ImmunoModulatory Activity of *Withania somnifera* and *Curcuma longa* in Animal Models: Modulation of Cytokines Functioning

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**ABSTRACT**

**Background and Objective:** The aim of the present study was to evaluate the immunomodulatory activity of *Withania somnifera* (WS) and *Curcuma longa* (C. longa) with their comparison and probable underlying cellular mechanism of action(s) in various animal models. **Materials and Methods:** Immunomodulatory activity of WS and C. longa was evaluated by use of pharmacological (Cyclophosphamide (CYP) induced immunosuppression, phagocytosis by carbon clearance, Delayed Type Hypersensitivity (DTH), haemagglutination titer), biological (thymus and spleen weight) and biochemical (estimation of cytokines TNF-α and IL-6) studies. **Results:** Pretreatment with WS and C. longa extract significantly increased total leukocytes during DTH, phagocytosis, HA titer, weight of thymus and spleen and prevention of edema formation in CYP induced immunosuppressed mice. Moreover, WS and C. longa treatment significantly increased the cytokines, TNF-α and IL-6 in both DTH and CYP induced immunosuppressed mice. **Conclusion:** The present experimental findings demonstrated that WS has superior immunomodulatory activity than C. longa. These effects presumably due to, greater ability of WS to boost the innate and adaptive immunity, the functioning arms of immune system.

**Key words:** *Curcuma longa*, *Withania somnifera*, immunomodulatory activity, TNF-α and IL-6

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**INTRODUCTION**

*Withania somnifera* is commonly called as ‘Ashwgandha’ in Indian traditional system of medicine and used as a tonic. It is traditionally used in the treatment of syphilis, as aphrodisiac, debility, dyspepsia, rheumatism and asthma and considered as an Indian Ginseng. Various pharmacological studies have been carried out and documented with *W. somnifera* viz., anti-inflammatory, anti-cancer, anti-stress, immunomodulatory, adaptogenic, central nervous system and cardioprotective activities. Effects of *Withania somnifera* on immune system was extensively studied. Its immunomodulatory activity was due to enhancement of total WBC count (17×125 cells: mm<sup>3</sup>) on 10th day, bone marrow cellularity (27×106 cells: Femur), increased α-esterase positive cell number (1800:4000 cells) and inhibition of delayed type hypersensitivity reaction on 10th day of treatment.

The cytokines role in immunomodulatory action of WS was demonstrated by Khan et al. they have demonstrated a dose-related potentiating IL-2 secretion by stimulated helper T cells (CD<sup>4</sup>+) and cytotoxic T-cells (CD<sup>8</sup>+). IL-2 promotes proliferation and differentiation of additional CD<sup>4</sup> cells, B cells and known to be a major mediator in immune reaction. It is reported that, increased expression of IL-2 is responsible for the enhanced IFN-γ expression. Activated macrophages are considered to be one of the important components of the host defense against tumor growth and the activation of macrophages are largely accomplished through the generation of cytokines, such as IL-1, TNF-α, IL-6 and IL-12 and suggested its participation in immunomodulatory activity. Furthermore, there was a significant increase CD4<sup>+</sup> and CD8<sup>+</sup> counts as compared to control and cyclosporin A treated groups, with a rapid recovery of CD4<sup>+</sup> T cells in immune suppressed animals. In addition, test treatment potentiated cellular and humoral immune responses in immune suppressed animals and was comparable to that of levamisole treated group. Such experimental findings clearly demonstrated selective Th<sub>1</sub> up-regulating activity of WS extract which further suggests its beneficial effect in the Th<sub>1</sub>/Th<sub>2</sub> modulation.
Curcuma longa (C. longa), called as 'Turmeric' in Ayurveda, is traditionally used in household as one of the ingredients of spice in food preparation. In Ayurvedic system of medicine, the rhizome of C. longa used as a stimulant, tonic, stomachic and carminative. Curcumin, an active constituent of C. longa found to inhibit matrix metalloproteinase (MMP-3) and MMP-13 gene expression by inhibiting the c-Jun-N-terminal kinase (JNK), activation protein-1 (AP-1), nuclear factor kappa B (NF-kB) pathways in human chondrocytes\(^\text{19}\), inhibits IL-8 production, monocyte inflammatory protein-1 (MIP-1 a), monocyte chemotactic protein-1 (MCP-1), IL-1β, TNF-α, 4-b-phorbol-12-b-myristate-13 a acetate (PMA) or lipo-polysaccharide(LPS), stimulated monocytes and macrophages\(^\text{20}\). Curcumin is also known to activate and regulate dendritic cells, inhibit IL-1, IL-6 and TNF-α, cell proliferation, along with inhibition of NF-kB activation\(^\text{21,22}\), acts as anti-inflammatory, cardio and hepato-protective through its antioxidant property in rats\(^\text{23}\). Therefore, with such background of information, the present study was undertaken to evaluate the immunomodulatory activity of WS and C. longa, their comparison and probable underlying mechanism of action(s) by studying the various pharmacological and biochemical paradigms in various experimental animal models.

