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Exploration Of Oculo-Antihypertensive Activity In *Panex ginseng* Extract : An Experimental Study

Mahendra Kumar[1], Ranjit Singh[2], Neeraj Masand[1], Kuldip Kumar Saxena[3]
[1]Dept of Pharmacy, LLRM Medical College Campus, Meerut,
[2]School of pharmacy Shobhit University Meerut,
[3]Deptt of pharmacology L.L.R.M. Medical College Meerut. Email:mahendra1129@yahoo.co.in

Abstract :

The present study was carried out to evaluate the oculo-hypotensive – antihypertensive effect of aqueous extract of Panex ginseng (PGE) in experimental model of glaucoma. The study was conducted in rabbits which were divided to constitute 3 experimental models viz. normotensive, water loading induced oculo-hypertensive (acute) model and steroid induced (chronic) model using Timolol 0.25% as standard. In normotensive model, effect of PGE (0.25%, 0.5% and 1.0% w/v) on intra-ocular pressure (IOP) was recorded every hour for 8 hours with the help of tonometer whereas in water loaded and steroid induced model, the effect of PGE (1% w/v) on IOP was evaluated after inducing ocular hypertension with the help of oral administration of water (70 mg/kg) and prednisolone 1% eye drops respectively. The effect of PGE (0.25%, 0.5% and 1.0% w/v) on intra-ocular pressure (IOP) was recorded in water loading model at every 15minfor two hours and in steroid induced model at every hour for 8 hours. The aqueous leaf extract of Panex ginseng showed reduction in normotensive animals when used topically in concentration of 0.25% (w/v), 0.5% (w/v) & 1.0% (w/v) concentration respectively. The maximum effect was observed in the concentration of 1.0% (w/v) PGE. This concentration was further evaluated in water loaded (acute) and steroid induced (chronic) ocular hypertensive model. It was observed that a maximum mean difference of 29.0 mm of Hg was found between vehicle treated eyes and 1% PGE treated eyes in acute model ($p < 0.0001$) after 75 minutes whereas a maximum mean IOP reduction of 35.4 mm of Hg was observed in chronic model ($p < 0.0001$) after 3 hours of test drug administration.

PGE (1% w/v) reduced IOP to the same extent as Timolol (0.25%, standard drug) produced in acute and chronic ocular hypertensive models in rabbits. The probable mechanism of action of this effect could be attributed to diuretic, antioxidant, nitric oxide scavenging anticholinesterase activity or suppression of aqueous humar production.

Keywords : Glaucoma, Ocular hypertension, Cannabis sativa, Timolol.

INTRODUCTION:

Glaucoma, a condition in which IOP gradually increases causing damage to the optic nerve and gradual deterioration of vision, is the second largest cause of blindness.(REFERANCE A-1) Loss of vision in glaucoma is gradual; hence glaucoma is nicknamed as "sneak thief of sight". There are about 67 million sufferers from glaucoma in the world and out of them; 12 million are only in India.(REFERANCE-3) The worldwide prevalence is increasing and it is estimated that about 79.6 million people will be affected by 2020 (REFERANCEA-2) [1] The aim of the treatment of glaucoma is to reduce the IOP by either improving aqueous humor outflow or reducing its production. The commonly used drugs are beta blockers (eg Timolol), alpha adrenergic agents (eg Brimonidine), carbonic anhydrase inhibitors (eg, Brinzolamide), prostaglandin analogues (eg Lanatoprost), and miotics (eg Pilocarpine). But the cost and side effects of and contra-indications to these agents limit their use.(REFERANCE-4,5,6) Various indigenous plants have been found to possess oculo-antihypertensive activity but perusal of literature indicates that sufficient scientific studies have not been carried out to establish their efficacy against glaucoma. The drugs from natural sources if found effective in reducing IOP without serious side effects in glaucomatous eyes, may prove to be an important tool for the scientific community in the treatment of glaucoma.

It has been reported that aqueous extract of Panex ginsengleaves possesses strong antioxidant (REFERENCE-9) and diuretic (REFERENCE-11)activity both in vivo and in vitro studies. Keeping in view of the above pharmacological actions of aqueous extracts of Panex ginseng(PGE) , present study was designed to evaluate its oculo-hypotensive effect in the treatment of Glaucoma.

