RESULTS

Following the inclusion criteria a total of hundred and five male subjects were enrolled in the study with 35 subjects in each group. Table 1 & Fig. 1, 2 & 3 shows the baseline characteristics of the patients from all the groups. All the subjects were age matched. Based on their FPG levels the patients were divided into the following groups

Group I. Including 35 Healthy males.

Group II. Including 35 Prediabetes males.

Group III. Including 35 Diabetes males.

Results according to the objective of the study

1. **Primary objective** : To estimate HPT axis hormones(TRH, TSH, T3, T4), HPA axis hormones(CRH, ACTH, Cortisol), HPG axis hormones(GRH, LH, FSH, Testosterone), pineal gland hormone (Melatonin) and Insulin levels in normal, prediabetes and diabetes male subjects and compare them between the groups.

Table 1: Comparison of Mean Age, fasting plasma glucose (FPG) and Insulin levels between the groups.

Variable	Group I (n=35)	Group II (n=35)	Group III (n=35)
Age (years)	36.71 ± 5.90	38.37 ± 3.54	39.43± 4.16
FPG (mg/dl)	86.49 ± 6.55[b,c]	110.77 ± 7.14[a,c]	189.69 ± 53.92[a,b]
Insulin (µIU/ml)	12.6 ± 2.88 [b,c]	35.41 ± 3.87 [a,c]	71.01 ± 14.8 [a,b]

*Results are shown as Mean ± SD, Group I - Healthy males, Group II - Prediabetes males, Group III - Diabetes males, n – number of subjects, FPG- fasting plasma glucose, p <0.05 is considered significant and denoted as **a** when Vs Group1, **b** when Vs Group 2, **c** when Vs Group 3.*

The data was analysed based on groups using ANOVA and the mean values were compared. The mean fasting plasma glucose and serum Insulin were found to be significantly increased in both group II and group III when compared with group I. Also both these parametes were found to be significantly increased in group III when compared with group II. The mean age was comparable between the three groups. Figure 1, 2, & 3 are graphical representation of the same.

Fig. 1: Comparison of Mean Age between the groups.

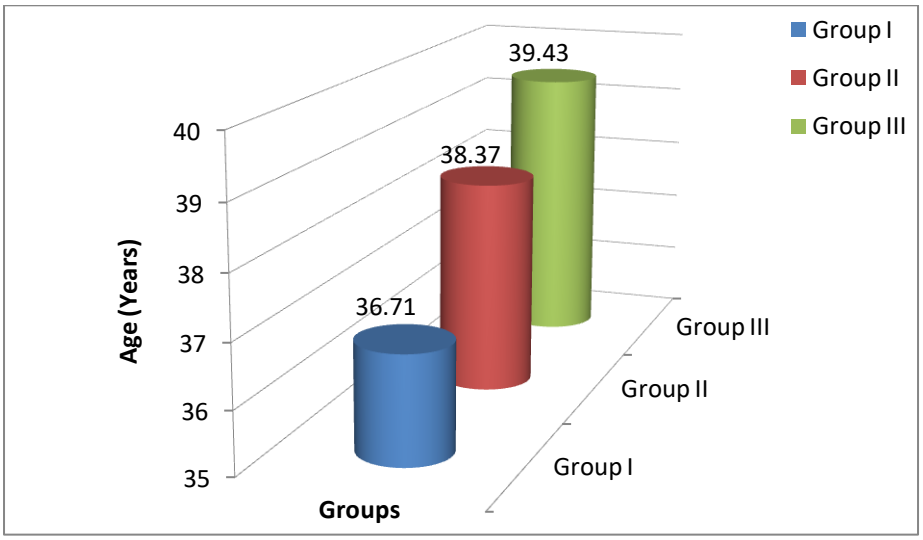


Fig. 2: Comparison of Mean fasting plasma glucose (FPG) between the groups.

