

CHAPTER IV

RESULTS

Part I: Subject characteristics

Thirty five 11778 LHON patients with chronic phase were participated in this study. Patients were divided into two group, placebo and curcuminoid group. Three patients in curcuminoid group were dropped from the study because they did not follow up at month 3 or 6 of treatment. The physical characteristics of LHON patients were presented in table 4.1. The other diseases have been found in some patients. In placebo group, four patients have one of the following diseases: hypertension, peptic ulcer and hemorrhoid. Whereas, in curcuminoid group found two cases of allergy, one case of anemia and one case of hypertension with hyperlipidemia. The liver enzyme, serum glutamic oxalocetic transaminase (SGOT) and/or serum glutamic pyruvic transaminase (SGPT) level at baseline in patients were higher than normal range (see appendices). The normal range of SGOT and SGPT level are 0-37 U/L and 0-40 U/L, respectively. In placebo group, only one case had elevated SGPT level (45 U/L). In curcuminoid group, we found both elevated enzyme in three patients, elevated SGOT level in 2 patients and elevated SGPT level in 2 patients. These eight patients were asked to stop smoking and drinking alcohol. Then the liver enzyme were monitored at 3 and 6 month and found that the enzyme of all cases were in normal range.

Table 4.1 Physical characteristics of LHON patients

| Parameters | Placebo (n=15) | Curcuminoid (n=17) |
|--------------------------|------------------------------|------------------------------|
| Age (years) ¹ | 29.3 ± 3.33 (15-52 years) | 33.8 ± 3.95 (12-67 years) |
| Sex (M/F) | 14 M / 1 F | 13 M / 4 F |
| Weight (kg) ¹ | 60.7 ± 1.50 | 53.7 ± 2.25 |
| Height (cm) ¹ | 168.0 ± 2.36 | 161.4 ± 2.07 |
| Smoking | 73.3 % | 23.5 % |
| Alcohol | 73.3 % | 29.4 % |
| Head injury | 60.0 % | 35.3 % |
| Family history | 46.7 % | 82.4 % |

¹ Values represent the mean ± SE.

Part II: Oxidative stress status

1. The Malondialdehyde (MDA) level

Measurement of MDA level is a marker to determine lipid peroxidation products and refer to oxidative stress. The MDA levels in two groups of LHON patients at 0, 3, 6 and 12 month were not significantly difference (Figure 4.1).

In placebo group, levels of MDA in 3, 6, and 12 month were significantly increased from base line (0M) at p value < 0.05 . But in the curcuminoid group, the MDA levels were not significant difference from baseline (Figure 4.1)