MATERIALS & METHODS:

The study was conducted on New Zealand white rabbits of either sex weighing 1.5-2.0 kg. Animals were kept in standard laboratory conditions and were provided with normal pellet diet and tap water ad libitum. The experimental protocol was approved by IAEC and was executed according to guidelines of CPPGEA, India. Only thPGE rabbits were selected for study which were found to

be normal in general and ophthalmic examinations. After one week of habituation in the animal house, training for acceptance of tonometry was given to the animals.

Test Drug: The leaves of Panex ginseng were collected and were identified at Indian Council of Agricultural Research, Pusa Campus, New Delhi. Leaves were dried in shade. Extract of Panex ginseng leaves was prepared by powdering 100 gm of dried leaves and extracting it with aqueous solvents by soxhlet extraction process. The extract was filtered and the solvent was evaporated by distillation. The concentrated extract was dried on water bath. It was powered and kept in amber coloured bottle at 4^0C. The percentage yield of the extract was 3.5%.

The Panex ginseng extract (PGE) was dissolved in 0.25% autoclaved hydroxy propyl methyl cellulose (HPMC), so as to give a concentration of 0.25%, 0.5% and 1.0% (w/v). HPMC was used to increase the viscosity of the eye drop so that the corneal contact time is increased. 0.01% Benzalkonium chloride was used as preservative and pH of the eye drop was adjusted by citric acid, sodium citrate & sodium phosphate.

Standard drug used was Timolol (a beta blocker) eye drop in the concentration of 0.25% (procured from FDC International Ltd) in which the active ingredient is Timolol maleate. This preparation also contains sodium dihydrogen phosphate dihydrate, disodium edetate, disodium phosphate dodecahydrate, benzalkonium chloride 0.01% (w/v), sodium hydroxide, sodium chloride and water for injection.

Estimation of IOP: Estimatimation of IOP was done by using Schiotz Tonometer (Rudolf Reister GmbH, Germany). Tonometries were repeated twice in a week. Too frequent tonometries were avoided in order to save the integrity of the corneal eithelium. Baseline tonometries were obtained when the ocular tension turned out to be constant.

Experimental Design: Effect of different concentrations of PGE eye drops were seen in normotensive rabbits The concentration of PGE showing maximum activity was further evaluated in water loading (acute) model and steroid induced (chronic) model. To induce acute ocular hypertension in the water loading model, tap water (70ml/kg) was rapidly administered in the conscious rabbits through an orogastric tube to raise IOP.(REFERENCE13 and 14)This elevated the IOP is rabbits which lasted for about 2 hours. The IOP was measured every 15 minutes. The water loading model mimics the acute model. In chronic model of ocular hypertension the IOP was elevated by topical instillation of 10µl of prednisolone (1% eye drops)(COMPANY) in both eyes, twice daily for a period of 40 days continuously [17,18.] During this period, the IOP was measured twice every week. Topical instillations of glaucocorticoids has been shown to cause elevation in IOP in rabbits [(17, 18, 19)].

Normotensive Model

The experimental study was performed in two steps:

1st Step: Normotensive rabbits were randomly divided into 4 groups of 6 rabbits each. Baseline IOP was measured with the help of tonometer between 8:30 AM to 9:30 AM on the day of experiment. In the animals of 1st group , 50µl of 0.25% PGE was instilled in the randomly chPGEn eye while the other eye of the same animal was treated with 50µl of 0.25% vehicle containing 0.25% HPMC solution containing citric acid, sodium phosphate and sodium citrate (for maintaining pH at 6.5 to 8.5) and 0.01% benzalkonium chloride (as preservative). Similarly, in the randomly chPGEn eye of the rabbit of group 2 & group 3, PGE in the concentration of 0.5% and 1% (dPGE 50µl) was instilled respectively whereas in the other eye of the same rabbit, equal volume of vehicle was instilled as was done in the animals of group 1 .In the fourth group of rabbits 50ul of 0.25% of Timolol eye drop was instilled T in the randomly chPGEn eye while the other eye of the same animal was treated with 50µl of 0.25% vehicle .The IOP was recorded every hour for next 8 hours.

2nd Step: The dPGE of PGE showing maximum reduction in IOP in comparison to baseline IOP in normotensive rabbits was further evaluated for its effect in acute (water loading induced) and chronic (steroid induced) models of ocular hypertension.