MATERIALS AND METHODS

Animals: Swiss albino mice (20-25 g) of either sex were procured from Haffkin Institute of Biopharmaceutical Sciences, Mumbai, India. Animals were maintained in animal house at desired conditions: Temperature of 23±1°C and relative humidity 50±5%. The animals were provided with standard laboratory diet (Amrut Laboratory Animal Feed, Nava Maharashtra Chakan Oil Mills and Pune, India) and water ad libitum. The animals were shifted from animal house to laboratory 2 h prior to experiments.

Institutional animal ethics committee approval: Experimental protocol was reviewed and approved by Institutional Animal Ethics Committee (IAEC) constituted under the Committee for Purpose of Control and Supervision of Experiments on Animals (CPCSEA), approval no. SIPS/IAEC/App/2011-12/16, Animal house registration with Govt. of India (962/c/06/CPCSEA) dated 27 July 2006.

Drugs and chemicals: Cyclophosphamide (Endoxan 500 mg) obtained from Zydxus Onco Sciences, Ahmedabad, India; Sheep red blood cells antigen (Serum institute of India, Pune, India), Purified and standard extract of WS, obtained from Natural Remedies Pvt Ltd, Bangalore, India Charak Pharmaceutical Pvt Ltd. Mumbai, India and C. longa obtained as a gift sample from. Standard cytokines kits viz TNF-α and IL-6 were purchased from Woborn, MA, USA.

HPTLC studies of WS and C. longa: The 150 mg of WS and 200 mg of C. longa were dissolved (using ultrasonicator) in 15 mL of methanol separately and samples of 5, 10 and 20 mL were applied as 8 mm wide bands, using Camag Linomat V automatic sample applicator. Samples were applied with a 100 mL syringe (Hamilton, Bonaduz, Switzerland) at a constant rate of 0.1 mL sec\(^{-1}\) and the distance between adjacent bands was 15 mm. The 10×10 cm aluminum backed HPTLC plates coated with 250 mm layers of silica gel G 60F254 (Merck India, India) were prewashed with methanol and activated at 110°C for 10 min and used as stationary phase. The plates were developed in an ascending manner with solvent system for WS consisting of ethyl acetate: Toluene: Acetic acid in a ratio of 9:1:1:0.6 and for C. longa Toluene: Ethyl acetate: Formic acid in a ratio of 5:1.5:0.5. HPTLC Plates of WS and C. longa were scanned at 214 and 254, respectively using scanner-4 (CAMAG) operated in reflectance absorbance, fluorescence and white light mode and controlled by Win CATS software (Version 1.4.3). The sources of radiation used were deuterium lamp (200-400 nm), mercury lamp (200-400 nm) and tungsten (400-800 nm) emitting continuous UV and fluorescence spectra. The plates were derivatised by spraying anisaldehyde sulphuric acid and heated (110°C for 5 min). The Rf values and the colors of the bands resolved were recorded.