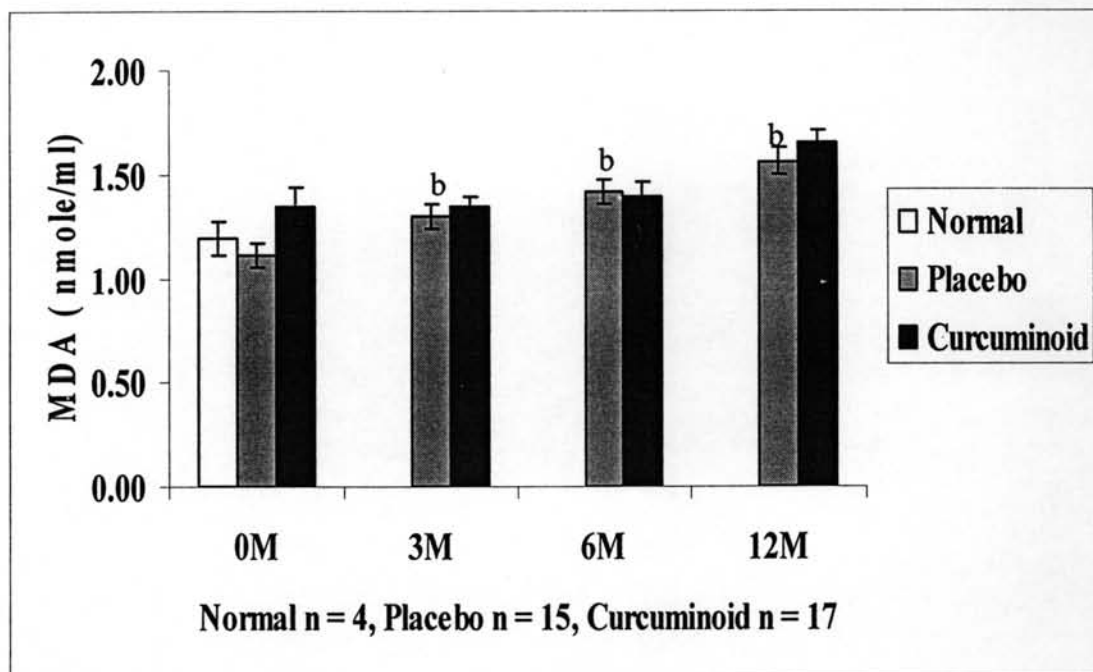


Figure 4.1 The malondialdehyde level in LHON patients. Results are expressed as nmole/ml. Each bar represents the mean value and the vertical lines show the SE.

^b Significant difference from baseline at p value < 0.05 .

2. The correlation between age and MDA level in LHON patients.

The positive correlation between age and MDA level in LHON patients were found with the r-value of 0.41 ($p < 0.05$) (Figure 4.2). Most older patients were female. The average MDA levels at baseline in females were 1.46 ± 0.22 nmole/ml, whereas those in males were 1.19 ± 0.06 nmole/ml.