Water loading induced ocular hypertensive model:

In this model, rabbits were divided in 3 groups of 6 animals each. The baseline IOP was estimated after overnight fasting between 8:30 AM.to 9.30 A.M. Animals of 1st group were instilled with 50µl of vehicle (0.25% HPMC and other substances) in one of the randomly chPGEn eye and 50µl of normal saline in other eye. The second group received 50µl of PGE (1% w/v) in one of the randomly selected eye and the same volume of normal saline was instilled in the other eye. The third group received 50µl of 0.25% Timolol in one of the randomly selected eyes and the same volume of normal saline in the other eye.

One hour after the Vehicle ,PGE/Standard Drug/ instillation, rabbits were administered with tap water (70ml/kg), through an oro-gastric tube. This was followed by IOP measurements at an interval of 15 minutes for a total period of 120 minutes

Steroid Induced ocular hypertensive model:

For the evaluation of efficiency of PGE in steroid induced model, 24 animals were trained to accept tonometry and IOP was measured for 15 days so as to make a record of baseline IOP. Then, these rabbits were instilled with prednisolone 1% eye drop (10µl) in one of the randomly chPGEn eye and in the other eye, normal saline (10µl) was instilled twice a day for a period of 40 days. During this period, IOP of the rabbits was measured twice a week. Then, these rabbits were randomly divided in 4 groups of 6

animals each. On the day of experiment, the baseline IOP was measured.

1st group was then instilled with 50µl of normal saline in both the eyes. This group served as control (normal saline gp).

2nd group was instilled with 0.25% HPMC solution (50µl) in one of the randomly chPGEn eye while normal saline (50µl) was instilled in the other eye.

3rd group which served as drug treated group and was instilled with 1.0% PGE (50µl) in one of the randomly chPGEn eye while 50µl of normal saline in the other eye.

4th group served as standard drug treated group and was instilled with 0.25% Timolol (50µl) into one of the randomly chPGEn eye of the rabbits while 50µl of normal saline in the other eye.

This was followed by IOP estimations at hourly interval for a total duration of 8 hours.

STATISTICAL ANALYSIS: The results were expressed as mean ± SD. One way ANOVA was used for determining the statistical significance of the differences between groups at probability level of 95%.

RESULTS: The estimation of reduction in IOP by PGE was carried out in the rabbits with normal IOP (Normotensive model) and experimentally induced raised IOP rabbits, with the help of oral administration of water (Water loading model) and by the instillation of corticosteroid eye drops (Steroid induced model).

Normotensive Model

In this model, ocular hypotensive effect was evaluated at 1 hour interval after the administration of standard drug (Timolol 0.25%) & PGE (0.25%, 0.5% & 1.0%) separately in different groups.

Maximum percent change in IOP was observed after 5 hours in 0.25% PGE and 0.5% PGE treatment (23.9± 4.42 and 30.4 ± 7.18 respectively) whereas the maximum IOP reduction by 1% PGE treatment was observed after 4 hours (32.6 ± 3.96). (Table1) Moreover, 1% PGE showed significant reduction in IOP from the very first hour when compared to control eye IOP (Table-1). Thus, 1% PGE is more efficacious than 0.25% and 0.5% PGE due to early onset of action and higher percent reduction in IOP. (Figure 1)

Further, significant difference in IOP reduction was observed in 4,5 and 7 th hour when effect of 1% PGE and standard drug (Timolol 0.25%) was compared. (Table -2) The comparison between different PGE concentrations at 4 hour is detailed in Table 3.

Table 1: Maximum Oculo-hypotensive Activity of Panex ginsengin Normotensive Rabbits (N=6).

