male Sprauge Dawely rats and laying hens was also designated by Kilany *et al*. (2020) and Shevchenko *et al*. (2021).

Monocyte count

The monocyte count of rats from group III (3.62%) and IV

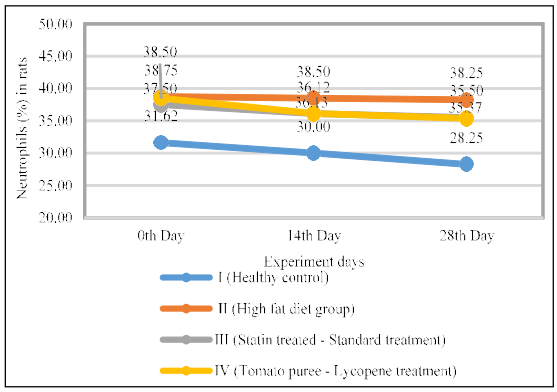

Fig 5: Neutrophil count (%) changes in rats Lymphocyte count.

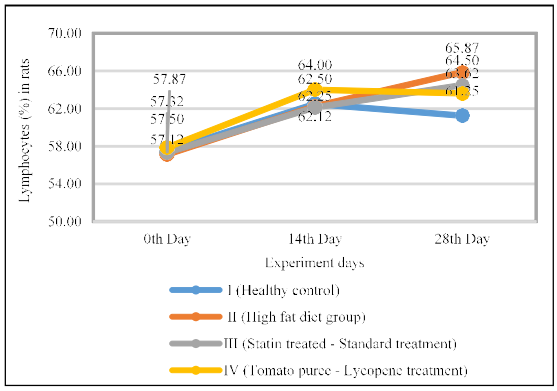

Fig 6: Lymphocyte count (%) changes in rats.

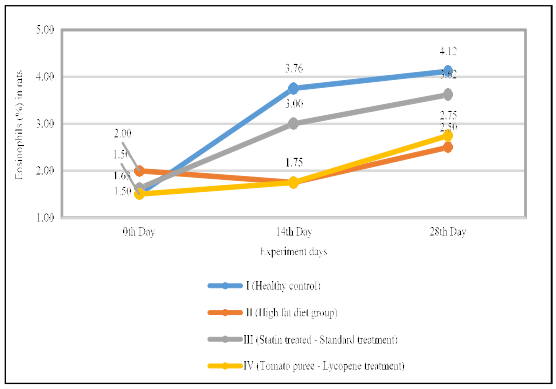

Fig 7: Eosinophil count (%) changes in rats.

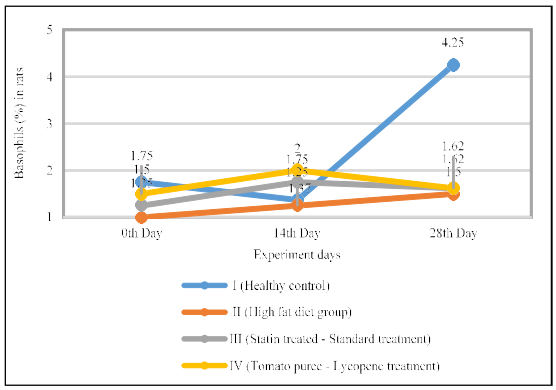

Fig 8: Basophil count (%) changes in rats.

(3.75%) on 28th day of treatment were observed to be at par against higher monocytes in group I rats (4.13%). The non-significant increase in monocyte count of rats from groups III (3.00 and 3.62%) and IV (3.00 and 3.75%) recorded on 14th and 28th day against their respective 0-day values (Fig 9) underlined the effect of lycopene and statin on monitoring the monocytes. The monocyte count of all the group rats on notified days (0, 14th and 28th day) were observed in the range of normal physiological limits. The similar trend of observations in monocyte count of experimental animals was also reported by earlier researchers (Moldovan *et al*., 2016; Kilany *et al*., 2020; Shevchenko *et al*., 2021).

