

The effects of intellan and cytacon on hematological and biochemical parameter in rabbits: A comparative study

Humera Anser², Raheela Najam¹, Saira Saeed Khan¹, Bushra Riaz² and Sana Sarfaraz²

¹Department of Pharmacology, University of Karachi, Karachi, Pakistan

²Department of Pharmacology, Jinnah University for Women, Karachi, Pakistan

Abstract: Several plants have been selected based on their use in traditional systems of medicine, and research has identified a number of natural compounds that could act as Nootropicagents. In this study a herbal product Intellan containing *Centella asiatica*, *Bacopa monniera*, *Coriandum sativum*, *Amomum subulatum*, *Emblica officinalis* and another product Cytacon (Cyanocobalamine) were selected The study was designed on animal models to explore the effects on different parameters. For this the animals were given chronic dosing for 6-8 weeks during and after which the parameters were observed to determine their effects. The purpose of focusing on such formulations is to do hematological screening in long-term use. The hematological parameter included hemoglobin/HCT, total leucocyte count, platelets. The lymphocytes and the monocytes counts were increased significantly by intellan, while cyanocobalamine increases RBC counts, platelet counts, monocyte counts, hematocrit etc significantly. The SGPT, SGOT were found increased in both of these drugs.

Keywords: *Centella asiatica*, *Bacopa monniera*, Red blood cell, Cyanocobalamin.

INTRODUCTION

Various plants are used in the traditional system because of their important constituents that are used as nootropic agent. One plant that has been used as brain tonic and curative in injured conditions is *Bacopa monniera* (BM). It is a climber, a plant found throughout the Indian subcontinent in wet, soggy and muddy areas (Chunekar, 1960; Satyavati *et al.*, 1976). *Bacopa monniera* (BM), a traditional ayurvedic medicine, used for centuries as a memory boosting, anti-inflammatory, analgesic, antipyretic, sedative and antiepileptic agent. The plant, plant extract and isolated bacosides (the major active principles) have been extensively investigated in several laboratories for their neuro pharmacological effects and a number of reports are available confirming their nootropic action. Various studies have been done by the researchers on anti inflammatory, cardiogenic effects of BM preparations/extract. It is also useful in renal disorders, blood diseases, cough, anemia and poisoning (Choudhuri *et al.*, 2002). The effect of BM in inhibiting experimentally induced inflammation was compared with that of indomethacin, a known anti-inflammatory drug. The results of this study showed that BM effectively suppressed experimentally induced inflammatory reactions, by inhibiting prostaglandin synthesis and partly by stabilizing lysosomal membranes, and did not cause gastric irritation at anti-inflammatory doses (Jain *et al.*, 1994).

Centella asiatica is distributed widely in South America and Asia especially in the damp places throughout India and is known as a therapeutic agent in folk medicine,

*Corresponding author: e-mail: humera124@gmail.com

capable of improving memory and treating several neurological disorders. *Centella asiatica* also act as an anti-oxidant and thus exert significant neuro protective effect and proved effective in protection of brain against age related oxidative damage (Subathra *et al.*, 2005). *Centella asiaticais* also claimed to possess memory improving effect (Veerendra Kumar and Gupta, 2002), anti-inflammatory, wound healing (Suguna *et al.*, 1996), anticancer (Babu *et al.*, 1995) and antioxidant properties (Zainol *et al.*, 2003).

MATERIAL AND METHODS

For hematological screening 30 healthy albino rabbits of either sex weighing from 1500 to 2000 grams were used. Rabbits were selected as experimental animals because of several reasons i.e. biochemical and hematological changes produced in rabbits are relatively similar as observed in humans (Irana *et al.*, 1979). All animals were equally divided into three groups, one group served as control while other two received normal doses according to their body weight for 30 day. Before administration of drug, apparent health of these animals was monitored during the conditioning period under the laboratory environments for a week before administration of drug specifically noticing loss of hair, diarrhea, edema, ulceration and lack of activity. They were housed individually in cages, under controlled condition of temperature $23\pm 2^{\circ}\text{C}$. Diet and water was provided ad libitum.