Percent PGE	Peak effect	IOP in control eye	IOP in treated eye	% Change in IOP
0.25%	5 hours	21.3 ± 0.42	16.2± 0.94	23.9± 4.42
0.5%	5 hours	21.4 ± 0.48	14.9 ± 1.53	30.4± 7.18
1.0%	4 hours	21.4 ± 0.97	14.4 ± 0.84	32.6 ± 3.96

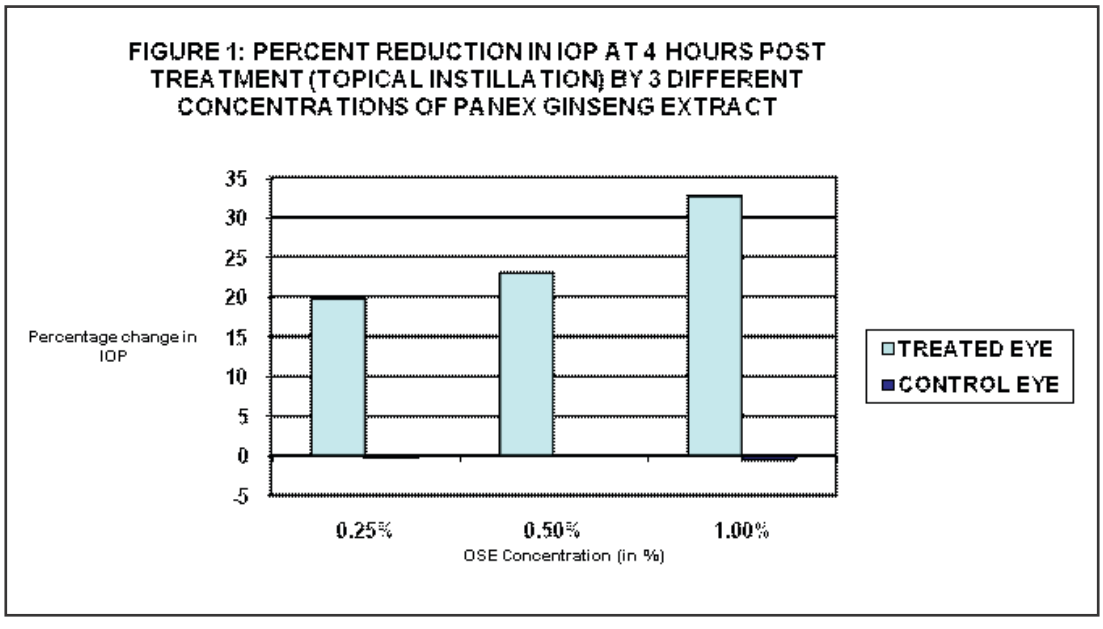

Table 2: Comparison of IOP reduction between different concentration of PGE and 0.25% Timolol in normotensive model
PGE (%) Mean±SD (mmHg)

Time (Hrs)	0.25%	0.5%	1.0%	Timolol (0.25%) Mean±SD
1	2.1 ± 10.83#	6.00 ± 3.09*	10.3 ± 7.95#	12.40 ± 4.64
2	10.4 ± 7.94*	13.9 ± 4.99#	18.4 ± 8.10#	19.50 ± 5.22
3	16.2 ± 6.44*	20.0 ± 6.38*	26.5 ± 7.82#	31.20 ± 6.01
4	19.7 ± 6.86*	23. 0 ± 7.71*	32.6 ± 3.96*	39.50 ± 4.93
5	23.9 ± 4.42*	30.4 ± 7.18#	28.6 ± 3.13*	36.40 ± 6.28
6	20.8 ± 9.42#	26.9 ± 9.25#	22.8 ± 6.28#	30.0 ± 6.92
7	15.2 ± 7.32#	20.8 ± 9.71#	18.3 ± 7.00*	21.6 ± 8.15
8	9.0 ± 5.55 #	12.0 ± 4.63 #	14.6 ± 6.41#	14.1 ± 9.93

* $p < 0.05$ and # $p > 0.05$. in comparision to Timolol 0.25%

Table 3: Comparison of P-value between three concentrations of PGE at fourth hour

Comparison Between Different Concentrations Of PGE	*t*-Value	*p*-Value	Significant / Non-Significant
0.25% *vs* 1.0%	-3.9 89	0.002	Significant
0.25% *vs* 0.50%	-0.783	0.451	Non -Significant
0.50% *vs* 1.0%	-2.713	0.021	Significant

Table 4: Comparison of percent change in IOP between PGE (1%) eye drop & control eye in water loading induced acute ocular hypertensive model.