Relative organ weight-Liver, kidney, and heart

The weight of liver, kidney, and heart of respective group of rats (group I to IV) were observed as 3.95, 0.92 and 0.36; 4.03, 0.93 and 0.33; 3.66, 0.91 and 0.39; 3.60, 0.89 and 0.44 g, respectively (Fig 10). No significant alterations in relative liver and kidney weight of rats were recorded in control and treatment groups. The similar (non-significant) alterations in weights of liver and kidney were also reported by Kilany *et al*. (2020) in experimental rats.

Histopathological assessment of liver, kidney and heart

Liver

The examination of liver histopathology of high fat diet fed rats has underlined mild to moderate, focal to multifocal congestion and dilatation of sinosidal spaces, indicating fatty and degenerative changes responsible for fatty diet caused illness. The liver samples of statin treated rats recorded improvement in histomorphology as compared to group IV. The histopathological changes of tomato puree treated rats exhibited focal degenerative changes and minimal focal fatty changes complementary to protective effect of lycopene treatment.

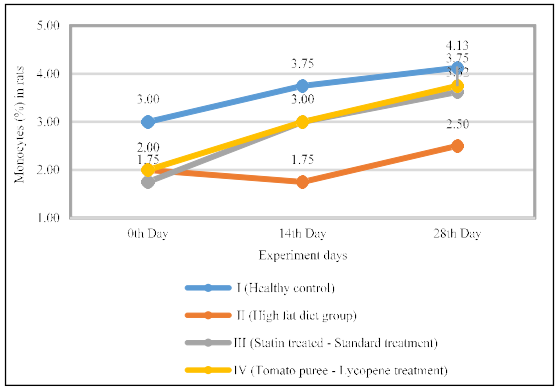

Fig 9: Monocyte count (%) changes in rats.

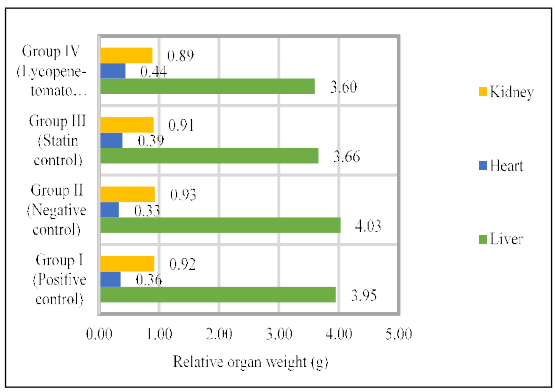

Fig 10: Relative organ weight (g) of experimental rats.

Kidney

The histopathological evaluation of kidneys of high fat diet fed rats has recorded mild to moderate degenerative changes, cystic degenerative changes, focal to multifocal congestion and mononuclear cell infiltration. The sections of kidney belonging to rats of tomato puree group did not show any specific change except, congestion and focal degenerative changes.

Heart

The heart sections of high fat diet fed rats projected minimal to mild congestion and mononuclear cell infiltration. However, no specific changes were recorded in histomorphology architecture of healthy control, statin treated and tomato puree fed rats.

CONCLUSION

The increased haemoglobin (Hb) level recorded on 14th and 28th day (17.18 and 17.43 g/dl) against 0-day (15.37g/dl) projected erythropoiesis stimulatory effect of lycopene in tomato puree fed rats. Increased TEC level high fat diet-tomato puree fed rats (7.44-7.52 million/cu mm) recorded on 14th and 28th day against healthy control, high fat diet and statin fed rats is also analogous to erythropoiesis stimulatory effect of tomato puree lycopene. Increased TLC level high fat diet-tomato puree fed rats recorded on 14th and 28th day against 0-day was observed to be complementary to project positive effect of tomato puree lycopene. The non-significant alterations in serum creatinine blood urea nitrogen levels of rats from healthy, HFD fed, statin treated and tomato puree fed groups recorded on notified days (0, 14th and 28th day) projected non responsible effect of treatment.

Histopathological assessment base changes in selective organs (heart, liver, and kidney) of tomato puree treated rats exhibited minimal degenerative changes as compared to high fat diet fed rats which is complementary to indicate protective effect of lycopene treatment. The haematological and blood biochemical specialty features monitoring the risk of physiological disorders and diseases of live entity (Wistar rats) outlined through animal study experimental data provided a logistic option to confirm the efficacy of tomato puree lycopene to protect wellbeing/ healthiness.

Conflict of interest: None.

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