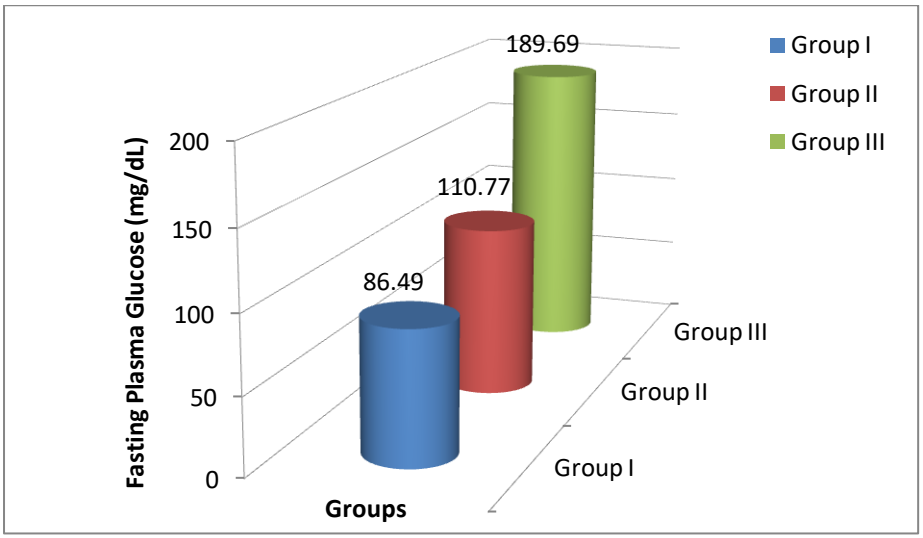


Fig. 3: Comparison of Mean Insulin levels between the groups.

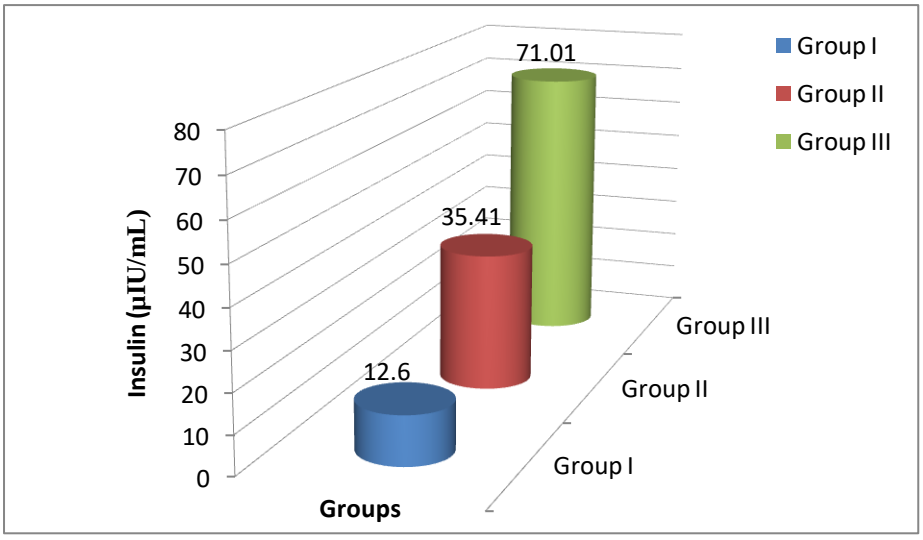


Table 2: Comparison of HPT (Hypothalamus - Pituitary – Thyroid) axis hormones between the groups.

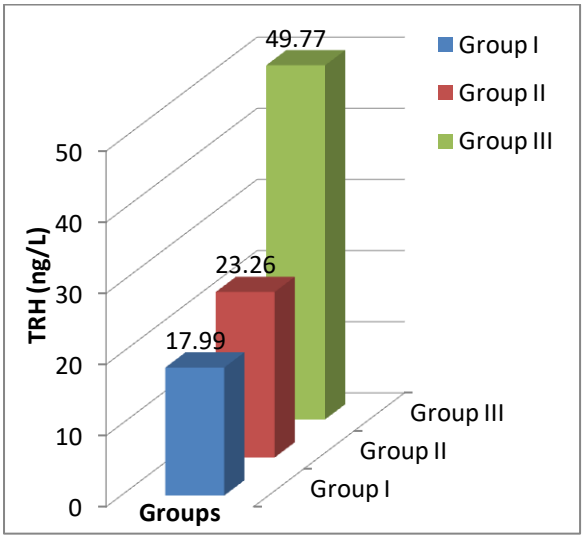
Variable	Group I (n=35)	Group II (n=35)	Group III (n=35)
TRH (ng/L)	17.99 ± 4.56 [b,c]	23.26 ± 3.15 [a,c]	49.77 ± 10.84 [a,b]
TSH (µIU/mL)	1.99 ± 1.14 [b,c]	4.38 ± 0.56 [a,c]	7.74 ± 3.04 [a,b]
Free T3 (pg/mL)	2.84 ± 0.83 [b,c]	2.37 ± 0.68 [a]	2.16 ± 0.48 [a]
Free T4 (ng/dL)	1.41 ± 0.37 [b,c]	1.17 ± 0.29 [a]	1.16 ± 0.22 [a]