Time min		Ctrl eye	%Change in IOP	Tre eye	%Change in IOP	TE-CE
15	MEAN	30.7	-43.6	26.0	-21.9	21.8
	SD	1.09	5.09	2.12	9.93	6.68
30	MEAN	32.8	-53.6	28.1	-31.5	22.1
	SD	2.57	12.05	2.02	9.45	2.72
45	MEAN	42.0	-96.8	36.9	-72.7	24.1
	SD	3.32	15.55	2.96	13.84	2.37
60	MEAN	42.6	-99.6	37.0	-73.1	26.5
	SD	2.46	11.52	2.68	12.56	1.70
75	MEAN	44.6	-108.8	38.4	-79.9	29.0
	SD	1.72	8.07	1.66	7.75	3.65
90	MEAN	38.5	-80.5	32.6	-52.8	27.6
	SD	2.10	9.81	2.19	10.27	1.18
105	MEAN	32.8	-53.4	27.9	-30.8	22.6
	SD	2.32	10.87	2.27	10.65	3.66
120	MEAN	28.5	-33.6	26.3	-23.3	10.4
	SD	4.44	20.80	4.53	21.20	3.81

Water Loading Model

The ocular antihypertensive activity was evaluated every 15 minutes after the administration of test drug (PGE, 1% w/v) and standard drug (Timolol, 0.25 %). Significant decrease in IOP was observed in PGE treated group as compared to control eye throughout the study period (p value 0.005 to 0.0001).(Table-4) Maximum percentage change in IOP in PGE (1%) treated group was 79.9%, which was observed after 75 minutes whereas in Timolol treated group it was 70.6% after 60 minutes. The percent reduction in IOP by PGE 1% as well as by Timolol 0.25%, when compared with control eye treated with normal saline is significant till 105 minutes).(Table 4&5) No significant comparision except for, at 75 minute was observed for percent change in IOP between PGE (1%) eye drop & Timolol (0.25% eye drop) in water loading induced acute ocular hypertensive model. (Table 6)

Time min		Ctrl eye	%Change in IOP	Tre eye	%Change in IOP	TE-CE
15	MEAN	29.6	-38.6	25.8	-20.7	17.9
	SD	2.49	11.63	1.96	9.16	5.39
30	MEAN	33.7	-62.6	29.8	-39.5	23.1
	SD	1.03	4.84	1.73	8.08	3.78
45	MEAN	42.3	-98.0	35.6	-66.8	31.2
	SD	4.12	19.31	4.09	19.13	4.33
60	MEAN	43.9	-105.5	36.5	-70.6	34.9
	SD	2.86	13.37	2.80	13.13	3.98
75	MEAN	40.5	-89.5	35.0	-63.8	25.7
	SD	2.00	9.36	1.43	6.69	4.14
90	MEAN	36.0	-68.5	31.3	-46.7	21.8
	SD	2.60	12.18	2.06	9.65	5.70
105	MEAN	31.6	-48.0	27.2	-27.3	20.8
	SD	2.69	12.60	2.46	11.50	4.56
120	MEAN	28.5	-33.3	24.8	-16.3	17.1
	SD	2.93	13.73	2.46	11.53	3.12

Table 6: Comparison of percent change in IOP between PGE (1%)eye drop & Timolol (0.25% eye drop) in water loading induced acute ocular hypertensive model.

Time (min.)	Percent change in IOP by 1% PGE (mean ± SD)	Percent change in IOP by Timolol 0.25% (mean ± SD)	p-value	Level of significance
15	-21.90 ± 9.93	-20.70 ± 9.16	0.8321	NS
30	-31.50 ± 9.45	-39.50 ± 8.08	0.1461	NS
45	-72.70 ± 13.84	-66.80 ± 19.13	0.5541	NS
60	-73.10 ± 12.56	-70.60 ± 13.13	0.7391	NS
75	-79.90 ± 07.75	-63.80 ± 6.69	0.0032	S
90	-52.80 ± 10.27	-46.70 ± 9.65	0.314	NS
105	-360.80 ± 10.65	-27.30 ± 11.50	0.5964	NS
120	-23.30 ± 21.20	-16.30 ± 11.53	0.4936	NS

S= Significant, NS= Non-Significant.