Cyclophosphamide induced immunosuppression in mice: Mice were divided into various groups (n = 6), Group I: Received gum acacia (1% w/v, 10 mL kg\(^{-1}\)) orally (p.o.), Group II: Received Cyclophosphamide (CYP) 30 mg kg\(^{-1}\), intraperitoneally (i.p.) for 10 days, Group III-IV: Received WS 200 and 400 mg kg\(^{-1}\), orally daily for 14 days and CYP (30 mg kg\(^{-1}\), i.p.) for 10 days, respectively. Group V-VII: Received C. longa 40, C. longa 60 mg kg\(^{-1}\) and WS 200+C. longa 40, respectively orally daily for 14 days and CYP (30 mg kg\(^{-1}\), i.p.) for 10 days, respectively. On day 14, blood was collected by retro-orbital plexus under mild ether anaesthesia. Total WBCs and Hb (hemoglobin) were determined in laboratory as per the method of Ziauddin et al.\(^\text{24}\).
Evaluation of phagocytosis by carbon clearance in mice: Mice of either sex were divided into various groups, Group I: received gum acacia (1% w/v, 10 mL kg⁻¹) orally (p.o.), Group II and III: Received WS 200 and 400 mg kg⁻¹, p.o., respectively daily for 7 days, Group V, VI and VII: Received C. longa 40, C. longa 60 mg kg⁻¹ and WS 200+C. longa 40, respectively orally daily for 7 days. Twenty four hour after the last treatment, all groups were injected with 0.1 mL (1% w/v) carbon suspension by intravenous route. For assessment of phagocytosis, blood samples were collected from by retro-orbital plexus under light ether anaesthesia, at 5 and 15 min after the administration of carbon suspension. The phagocytic clearance rate was calculated as per the method of Hudson and Hay²⁵ by the equation mentioned as follows:

\[
K = \frac{\log (\text{OD}_5) - \log (\text{OD}_15)}{t_5 - t_{15}} \times 100
\]

whereas, \(\log (\text{OD}_5)\) mean absorption at 5 min, \(\log (\text{OD}_15)\) mean absorption at 15 min, \(t_5\) is absorption time at \(t_5\), \(t_{15}\) is absorption time at \(t_{15}\).

Delayed Type Hypersensitivity (DTH) in mice: Mice of either sex were divided into various groups (n = 6), Group I: Received gum acacia (1% w/v, 10 mL kg⁻¹) orally (p.o.), for 11 days, Group II: Received CYP 30 mg kg⁻¹, intraperitoneally (i.p.) on day 6, Group III and IV: Received WS 200 and 400 mg kg⁻¹, p.o. for 11 days and CYP (30 mg kg⁻¹, i.p.), respectively on day 6, Group V, VI and VII: Received C. longa 40, C. longa 60 mg kg⁻¹ and WS 200+C. longa 40, respectively orally daily for 11 days and CYP (30 mg kg⁻¹, i.p.) on day 6. To all groups, SRBCs was injected (1×10⁶ cells/mouse) by i.p., 2 h after the CYP administration and on 11th day into right hind paw (50 µL). The DTH reaction was determined by measuring the paw edema as a parameter by volume displacement method using plethysmometer (Orchid Scientific, India) and results were expressed as percentage (%) of paw edema formation.

Heamagglutination titer: Mice of either sex were divided into various groups (n = 6), Group I: Received gum acacia (1% w/v, 10 mL kg⁻¹) orally (p.o.), for 6 days; Group II: Received CYP 30 mg kg⁻¹, intraperitoneally (i.p.) on day 6; Group III and IV: Received WS 200 and 400 mg kg⁻¹, orally daily for 6 days, respectively and CYP (30 mg kg⁻¹, i.p.) on day 6, Group V, VI and VII: Received C. longa 40, C. longa 60 mg kg⁻¹ and WS 200+C. longa 40 orally daily for 6 days, respectively and CYP (30 mg kg⁻¹, i.p.) on day 6. On day 7, blood was collected by retro-orbital plexus under anaesthesia and serum was separated. Two fold dilution of serum was done in 0.15 M Phosphate Buffer Solution (PBS) and 25 µL of each dilution was aliquoted in 96 well microtitre plates (Tarsons, India). To this, a freshly prepared SRBCs suspension (25 µL) in PBS was dispensed in to each well and mixed thoroughly. The plates were incubated at 37±0.5°C for 1 h and examined for button formation and Haemagglutination titer was calculated²⁵.

Effects of WS and C. longa on lymphoid organ and Total Leukocyte Counts (TLC): Animals were divided in to various groups (n = 6) and WS and C. longa were administered daily for 21 days on 22nd day, blood was collected for TLC determination and animals were sacrificed under ether anaesthesia. Spleen and thymus were isolated and their weights were recorded.

Measurement of TNF-α and IL-6: The cytokines viz TNF-α and IL-6 were measured in two experimental condition (a) CYP induced immunosuppression and (b) Delayed type hypersensitivity reactions using ELISA reagent kits (Woborn, MA, USA) following manufacturer's instructions by using ELISA reader (Biotek, Germany) at pharmacology laboratory.

Statistical analysis: Experimental results were presented as Mean±SEM and analyzed by using one-way ANOVA followed by Dunnett’s test. A value p<0.05 was considered as statistically significant.