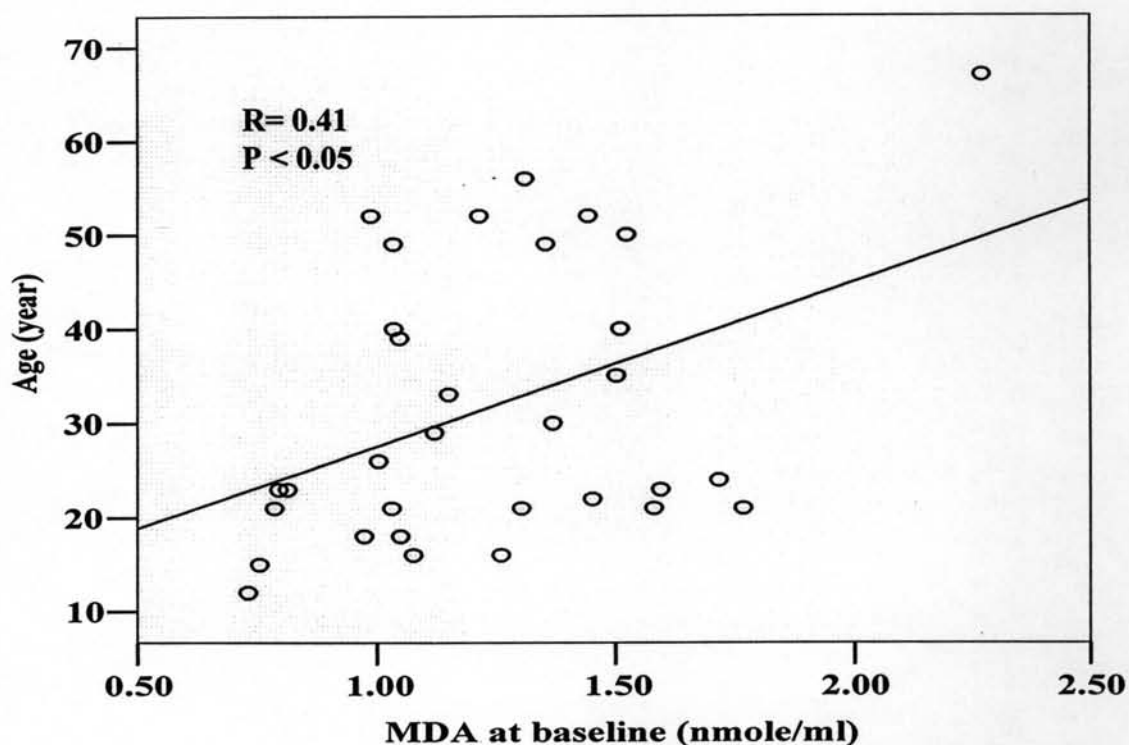


Figure 4.2 The correlation between age and MDA level in LHON patients. The data analyses from thirty two LHON patients.

Part III: Antioxidant enzyme activity

1. Superoxide dismutase (SOD)

The SOD activities were not significant difference between curcuminoid and placebo group (Figure 4.3). Within curcuminoid group, SOD activities at 3 month were decreased 12.6% from baseline (Figure 4.4).

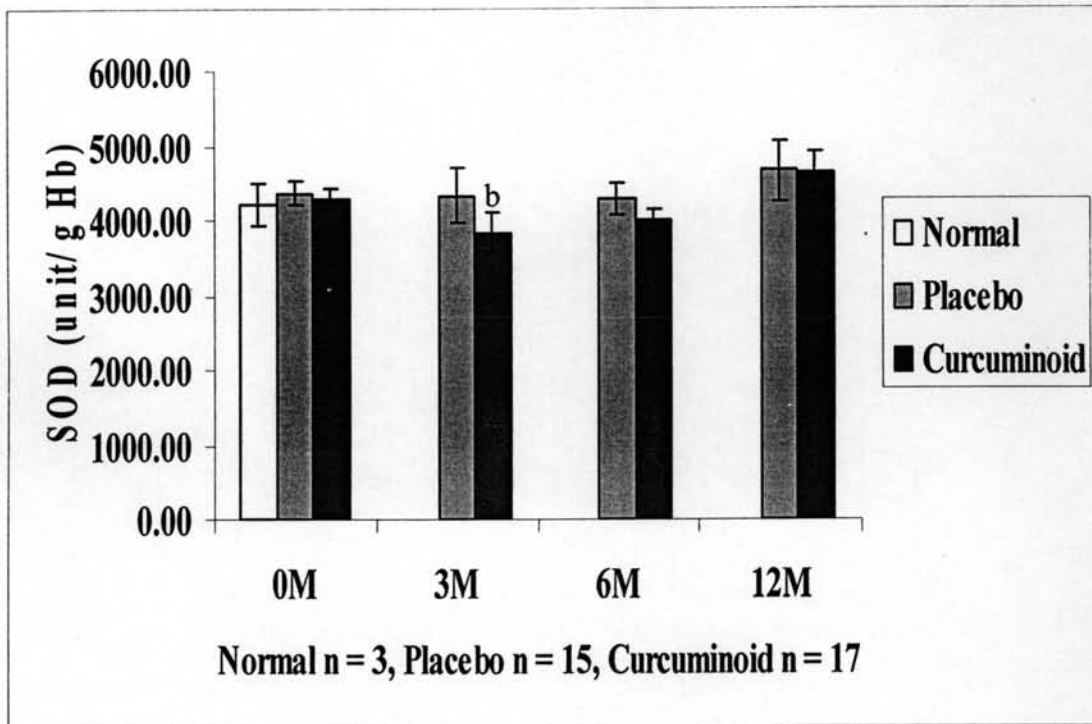


Figure 4.3 Superoxide dismutase enzyme activities in LHON patients. Results are expressed as unit/g Hb. Each bar represents the mean value and the vertical lines show the SE.

^b Significant difference from baseline at p value < 0.05.