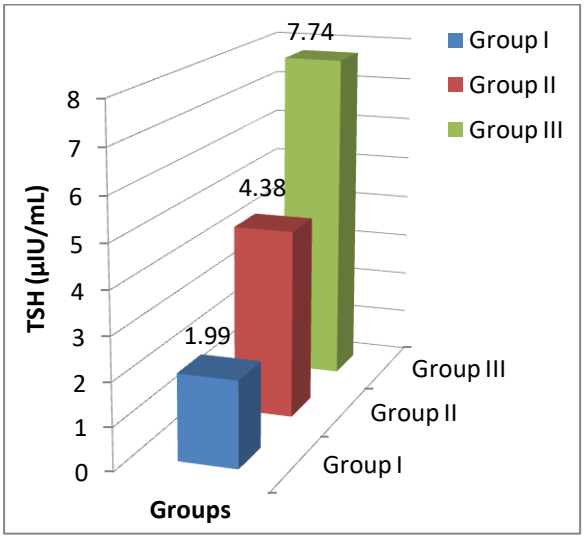
*Results are shown as Mean ± SD, Group I - Healthy males, Group II - Prediabetes males, Group III - Diabetes males, n – number of subjects, TRH - Thyrotrophin releasing hormone, TSH – Thyroid stimulating hormone, T3 – Triiodothyronine, T4 – Tetraiodothyronine, p <0.05 is considered significant and denoted as **a** when Vs Group1, **b** when Vs Group 2, **c** when Vs Group 3.*

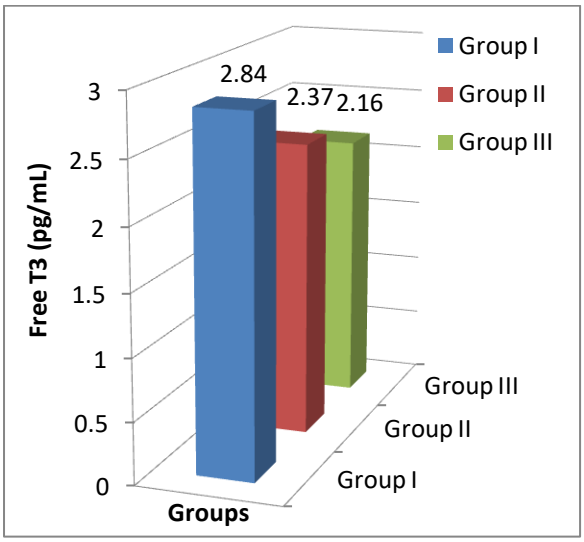
The data was analyzed based on groups using ANOVA and the mean values were compared. The mean thyrotrophin releasing hormone (TRH) and thyroid stimulating hormone (TSH) were found to be significantly increased in both group II and group III when compared with group I. Also both these parametes were found to be significantly increased in group III when compared with group II.

While triiodothyronine (T3) and tetraiodothyronine (T4) were found to be significantly decreased in both group II and group III when compared with group I. There was no significant difference between group II and group III for the mean values of triiodothyronine (T3) and tetraiodothyronine (T4). Figure 4 is graphical representation of the same.

Fig 4: Comparison of HPT (Hypothalamus - Pituitary – Thyroid) axis hormones between the groups.







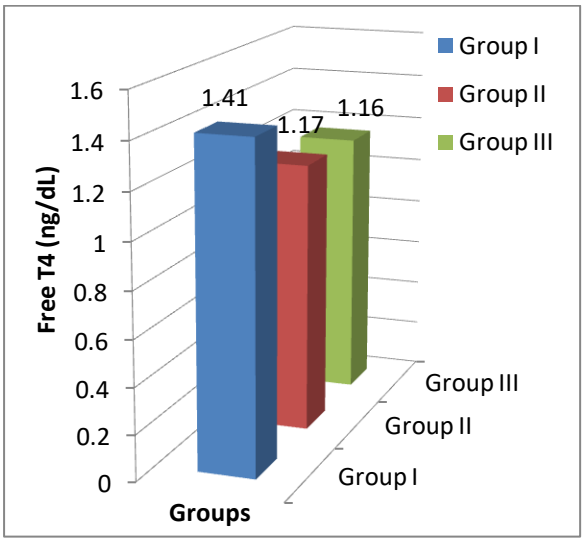


Table 3: Comparison of HPA (Hypothalamus - Pituitary – Adrenal) axis hormones between the groups.