RESULTS

HPTLC studies: The WS and C. longa showed the presence of 7 and 6 components, respectively on HPTLC chromatogram at 214 nm for WS and 254 nm for C. longa (Fig. 1 and 2). HPTLC chromatogram of WS samples (Fig. 1a) exhibited violet (visible) bands at the R₅ value 0.56 similar to that of standard withaferin A (R₅ = 0.56). C. longa chromatogram showing orange-violet bands at the R₅ value 0.28 similar to curcumin standard R₅ value 0.28 (Fig. 2a). Therefore, the chromatogram finger print clearly indicates the presence of withaferin A and curcumin.

Effects of WS and C. longa on cyclophosphamide induced immunosuppression in mice: Mice treated with CYP induced immunosuppression reflected in significant (p<0.001) decrement in total leukocyte counts (Fig. 3). Pretreatment with WS improved the
TLC significantly (p<0.05) on 14th day compared to CYP treated immunosuppressed mice. Further, the combined treatment of WS 200+C. longa 40 restored significant (p<0.05) the TLC compared to WS 200 treated animals (Fig. 3).

**Effects of WS and C. longa on phagocytosis activity in mice by carbon clearance:** The significant (p<0.001) increased carbon clearance rate (K) in mice treated with WS (40 and 60 mg kg⁻¹) compared to vehicle treated mice was observed; whereas, C. longa pretreatment did not elicit significant effect on phagocytosis (Fig. 4). Combined treatment of WS 200+C. longa 40 elicited greater carbon clearance than WS 200 (Fig. 4).

**Effects of WS and C. longa on Delayed Type Hypersensitivity (DTH) in mice:** SRBC sensitized mice when challenged with same antigen elicit paw edema formation which is an index of DTH reactions. The animals treated with CYP induced a significant (p<0.001) increase in edema formation compared to vehicle treated control animals. Pretreatment with WS and C. longa prevented the edema formation significantly (p<0.001) at various time intervals compared to CYP treated mice. The prevention of edema formation with WS was comparatively higher than that of C. longa (Fig. 5). Furthermore, the combined treatment of WS 200+C. longa 40 prevented the edema formation and the results were comparable to that of WS 400 treated animals (Fig. 5).
**Effects of WS and *C. longa* on heamagglutination titer in mice:** Mice treated with CYP showed significant reduction in HA titer compared to vehicle treated animals. Pretreatment of WS elicited significant rise in HA titer than *C. longa* compared to CYP treated mice. The animals treated with WS 200+*C. longa* 40 significantly increased the HA titer and was equivalent to WS 400 treated group (Fig. 6).

**Effects of WS and *C. longa* on lymphoid organ and TLC:** Treatment of WS+*C. longa* extract for 21 days significantly increased TLC count, thymus and spleen weight compared to vehicle treated mice. The extent of increase in TLC count, thymus and spleen weight were higher with the treatment of WS than *C. longa*. Further, the combined treatment WS 200+*C. longa* 40 elevated the weight of thymus, spleen and the TLC count. The effect of combined treatment was comparable to that of WS 400 treated animals (Table 1).

**Effects of WS and *C. longa* on cytokines (TNF-α and IL-6):** Pretreatment of WS and *C. longa* increased the cytokines, TNF-α and IL-6 due to DTH reaction. Further, in CYP induced immunosuppression, there was
Fig. 5: Effects of WS and C. longa on paw edema formation in CYP induced immunosuppression in mice ** p<0.01 when compared to vehicle treated group, *p<0.05 when compared to CYP treated group, † p<0.05 when compared to C. longa 60 treated group.

Fig. 6: Effects of WS and C. longa on HA titer * p<0.05 when compared to vehicle treated group, ** p<0.01, * p<0.05, when compared to CYP treated group.