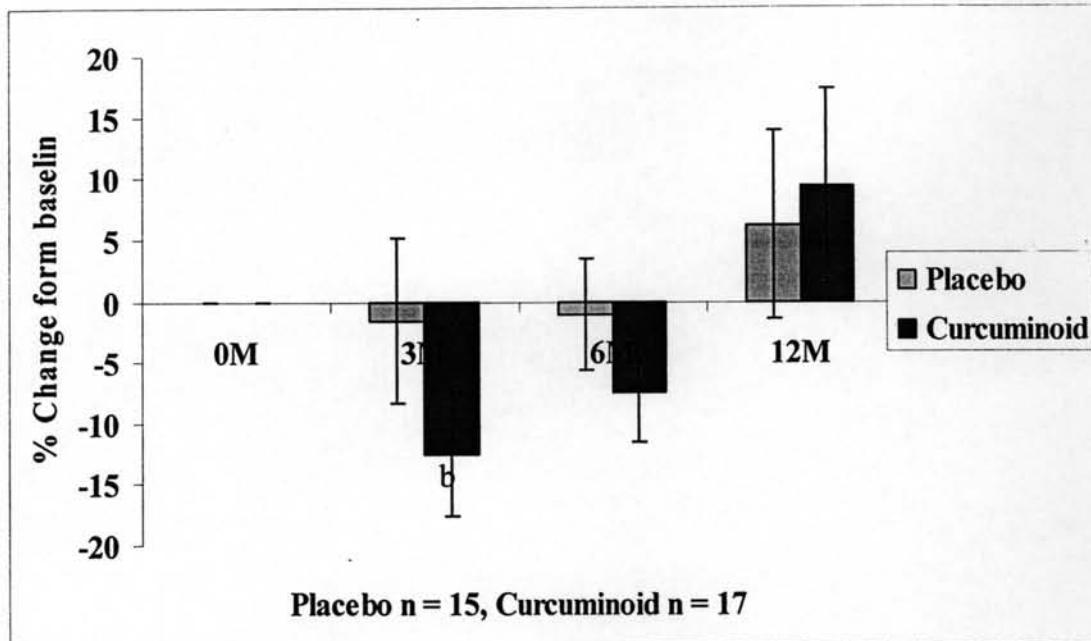


Figure 4.4 Percent change from baseline of superoxide dismutase activities. Each bar represents the mean value and the vertical lines show the SE.

^b Significant difference from baseline at p value < 0.05 .

2. Catalase enzyme (CAT)

The catalase activities were not significant difference between curcuminoid and placebo group. However, at 12 month the catalase activities in both groups were increase significantly from baseline at p value < 0.05 (Figure 4.5).