Animal dosing

For hematological and biochemical analysis, all animals received drugs orally. The dosage form selected was

syrup. The adult dose of Intellan is 10 ml that contain 500 mg so the dose of intellan given was 12.5mg/1.5 kg which is 0.25ml of the syrup, the adult dose of Cynocobalamin (cytacon liquid) was 800µg while 5ml contain 35µg of cyanocobalamin. The animal dose given was 20µg/1.5kg i.e 2.85ml of the syrup. These drugs were given for 30 days. The animal doses were calculated on the basis of their body weight from adult dose i.e. 500mg of intellan, and 35µg of Cynocobalamin.

Sample collection

Blood samples of 7ml were collected once at the end of dosing period i.e. 30 days by cardiac puncture. The 2 ml blood was taken in EDTA.K3 tubes for hematological study and remaining 5 ml was used for biochemical study.

RESULTS

Table 1 showed the RBC count, Hematocrit(Hct), Hemoglobin(Hb), MCH, MCV, MCHC values. This table showed that RBC counts of Intellan treated rabbits was increased non-significantly as compared to control rabbits. But in Cynocobalamin treated group RBC counts were increased significantly after day 30 (P<0.05). Hemoglobin of Intellan treated rabbits and Cynocobalamin was increased non-significantly as compared to control rabbits after day 30. Hematocrit of Intellan and cyanocobalamin treated rabbits was increased significantly as compared to control rabbits (p<0.05) after 30 days. Mean cell volume (MCV) of both groups i.e.

Intellan and Cyanocobalamin treated rabbits were increased non-significantly as compared to control rabbits after day 30. MCHC of Intellan as well as Cyanocobalamin treated rabbits were decreased non-significantly as compared to control rabbits after 30 days dosing. MCH of Intellan and Cyanocobalamin treated rabbits were decreased non-significantly as compared to control rabbits.

Table 2 showed that WBC count of Intellan and Cyanocobalamin treated rabbits after 30 days was increased non-significantly as compared to control rabbits. Platelets count of Intellan treated rabbits were decreased non-significantly as compared to control rabbits. While Platelets count was decreased significantly in Cytacon treated group (p<0.05).

Table 3 showed SGOT, SGPT, glucose, total cholesterol, HDL-C, LDL-C, triglycerides values at day 30. The SGOT values of Intellan and Cyanocobalamin treated rabbits was increased non-significantly as compared to control rabbits After 30 days dosing. Glucose values of Intellan treated rabbits was decreased significantly as compared to control rabbits. (p=0.001) while glucose values was increased non-significantly in cytacon treated group.

Total cholesterol values in both groups i.e. Intellan and Cyanocobalamin treated rabbits were increased non-significantly as compared to control rabbits after day 30.

Table 1: Variations in RBC’s profile among different groups.

Groups	30 Day					
	RBC	Hb	Hct	MCV	MCHC	MCH
Control	5.52±0.21	10.2±0.55	34.5±1.03	62.6±0.83	29.3±0.94	18.4±0.61
Intellan	6.39±0.38	11.3±0.60	41.19±2.44*	64.5±0.65	27.65±0.25	17.8±0.21
Cytacon	7.29±0.60*	12.4±0.97	46.8±4.27*	63.8±0.66	26.76±0.48	17.07±0.32

Table 2: Variations in WBC’s profile and platelets among different groups.

Groups	30 Day				
	WBC	Granulocyte	Monocyte	Platelet	Lymphocyte
Control	5.94±0.468	46.8±4.11	5.65±0.98	509±67.04	37.8±5.34
Intellan	7.12±0.822	47.19±2.06	10.55±0.74*	441±40.87	42.26±2.12
Cytacon	6.21±1.00	50.8±3.98	9.72±0.72*	315±39.85	39.4±3.95

Table 3: Variations in biochemical parameter among different groups.