Variable	Group I (n=35)	Group II (n=35)	Group III (n=35)
CRH (ng/mL)	17.8 ± 6.08 [b,c]	11.47 ± 1.58 [a,c]	42.67 ± 8.42 [a,b]
ACTH (ng/L)	18.25 ± 6.11 [c]	17.58 ± 4.37 [c]	44.46 ± 9.52 [a,b]
Cortisol (ng/mL)	131.01 ± 37.3 [c]	140.27 ± 28.82 [c]	183.7 ± 38.84 [a,b]

*Results are shown as Mean ± SD, Group I - Healthy males, Group II - Prediabetes males, Group III - Diabetes males, n – number of subjects, CRH- Corticotrophin releasing hormone, ACTH – Adrenocorticotrophic hormone, p <0.05 is denoted as **a** when Vs Group1, **b** when Vs Group 2, **c** when Vs Group 3.*

The data was analyzed based on groups using ANOVA and the mean values were compared. The mean corticotrophin releasing hormone (CRH) was found to be significantly increased in both group II and group III when compared with group I. Also it was found to be significantly increased in group III when compared with group II.

While adrenocorticotrophic hormone (ACTH) and cortisol were found to be significantly increased in group III when compared with group I and group II. There was no significant difference between group I and group II for the mean values of adrenocorticotrophic hormone (ACTH) and cortisol. Figure 5 is graphical representation of the same.

Fig.5: Comparison of HPA (Hypothalamus - Pituitary – Adrenal) axis hormones between the groups.

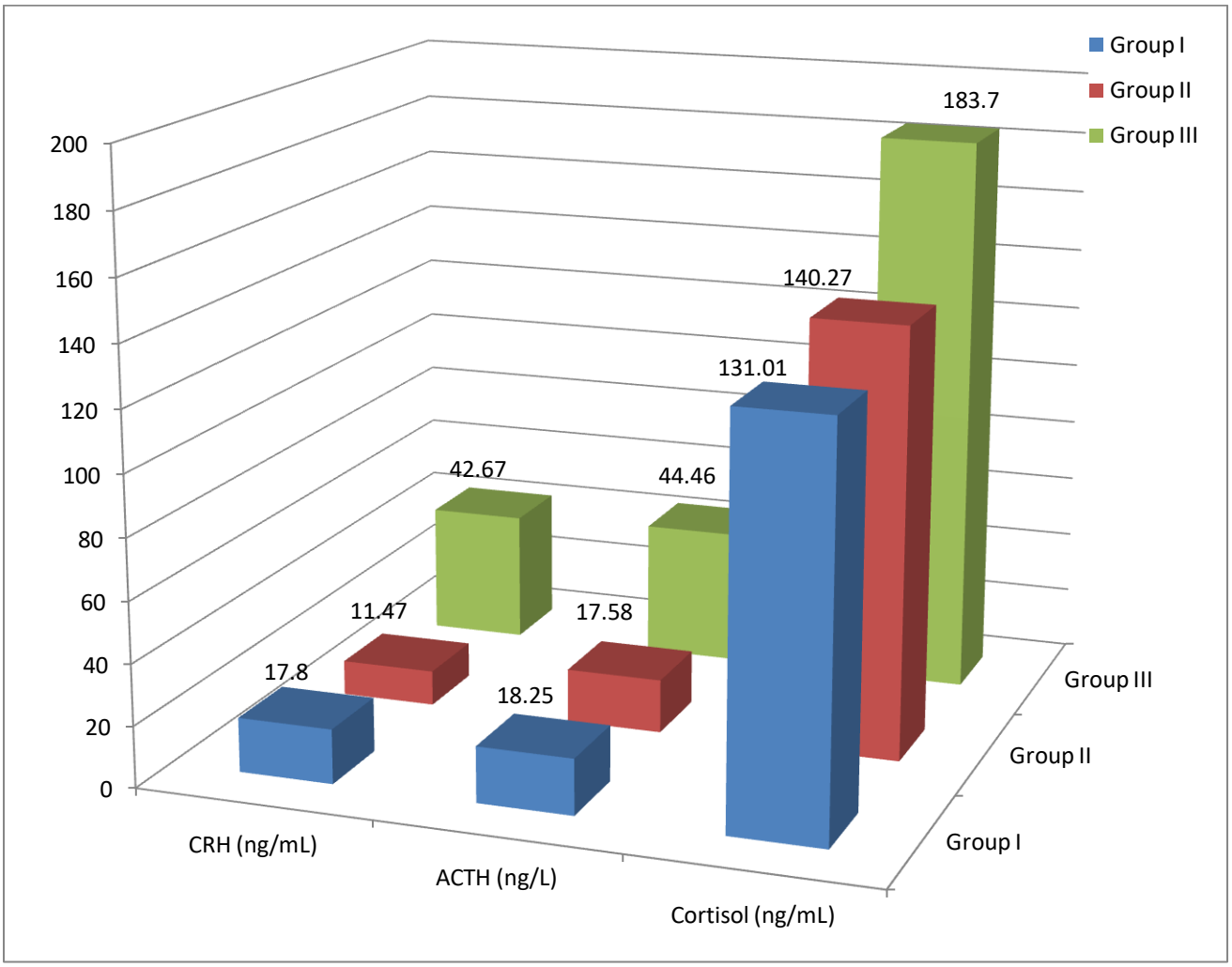


Table 4: Comparison of HPG (Hypothalamus - Pituitary – Gonadal) axis hormones between the groups.