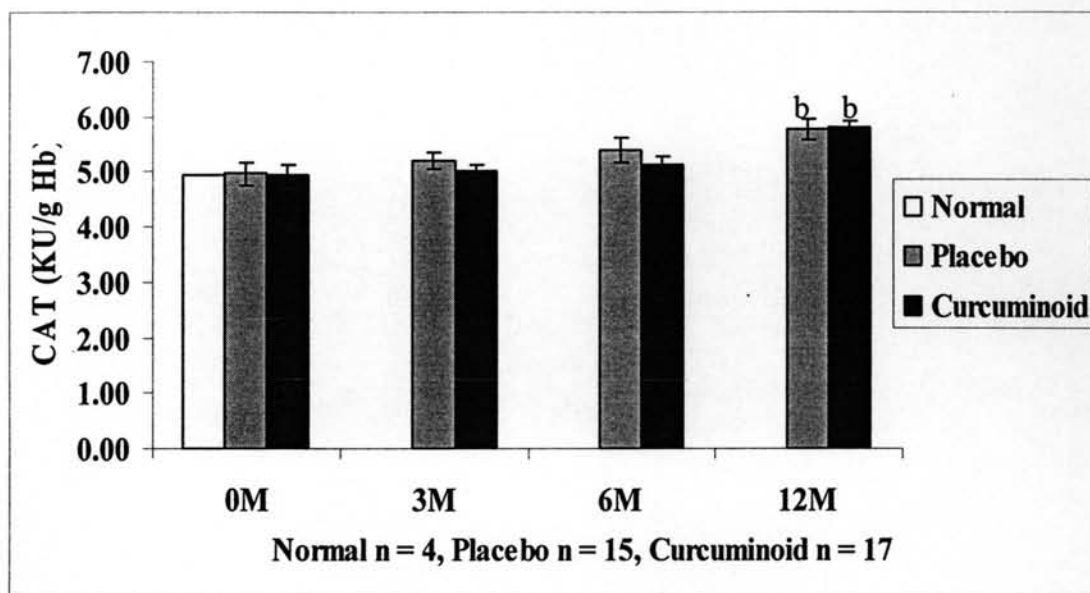


Figure 4.5 The CAT activities in LHON patients. Results are expressed as KU/g Hb. Each bar represents the mean value and the vertical lines show the SE.

^b Significant difference from baseline at p value < 0.05 .

3. Glutathione peroxidase (GPx)

The activities of glutathione peroxidase at 3 month in curcuminoid group were significantly decreased when compare to placebo group ($p < 0.05$) (Figure 4.6). In addition, the GPx activities at 3 and 6 month in curcuminoid group were significantly decreased 24.1% and 12.6% respectively, from baseline (Figure 4.7).

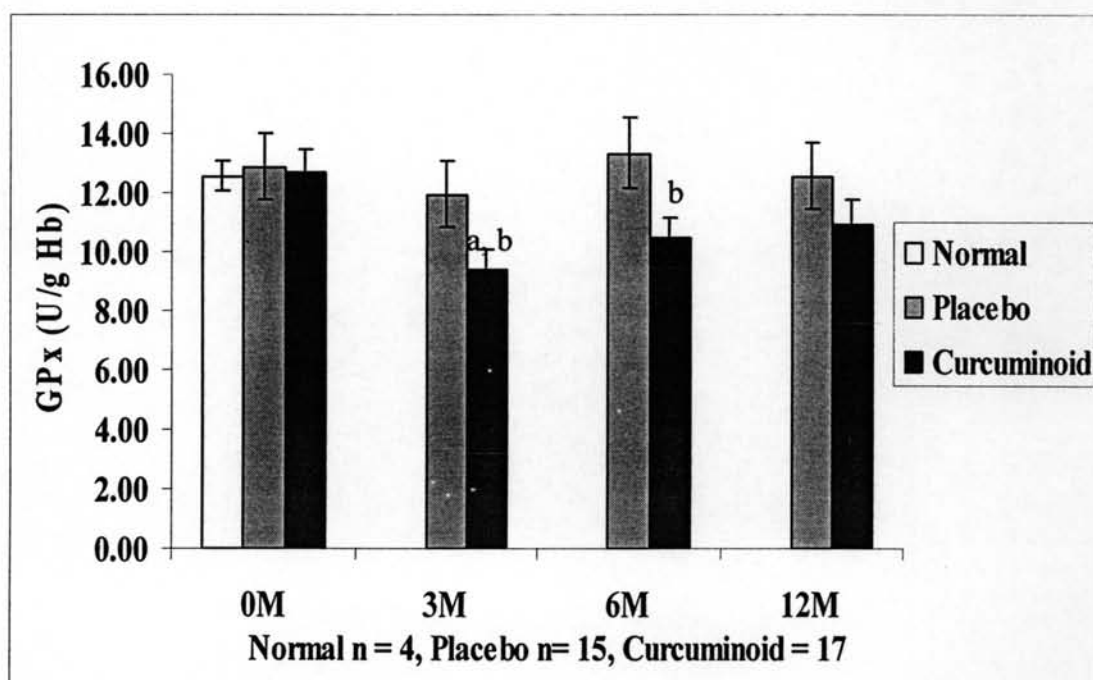


Figure 4.6 Glutathione peroxidase enzyme activities in LHON patients. Results are expressed as U/g Hb. Each bar represents the mean value and the vertical lines show the SE.