Drugs	30 Day						
	Total cholesterol	HDL-C	LDL-C	Triglyceride	SGPT	SGOT	Glucose
Control	26.75±3.10	1.56±0.37	15.89±2.13	55.2±6.2	80.4±12.33	56.7±11.35	160.9 ±10.82
Intellan	21.2±4.01	0.88±0.05	16.2±3.8	21.8±4.04	86.9±11.07	64.6±8.28	85.9 ±12.7***
Cytacon	12.8±6.61	1.6±0.18	24.4±5.63	105.4±23.40	91.6±15.56	103.1±24.47	204.8 ±20.62

HDL cholesterol values of Intellan treated rabbits was decreased non-significantly as compared to control rabbits. But in Cyanocobalamin treated group HDL cholesterol values was increased non-significantly after 30 days. Results showed that LDL cholesterol values of Intellan as well as Cyanocobalamin treated rabbits was increased non-significantly as compared to control rabbits. Triglyceride values of Intellan treated rabbits was decreased significantly as compared to control rabbits. ($p < 0.05$), while triglyceride levels was increased non-significantly by the Cyanocobalamin after 30 days.

DISCUSSION

The level of Hemoglobin was increased by all the drugs used, as well as the erythrocyte RBC's counts are increased correlating the two parameters. The increase in the count by Intellan is due to the ingredient *Bacopa monniera* which has neuro-protective effects as well as reported memory enhancing abilities. It can improve the cell viability and increase the life span of neuron, also it can protect the neurons and cells from oxidative stress (Nanteetip *et al.*, 2008). This suggest that the RBCs may also have been protected against oxidative stress and their increase in number and correspondingly increase hemoglobin level. *Centella asiatica* also possess antioxidants activity and that may also contribute here.

The increase in count and hemoglobin level by Cynocobalamin is very obvious, since vitamin B₁₂ is required for the synthesis of hemoglobin and its deficiency can lead to maturation failure anemia. If vitamin B₁₂ is given then there will be increased synthesis of RBCs and also, better incorporation of iron and increase in the hemoglobin concentration. These all effects are also related to their ability to increase the overall performance and cognitive function because in order to enhance memory and learning, the oxygen delivery is required, which is correlated to the counts of erythrocytes and then to the levels of hemoglobin concentration. Both of these effects contributed to increase the overall mental activities also. Since the hemoglobin concentrations are increased so the hematocrit value, mean hemoglobin concentration (MCH) was decreased by both drugs and the decrease were all statistically non-significant. The mean corpuscular hemoglobin concentration (MCHC) was also decreased but each erythrocyte was better saturated with oxygen, and this saturation will be utilized to deliver more oxygen, especially for increased mental or physical work. Even the value is too low but the number of RBCs are increased which is responsible for this effect.

The effect of Intellan on WBCs count is also significant and we have observed that WBCs. count was increased by both drugs. This is a very good finding indicating that these drugs can help body to fight different infections as

well as improve the immunity. The fractional percentages of different leucocytes support these observations as the number of monocytes and lymphocytes increase significantly. There was a slight increase in the counts of granulocytes also and this supports the fact that with these drugs the defense system of the body will also become strong. The increase was also seen after Intellan as it also contains *Coriander*, *Centella asiatica*, *Bacopa monniera*, which improve cell viability and also enhance the effects of growth inducers and differentiators to significant levels and increase the concentration of leucocytes. The effect seen after cynocobalamin administration was not significant but the count was slightly increased, because usually vitamin B₁₂ can act as erythropoiesis stimulator only, not a complete bone marrow stimulator.