Variable	Group I (n=35)	Group II (n=35)	Group III (n=35)
GnRH (mg/L)	34.22 ± 7.65 [c]	40.24 ± 5.08 [c]	58.28 ± 21.98 [a,b]
FSH (mIU/mL)	9.76 ± 2.59	10.67 ± 2.0	10.86 ± 1.95
LH (mIU/mL)	5.2 ± 1.39 [b,c]	6.58 ± 1.26 [a]	6.67 ± 2.44 [a]
Testosterone (ng/mL)	6.54 ± 1.83 [b,c]	2.69 ± 0.38 [a,c]	0.77 ± 0.36 [a,b]

*Results are shown as Mean ± SD, Group I - Healthy males, Group II - Prediabetes males, Group III - Diabetes males, n – number of subjects, GnRH- Gonadotrophic releasing hormone, FSH - Follicle stimulating hormone, LH – Leutenising hormone, p <0.05 is denoted as **a** when Vs Group1, **b** when Vs Group 2, **c** when Vs Group 3.*

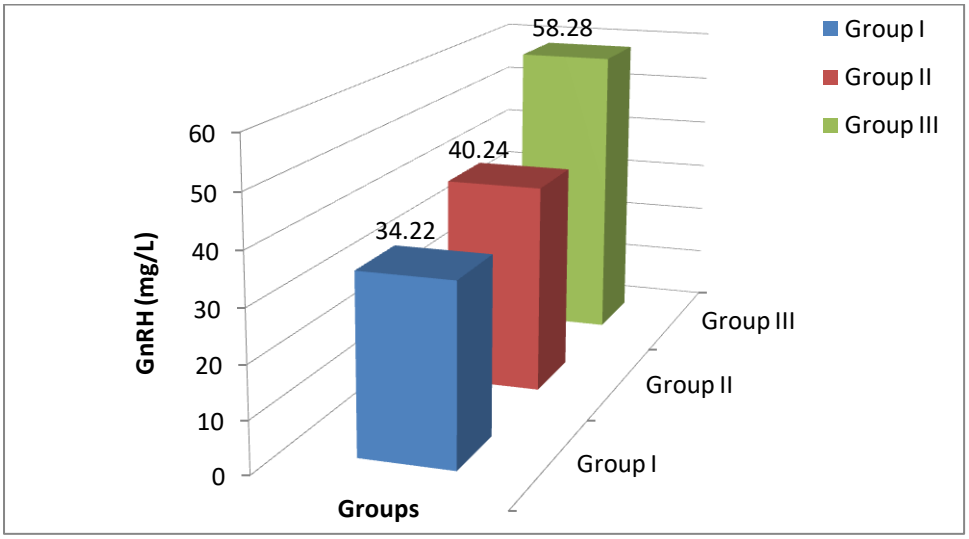
The data was analyzed based on groups using ANOVA and the mean values were compared. The mean gonadotrophic releasing hormone (GnRH) was found to be significantly increased in group III when compared with group I and group II. There was no significant difference between group I and group II for the mean values of gonadotrophic releasing hormone.