^a Significant difference from the placebo at p value < 0.05 .

^b Significant difference from baseline at p value < 0.05 .

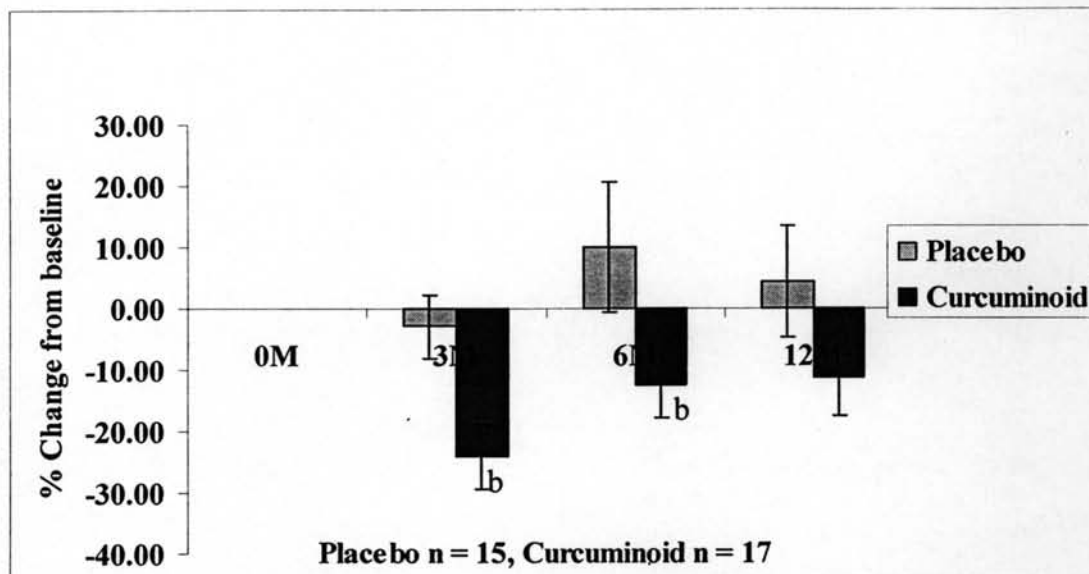


Figure 4.7 Percent change from baseline of glutathione peroxidase activities. Each bar represents the mean value and the vertical lines show the SE.

^b Significant difference from baseline at p value < 0.05 .

4. Total glutathione (GSH)

The total glutathione levels were not significantly difference between curcuminoid and placebo group. However, the level of GSH at 3 and 6 months in both groups were increased significantly from baseline (Figure 4.8).