Steroid Induced Model: The ocular antihypertensive activity was evaluated every hour after the administration of PGE 1% and standard drug Timolol (0.25%) for next 8 hours. Highly significant decrease in IOP was observed in PGE treated group as compared to control eye in all the time intervals (p value 0.006 to 0.0001)(Table-7). The maximum percent change in IOP in PGE treated eye was 35.7% after 3 hours whereas the maximum percent change in IOP in Timolol treated eye after 3 hours was 33.4%. Moreover, no significant difference was noted when effect of PGE (1%) was compared with Timolol (0.25%) from 1st to 8th hour. (Table 8)

Table 7: Comparison of percent change in IOP between PGE(1% eye drop) & control eye in Steroid Induced chronic ocular hypertensive model Model:

Timemin		Ctrl eye	%Change in IOP	Tre eye	%Change in IOP	TE-CE
1	MEAN	21.4	-0.2	24.9	-16.4	-16.2
	SD	1.32	6.17	1.23	5.77	9.29
2	MEAN	21.4	-0.2	27.0	-26.5	-26.4
	SD	1.32	6.17	1.27	5.96	9.36
3	MEAN	21.4	-0.2	29.0	-35.7	-35.4
	SD	1.43	6.72	1.00	4.68	8.09
4	MEAN	21.4	-0.2	29.0	-35.7	-35.4
	SD	1.43	6.72	1.00	4.68	7.64
5	MEAN	21.5	-0.5	28.3	-32.6	-32.1
	SD	1.47	6.89	1.06	4.95	7.61
6	MEAN	21.3	0.2	26.6	-24.7	-24.9
	SD	1.50	7.04	0.69	3.21	8.07
7	MEAN	21.4	-0.3	24.8	-16.1	-15.8
	SD	1.22	5.71	1.40	6.57	9.79
8	MEAN	21.3	0.2	23.9	-12.0	-12.3
	SD	1.33	6.21	1.29	6.04	8.52

Table 8: Comparison of percent change in IOP between PGE(1% eye drop) & Timolol (0.25% eye drop) in steroid induced ocular hypertensive model.

Time (hours)	Percent change in IOP by 1% PGE (Mean ± SD)	Percent change in IOP by Timolol 0.25% (Mean ± SD)	P-value	Level of significance
1	-16.4 ± 5.77	-13.40 ± 5.00	0.3585	NS
2	-26.5 ± 5.96	-25.00 ± 2.63	0.5852	NS
3	-35.7 ± 4.68	-33.40 ± 5.52	0.4543	NS
4	-35.7 ± 4.68	-31.70 ± 5.54	0.2065	NS
5	-32.6 ± 4.95	-29.10 ± 10.00	0.4600	NS
6	-24.7 ± 3.21	-22.30 ± 5.04	0.3484	NS
7	-16.1 ± 6.57	-20.90 ± 4.22	0.1631	NS
8	-12.0 ± 6.04	-16.60 ± 5.51	0.1982	NS

S= Significant, NS= Non-Significant.

Discussion :

The present study was designed to evaluate the effect of PGE in different concentrations on IOP in experimental models of glaucoma in rabbits. The experimental elevation in IOP was achieved by oral administration of water (which mimics the acute rise in IOP) and topical administration of steroids (which mimics the chronic elevation in IOP). Water (70ml/kg body weight) was given orally by orogastric tube for acute model and effect of PGE (1% of 50µl topical) was evaluated for 2 hours at every 15 minutes interval. In another set of experiments, Prednisolone (1% eye drops) was given in chronic ocular hypertensive model for 40 days and effect of PGE (1% of 50µl topical) was evaluated for 8 hours at every one hour interval. Timolol (0.25% topical) was used as standard drug.

The study concluded following outcomes:

1) Significant difference in mean peak IOP was observed when 0.25%, 0.5% and 1.0% PGE treated groups were compared with the control groups in all time intervals.

2) Significant difference in mean peak IOP was observed when 0.25 % and 0.5% PGE treated group was compared with the rabbits treated with 1.0% PGE ($p<0.05$).

3) In water loading model, 1.0% PGE showed a significant decrease in IOP between control and PGE treated group till 105 minutes. ($p<0.05$)

4) Effect of 1% PGE against acute rise in response to water loading was almost equal to that caused by Timolol (maximum percent change in IOP was 79.9 after 75 minutes for PGE treated group where for Timolol, maximum percent change was 70.6 after 60 minutes).

5) The maximum difference in IOP between control and treated eyes in PGE treated group was observed after 75 minutes of water loading.