Table 1: Effect of WS and Curcuma longa on lymphoid organ and TLC

<table>
<thead>
<tr>
<th>Treatment and dose (mg kg⁻¹ p.o)</th>
<th>Thymus</th>
<th>Spleen</th>
<th>TLC count (cell/cu mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vehicle control (1 mL/100 g)</td>
<td>0.15±0.001</td>
<td>0.35±0.02</td>
<td>6965.0±700.3</td>
</tr>
<tr>
<td>WS 200</td>
<td>0.30±0.03*</td>
<td>0.70±0.004*</td>
<td>8845.8±910.2*</td>
</tr>
<tr>
<td>WS 400</td>
<td>0.50±0.03**†</td>
<td>0.70±0.05*‡</td>
<td>9429.4±662*†</td>
</tr>
<tr>
<td>C. longa 40</td>
<td>0.20±0.02*</td>
<td>0.30±0.04*</td>
<td>7158.4±83.61</td>
</tr>
<tr>
<td>C. longa 60</td>
<td>0.30±0.04**</td>
<td>0.40±0.05*</td>
<td>7819.2±678.1</td>
</tr>
<tr>
<td>WS 200+C. longa 40</td>
<td>0.50±0.05**‡</td>
<td>0.70±0.02*</td>
<td>9540.0±524*</td>
</tr>
</tbody>
</table>

* p<0.05, ** p<0.01 when compared to vehicle treated group, † p<0.05 when compared to C. longa 60 treated group, ‡ p<0.05 when compared to WS 200, values are taken as Mean±SEM (n = 6)

a significant (p<0.05) reduction in both TNF-α and IL-6 compared to vehicle treated mice. Pretreatment of WS significantly (p<0.05) elevated TNF-α and IL-6.

The combined treatment restored (p<0.05) TNF-α and IL-6 due to CYP and the effect was greater than WS 200 treated group (Table 2).
Table 2: Effects of WS and Cuminum longa on TNF-α and IL-6 in delayed type hypersensitivity and CYP-induced immunosuppression in mice

<table>
<thead>
<tr>
<th>Treatment and dose</th>
<th>TNF-α (pg mL⁻¹)</th>
<th>IL-6 (pg mL⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>During delayed type hypersensitivity</td>
<td>CYP-induced immunosuppression</td>
</tr>
<tr>
<td>Vehicle control</td>
<td>20.1±1.3</td>
<td>10.6±1.7</td>
</tr>
<tr>
<td>CYP (30, i.p.)</td>
<td>9.9±1.6</td>
<td>4.4±0.8</td>
</tr>
<tr>
<td>WS 200</td>
<td>14.4±1.6</td>
<td>7.1±0.6</td>
</tr>
<tr>
<td>WS 400</td>
<td>18.8±2.2**</td>
<td>9.5±1.2</td>
</tr>
<tr>
<td>C. longa 40</td>
<td>10.9±1.8</td>
<td>5.1±0.7</td>
</tr>
<tr>
<td>C. longa 60</td>
<td>11.2±1.3*</td>
<td>6.2±0.4</td>
</tr>
<tr>
<td>WS 200+C. longa 40</td>
<td>18.2±2.1**</td>
<td>9.2±0.5</td>
</tr>
</tbody>
</table>

**p<0.01 when compared to vehicle treated group. *p<0.05, **p<0.01 when compared CYP treated group, †p<0.05 when compared to C. longa 60 treated group, ‡p<0.05 when compared to WS 200, values are taken as Mean±SEM (n = 6)

DISCUSSION

In the present experiments, the immunomodulatory activity of WS + C. longa and their per se treatment in animal models, WS elicited a dose dependent reversal of the depletion of proliferation of immune cells viz leukocytes. Pretreatment with WS and C. longa reversed the regressed organ weight due to CYP (thymus and spleen) on 14th day. Reversal of suppressed immune system with WS and C. longa may be due to (a) Differentiation of stem cells, (b) Stimulation of production of immune cells and (c) Stimulation of hematopoietic system. Such cellular events collectively lead to reversal of CYP induced immune suppression in mice. The documented report reveals the inhibition of natural killer cells (NK cells) results into amelioration of cytotoxicity, reactive oxygen species, nitric oxide and cytokines production with C. longa treatment. It is believed that, similar effects may also be responsible for the reversal of CYP induced immune suppression following WS treatment. Therefore, aforementioned cellular and molecular that effects with treatment of WS and C. longa may be responsible for immunomodulatory activity observed in the present study.

In another set of experiments, the effect of WS and C. longa extract was studied on phagocyte mediated clearance rate (a test indicate antigen clearance phagocytosis) of circulating mononuclear phagocytic cells, leukocyte, neutrophils and tissue macrophages which are responsible for phagocytosis. The experimental findings clearly indicate the WS and C. longa treatment enhanced phagocytosis which is considered to be an index of improved functioning of the macrophage-phagocyte in the reticuloendothelium system.