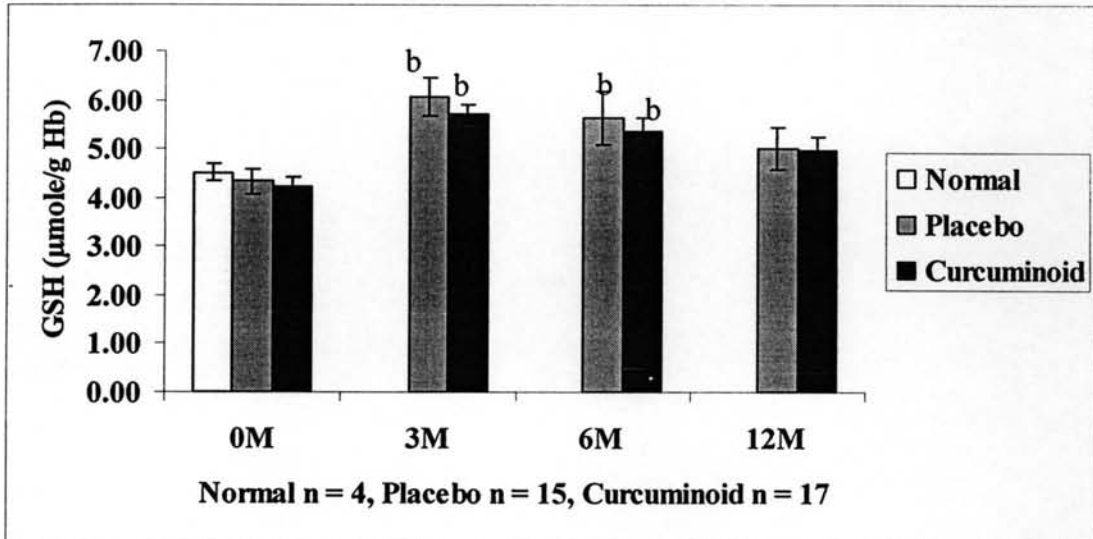


Figure 4.8 The GSH level in LHON patients when treatment with curcuminoid capsule or placebo at 0, 3, 6 and 12 month. Results are expressed as $\mu\text{mole/g Hb}$. Each bar represents the mean value and the vertical lines show the SE.

^b Significant difference from baseline at p value < 0.05 .

5. Oxidized glutathione (GSSG)

The amounts of oxidized glutathione were not significant difference between curcuminoid and placebo group. In placebo group, oxidized glutathione levels at 6 month were significant higher than those at base line. In curcuminoid group, oxidized glutathione levels at 12 month were significant higher than those at baseline too (Figure 4.9).

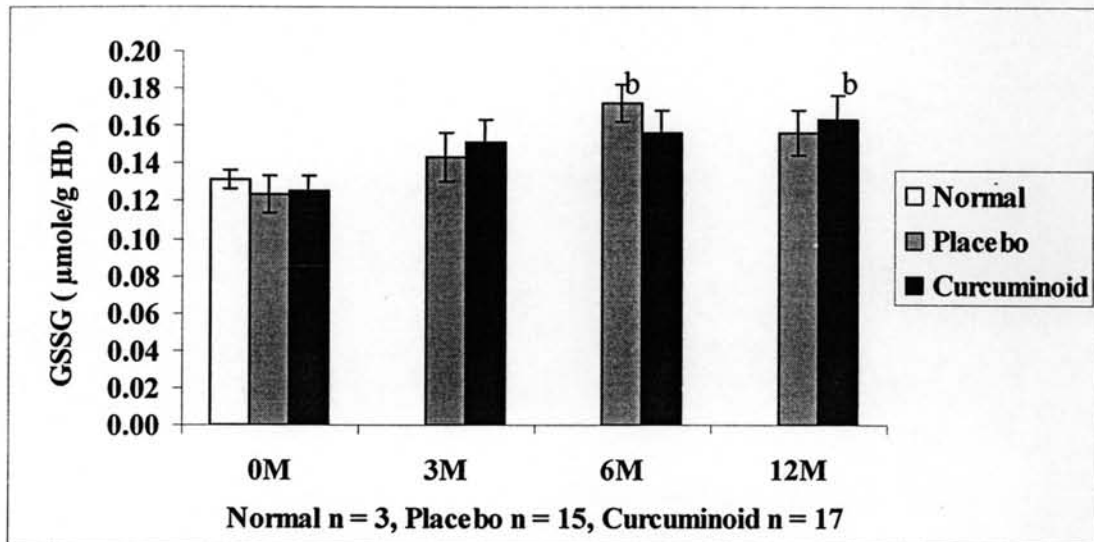


Figure 4.9 The GSSG level in LHON patients. Results are expressed as $\mu\text{mole/g Hb}$. Each bar represents the mean value and the vertical lines show the SE.

^b Significant difference from baseline at p value < 0.05 .

6. The percentage of oxidized glutathione (GSSG) with respect to total glutathione (GSH)

There was not significant difference in percentage of oxidized glutathione (GSSG) with respect to total glutathione (GSH) between placebo and curcuminoid group (Figure 4.10).