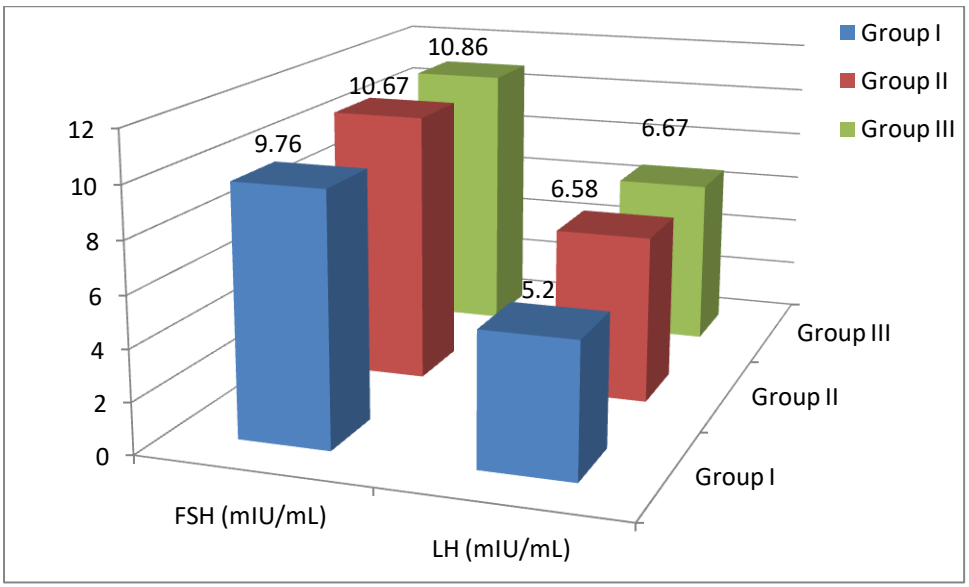
No significant difference was observed in the mean values of follicle stimulating hormone (FSH) between the groups.

Leutenising hormone (LH) mean values were found to be significantly increased in both group II and group III when compared with group I, but no significant difference was found between group II and group III.

The mean testosterone levels were found to be significantly decreased in both group II and group III when compared with group I. It was also observed to be significantly decreased in group III when compared with group II. Figure 6 is graphical representation of the same.

Fig. 6: Comparison of HPG (Hypothalamus - Pituitary – Gonadal) axis hormones between the groups.