In order to understand the possible mechanism(s) of action at cellular level, the effects of WS and C. longa were studied on cell mediated host defense system by Delayed Type Hypersensitivity reactions (DTH) in mice model, since it is predominantly T-cell mediated immune reactions. DTH response is triggered by IFN-γ produced by CD₄⁺, a Th1 (thymus derived helper cell) cells or CD₈⁺ cells. These cells take at least 24-72 h for the induction as well as T-cells activation which subsequently recruits monocytes and lymphocytes to the desired site for enhanced immune responses. Furthermore, DTH response is known to be initiated by interactions between antigen specific T-cells and antigen, causing the secretion of lymphokines which affects immune cells, especially macrophages. The DTH response largely represents the enhancement of lymphoproliferative events. In these experimental findings too, reduction of DTH response with the treatment of WS and C. longa, suggesting the increased proliferation and differentiation of Th₁ cells. Such cellular events finally results in increased production of cytokines viz. IL-2, IL-6, IL-12, IFN-γ and TNF-α. It is established that, IL-12 and TNF-α plays a vital role in both innate and adaptive immunity, therefore, it is worthwhile to mention here that enhanced production of TNF-α and Nitric Oxide (NO) are vital component of immune system which are participating in the protection of intracellular infections. Hence, it is likely that both WS and C. longa treatment elicit significant immunomodulatory activity.

In addition, the effects of WS and C. longa on antibody mediated humoral immune response were studied by determining antigen-antibody HA titers. Scientific evidence suggests that, there is an increased immunoglobulin formation which is considered to be an indicator of primary effector function of B-cells. The augmentation of humoral response to SRBCs with the treatment of WS and C. longa was manifested in the form of increased HA titers. Such improved HA response can be explained by taking into account that IgM is more effective than IgG in agglutinating red blood cells. The increased HA titers with the treatment of WS and C. longa likely to prove an important paradigm for evaluating improved immune reactions, that might have
been achieved through the activation of lymphoid cells. Thus, present experimental findings with DTH and HA titer clearly demonstrated that the treatment of WS and C. longa enhanced the proliferation of T cell and B-lymphocytes, ultimately leading to improvement of both the arms of immunity (innate and adaptive immunity).

Herbal drugs entitled as “Rasayana” are known to possess immunomodulatory properties and known to act by stimulating both specific and non-specific immunity. Both WS and C. longa were shown to stimulate humoral and cellular immunity which are the two cellular components of adaptive immunity. In DTH, antibody formation against the antigen, SRBCs requires co-operation of immune cells derived from T, B lymphocytes and macrophages. The short lived suppressor T-cells population is damaged by CYP which is the principal mechanism of action for the induction of DTH reaction. Furthermore, SRBCs cause mature T-cells to differentiate into 3 distinct functioning subsets, discriminated according to the array of cytokines they produce: Th1 cells secrete IL-2, IFN-γ, and tumor growth factor β; Th2 cells produce IL-4, IL-5, and IL-6. Th1 cells have the capacity to produce both Th1 and Th2 cytokines, since they represent a stage of differentiation prior to their commitment to the Th1 or Th2 lineage. In context to the aforementioned events, macrophages are responsible for secretion of IL-1, IL-6, IFN-γ, and TNF-α upon their activation. TNF-α is a principal mediator of acute inflammation inducing cytokines release following CYP administration. The level of TNF-α in the serum of both immune suppressed and SRBCs challenged rats were found to be reduced with the WS treatment which is contradictory to the above statement. Since, such findings have been reported which may be explained on the basis of biphasic action of TNF-α in immune modulation process. The rats treated with C. longa also showed a decreased TNF-α level which may also be attributed to similar action. The level of IL-6 in serum was decreased in rats treated with WS and C. longa compared to CYP induced immune suppressed and SRBCs challenged mice.

CONCLUSION

The present experimental findings demonstrate that WS possesses superior immunomodulatory activity mainly through improving the cellular mediators (cytokines) compared to C. longa. Such effects presumably due to, greater ability of WS to boost up the innate and adaptive immunity, the functioning arms of immune system. Further, it is confirmed that the combined effect of WS + C. longa produced a greater effect than per se effect, this may be due to improved activation of immune system and other associated cellular events.

Additional studies are required to explore the beneficial therapeutic implication of WS and C. longa either alone or in combination as immunomodulatory agents in clinical medicine for the treatment of immune related diseases/disorders viz., AIDS, HIV, cancer, tuberculosis, leprosy etc.

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