6) Significant difference in peak IOP reduction in 1% PGE group and Timolol group were observed in steroid induced model.

7) Both Timolol treated group and 1% PGE treated group showed maximum reduction in IOP after 3 hours in steroid induced model. But reduction in IOP was slightly higher in case of PGE treated group (35.7) as compared to Timolol treated group (33.4).

8) It was observed that no effect was produced in the eyes of control group which received same volume of inert vehicle.

References :

Quigley H.A., Broman A.T., (2006): The number of people with glaucoma worldwide in 2016 and 2020. *Br.J.Ophthalmol*. **90**:262-267

Radius R.L., Diamond G.R, Pollock I.P., Langhum M.E.(1998): Timolol a new drug for the management of chronic simple glaucoma. *Arch Ophthalmol.* **96**:1003.

Trevisan M.T., Vasconcelor Silva M.G., Pfundstein B., Spiegelhalder B., Owen RW. (2006): Characterization of the volatile pattern and antioxidant capacity of essential oils from different species of genus Ocimum. *J.Agric Food Chem.*; **54**:4778-82.

Gupta S., Mendiratta P.K., Singh S., Sharma K.K., Shukla R. (2006): Antidiabetic, antihyper cholesterolaemic and antioxidant effect of Panex ginseng(Linn) seed oil. *Indian J. Exp. Biol.* **44**: 300-4.

Bhattacharya Supriya Kumar (2004): Handbook of aromatic plants. Pointer publishers, Jaipur (Raj.), pp.332.

Pandey A.S., D.N. Pant M.C. (1994): Changes in the blood lipid profile after administration of Panex ginsengleaves in normal albino rats. *Indian J. Physiol. Pharmacology.* **38 (4)** :311-312.

Singh S., Taneja M., Majumdar D.K. (2007): Biological activities of Panex ginsengL. fixed oil – an overview. *Indian J. Exp. Biol*. **45(5)**: 403-12.

Yanpallewar S.U., Sunita Rai, Mohan Kumar, Acharya S.B. (2004):Evaluation of antioxidant and neuroprotective effect of Ocimum sanctum on transient cerebral ischemia and long term cerebral hypoperfusion: *Pharmacology Biochemistry and Behavior*. **79** :155-164.

Geetha, R.Kedlya, Vasudevan D.M. (2004): Inhibition of lipid peroxidation by botanical extracts of Ocimum sanctum: In vitro and in vivo studies. *Life sciences*. **76(1)**: 21-28.

Muglikar A.G., Kothekar M.A., Chilwant K.S. and Jaju J.B. (2004): Effect of *Panex ginseng* aqueous leaf extract on gentamycin induced nephrotoxicity in rats. *Indian J. Pharmacol*. **36(2)**: 123.

Pandey B.P., Anita (1999): In Economic botany. Chand and Company Ltd., Ramanagar, New Delh, pp.294.

Gulcim I., Elmastas M. and Aboul-Enein H.Y. (2007): Determination of antioxidant and radical scavenging activity of basil assayed by different methodologies. *Phytotherapy Research*. **21(4)** : 354-361.

Sue Stevens, Clare Gilbert and Nick Astbury (2007): *Community eye health*. **20 (64)**: 74-75.

Thorpe R.M., Kolker A.E. (1967): A tonographic study of water loading in rabbits. *Arch. Ophthalmol*. **77**:238.

McDonald T.O., Hodges J.W., Burgmann A.R.(1969): The water loading test in rabbits. *Arch. Ophthalmol*. **82**:381.

Goldman H. (1962): Cortisone Glaucoma. *Arch.*

Ophthalmol. **68**:621-626.

Armaly M.F. (1963): Effect of corticosteroid on intraocular pressure and fluid dynamics. *Arch. Ophthalmol* ; **70:**482-491.

Jackson R.T., Waitzman M.B.(1965): Effects of some steroids on aqueous humor dynamics. *Exp: Eye Res*. **4**:112-123.

Lorenzetti O.J.(1970): Effects of corticosteroids on ocular dynamics in rabbits. *J. Pharma Exp: Ther* **175**:763-772.

Levene R.I., Rothberger M., RPGEnberg S. (1974): Corticosteroid glaucoma in the rabbits. *Ann. J. Ophthalmol.* **78**:505-510.