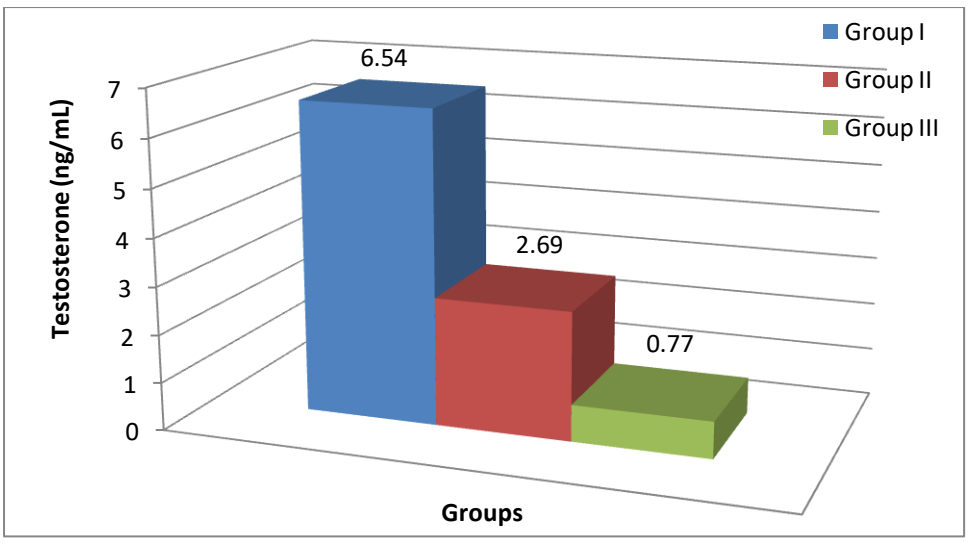


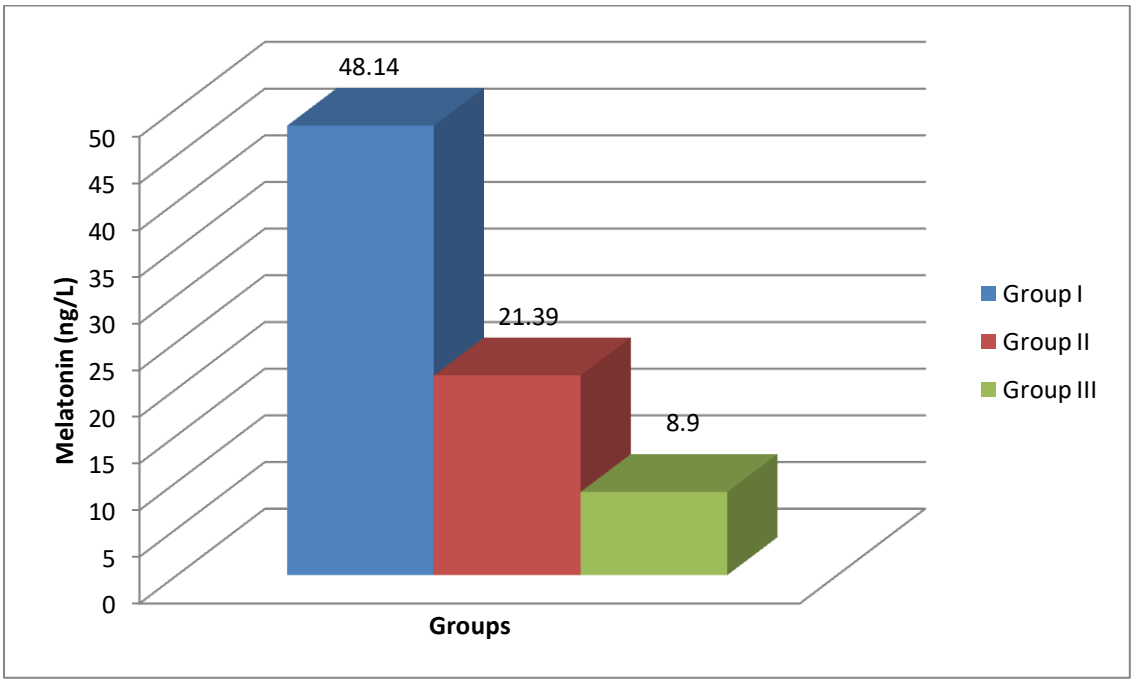
Table 5: Comparison of Pineal gland hormone between the groups.

Variable	Group I (n=35)	Group II (n=35)	Group III (n=35)
Melatonin (ng/L)	48.14 ± 16.57 [b,c]	21.39 ± 3.9 [a,c]	8.9 ± 2.97 [a,b]

Results are shown as Mean ± SD, Group I - Healthy males, Group II - Prediabetes males, Group III - Diabetes males, n – number of subjects, p <0.05 is denoted as ***a*** *when Vs Group1,* ***b*** *when Vs Group 2,* ***c*** *when Vs Group 3.*

The mean melatonin levels were found to be significantly decreased in both group II and group III when compared with group I. It was also observed to be significantly decreased in group III when compared with group II. Figure 7 is graphical representation of the same.

Figure 7: Comparison of Pineal gland hormone between the groups.



Results according to the objective of the study

2. **Secondary objective :** To correlate HPT, HPA, HPG axis hormone and pineal gland hormone levels with FPG and Insulin in all the groups.

Table 6: Correlation of HPT (Hypothalamus - Pituitary – Thyroid) axis hormones with FPG and Fasting Insulin between the groups.

Parameter			TRH (ng/L)	TSH (µIU/mL)	Free T3 (pg/mL)	Free T4 (ng/dL)
FPG (mg/dl)	Group I	r value	-0.271	0.214	-0.206	-0.137
		p value	0.116	0.217	0.236	0.431
	Group II	r value	-0.131	- 0.176	-0.165	-0.142
		p value	0.454	0.311	0.342	0.416
	Group III	r value	0.028	0.056	0.011	0.199
		p value	0.872	0.749	0.948	0.252
Fasting Insulin (µIU/mL)	Group I	r value	0.185	-0.325	0.048	-0.123
		p value	0.287	0.057	0.783	0.482
	Group II	r value	**0.473***	0.262	0.148	0.257
		p value	**0.004**	0.128	0.396	0.137
	Group III	r value	0.055	**0.681***	**0.614***	0.174
		p value	0.756	**0.000**	**0.000**	0.317

*Group I - Healthy males, Group II - Prediabetes males, Group III - Diabetes males, FPG- fasting plasma glucose, TRH - Thyrotrophin releasing hormone, TSH – Thyroid stimulating hormone, T3 – Triiodothyronine, T4 – Tetraiodothyronine, , r value – Pearson's correlation, *p <0.05 is significant*

When Pearson's correlation was applied for FPG and fasting insulin versus serum thyrotrophin releasing hormone (TRH), thyroid stimulating hormone (TSH), triiodothyronine (T3) and tetraiodothyronine (T4) in all the three groups. No significant correlation was seen of TRH with FPG in all the three groups, but TRH showed significant correlation with Fasting insulin in only group II (r=0.473, p = 0.004)

In case of TSH, similar to TRH, no significant correlation was observed with FPG in all the three groups, in contrast to TRH, TSH showed significant correlation with fasting insulin with group III.

Free T3 and Free T4 does not have any significant correlation with FPG in all the three groups. But fasting insulin shows significant correlation with free T3 only in group III and no correlation with free T4 in all the three groups.

Table 7: Correlation of HPA (Hypothalamus - Pituitary – Adrenal) axis hormones with FPG and Fasting Insulin between the groups.