The effect of two drugs on platelets was non-significant, as all slightly decreased the level of platelets. This may be due to increased utilization of platelets in the body, because the animal was not restricted in the activity and may be exposed to certain traumas. This could be a useful finding also, as the concentration of platelets was decreased but not very significantly and the hematocrit value of the blood was not increased by the platelets, it was increased by RBCs. So there are less chances of excessive platelet aggregation in the microvasculature and less chances of formation of micro thrombi. This could be beneficial in those people who need a drug that should improve their memory, and cognition but simultaneously should not predispose to thrombus formation.

The levels of SGOT was non-significantly increased by Intellan, but significantly increased by cytacon. SGOT or Serum glutamic oxaloacetic transaminase is an enzyme which help liver to build and break down proteins. It is found in many tissues and if any of these organs are affected, it will be released in the blood, so it is not a specific indicator of liver damage as SGPT. It is also present in RBCs which could be the reason of increase in SGOT level after the administration of cyanocobalamin. The increased use of RBCs may lead to the breakdown and release of SGOT which is contributed to this increased level of SGOT and is not a direct indicator of liver damage. This is supported by the finding that SGPT is not significantly elevated by cyanocobalamin supporting our discussion. SGPT/ALT, or Serum glutamic pyruvate transaminase is commonly associated with liver, however fluctuations in the ALT level is normal over the course of the day, and the ALT levels can also increase in response to sternous physical exercise (Paul and Giboney). In our finding there was non-significant increase in the SGPT level by both drugs suggesting that neither for them has produced any liver toxicity, and only the slight increase which was observed is considered normal.

Glucose is decreased after administration of Intellan possibly due to increased utilization to provide energy for different activities, especially mental work, as Intellan is acting as memory enhancers requiring more energy at the brain level, thereby decreasing the glucose level in the blood. Also our finding is supported by hypoglycemic effect of coriander as reported by Chitra and Leelamma, (2000). Coriander is an ingredient in Intellan which could explain the hypoglycemia observed after the administration of these drugs. The increase in the concentration of glucose after the administration of cyanocobalamin may be related to the fact that if there is increased requirement of energy, then due to increased RBCs count there is increased glucose level. Also according to the Mauro and Isaaks (1989) nucleated RBCs of most vertebrates rely less on TCA for energy production and more on the glycolytic mechanism of carbohydrate, which is related to our observation. The metabolism of glycogen leads to increase glucose level in blood which gives intense energy availability for increased performance in physical and mental activities. This also supports our precious findings that after the administration of cyanocobalamin the mental function and ability to accomplish task is increased. It is important to mention here that the increase in glucose is not so much that it could be considered as hyperglycemia.

As far as the effect on lipid profile is concerned, it was observed that after the administration of intellan the level of triglycerides was decreased, and the level of LDL-cholesterol is nearly the same as that of control. The level of HDL-cholesterol is non-significantly decreased, and overall total cholesterol is decreased which is considered to be a beneficial effect. Since these drugs are given for a long period of time, and if the effect on total cholesterol is such that the levels are within the normal values, then it is considered to be an advantage. This effect could be attributed to the presence of Centella, Bacopa and Coriander all of which possess antioxidant properties. According to Radhika *et al.*, (2008) Bacopa Monniera could be responsible for the restoration of metabolic activities, and Bacopa is one of the ingredients of intellan which is responsible for this action. The effect of Cyanocobalamin on lipid profile is that it significantly increased triglyceride level possibly after the cleavage of chylomicrons, because the level of LDL-C was not highly increased. There was also slight increase in the level of HDL-C and the total cholesterol was significantly decreased by Cyanocobalamin indicating that initially there was increase in triglyceride level but finally the total cholesterol will be decreased and the lipid profile will remain normal or will be decreased. This will also prove beneficial, because it supports our finding that if Cyanocobalamin is used for prolong period, it will not disturb the lipid profile, rather it can be safely used for a long period even in people who has high cholesterol level.

CONCLUSION

From the research data it is concluded that Cyanocobalamin is better than as compared to intellan if taken for long period of time and has good drug profile, less adverse effects than Intellan. Still several aspects are yet to be determined and need further research.

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