Parameter			CRH (ng/mL)	ACTH (ng/L)	Cortisol (ng/mL)
FPG (mg/dl)	Group I	r value	0.085	-0.140	-0.025
		p value	0.629	0.423	0.885
	Group II	r value	-0.129	0.002	-0.082
		p value	0.462	0.99	0.641
	Group III	r value	0.037	0.168	-0.100
		p value	0.835	0.336	0.567
Fasting Insulin (µIU/mL)	Group I	r value	0.265	0.017	0.087
		p value	0.124	0.922	0.617
	Group II	r value	**-0.381***	-0.303	**0.688***
		p value	**0.024**	0.077	**0.000**
	Group III	r value	0.074	-.0291	0.098
		p value	0.672	0.090	0.577

*Group I - Healthy males, Group II - Prediabetes males, Group III - Diabetes males, FPG- fasting plasma glucose, CRH- Corticotrophin releasing hormone, ACTH – Adrenocorticotrophic hormone r value – Pearson's correlation, *p <0.05 is significant*

When Pearson's correlation was applied for FPG and fasting insulin versus serum corticotrophin releasing hormone (CRH), adrenocorticotrophic hormone (ACTH) and cortisol in all the three groups. No significant correlation was seen of CRH with FPG in all the three groups, but CRH showed significant correlation with Fasting insulin in only group II (r=-0.381, p = 0.024)

In case of ACTH, no significant correlation was observed with FPG and fasting insulin levels in all the three groups.

Serum cortisol does not have any significant correlation with FPG in all the three groups. But has significant correlation with fasting insulin in group II and not with group I and group III.

Table 8: Correlation of HPG (Hypothalamus - Pituitary – Gonadal) axis hormones with FPG and Fasting Insulin between the groups.

Parameter			GnRH (mg/L)	FSH (mIU/mL)	LH (mIU/mL)	Testosterone (ng/mL)
FPG (mg/dl)	Group I	r value	-0.243	0.051	0.045	-0.140
		p value	0.159	0.771	0.798	0.424
	Group II	r value	**-0.352***	0.079	0.319	0.269
		p value	**0.038**	0.654	0.062	0.118
	Group III	r value	0.231	-0.184	0.082	-0.258
		p value	0.183	0.289	0.638	0.135
Fasting Insulin (µIU/mL)	Group I	r value	0.164	0.042	0.005	-0.011
		p value	0.345	0.809	0.977	0.951
	Group II	r value	**0.447***	-0.056	0.002	-0.016
		p value	**0.007**	0.751	0.99	0.929
	Group III	r value	-0.124	0.131	-0.102	0.162
		p value	0.478	0.453	0.559	0.354

*Group I - Healthy males, Group II - Prediabetes males, Group III - Diabetes males, FPG- fasting plasma glucose, GnRH- Gonadotrophic releasing hormone, FSH - Follicle stimulating hormone, LH – Leutenising hormone, r value – Pearson's correlation, *p <0.05 is significant*

When Pearson's correlation was applied for FPG and fasting insulin versus serum gonadotrophic releasing hormone (GnRH), follicle stimulating hormone (FSH), Leutenising hormone (LH) and testosterone in all the three groups. GnRH showed significant correlation with FPG (r --0.352*,p - 0.038) and fasting insulin (r - 0.447*, p - 0.007) in group II only. No significant correlation was seen of GnRH with group I and group III.

In case of FSH and LH, no significant correlation was observed with FPG and fasting insulin levels in all the three groups.

Serum testosterone also does not present with any significant correlation with FPG and fasting insulin levels in all the three groups.

Table 9: Correlation of Pineal gland hormone with FPG and Fasting Insulin between the groups.

Parameter			Melatonin (ng/L)
FPG (mg/dl)	Group I	r value	0.028
		p value	0.872
	Group II	r value	0.184
		p value	0.289
	Group III	r value	0.039
		p value	0.823
Fasting Insulin (µIU/mL)	Group I	r value	0.198
		p value	0.254
	Group II	r value	**-0.459***
		p value	**0.006**
	Group III	r value	-0.089
		p value	0.612

*Group I - Healthy males, Group II - Prediabetes males, Group III - Diabetes males, FPG- fasting plasma glucose, r value – Pearson's correlation, *p <0.05 is significant*

When Pearson's correlation was applied for FPG and fasting insulin versus serum melatonin in all the three groups, Melatonin showed significant correlation with fasting insulin (r – - 0.459*, p - 0.006) in group II only. No significant correlation was seen of melatonin with group I and group III fasting insulin. Also there was no significant correlation of melatonin with FPG in all the three groups.