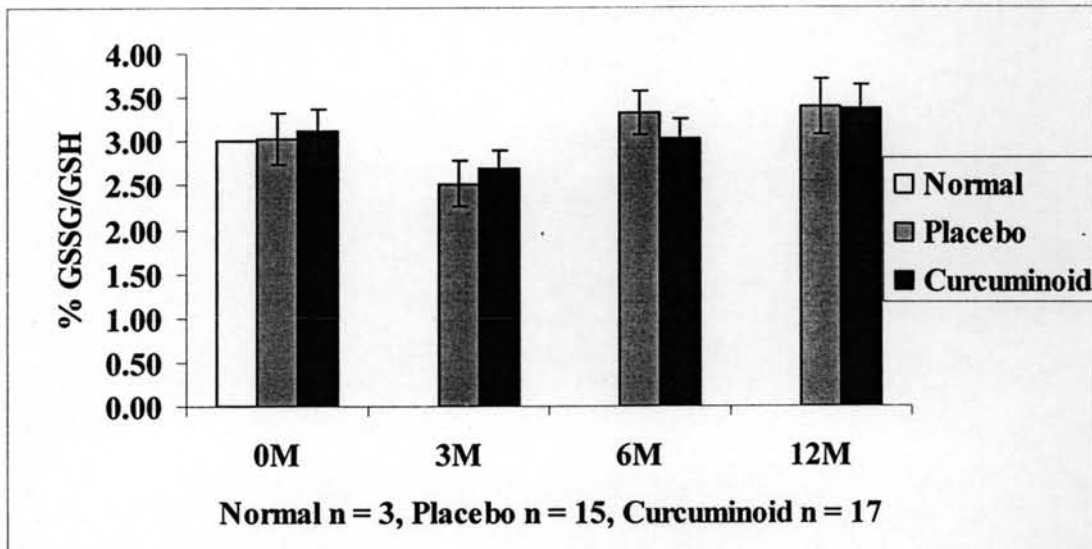
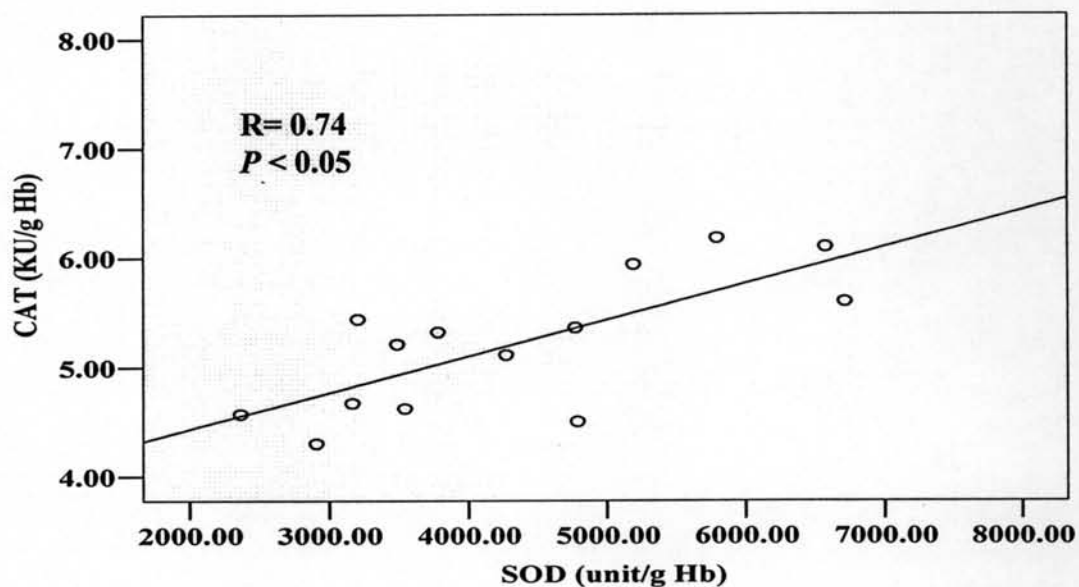
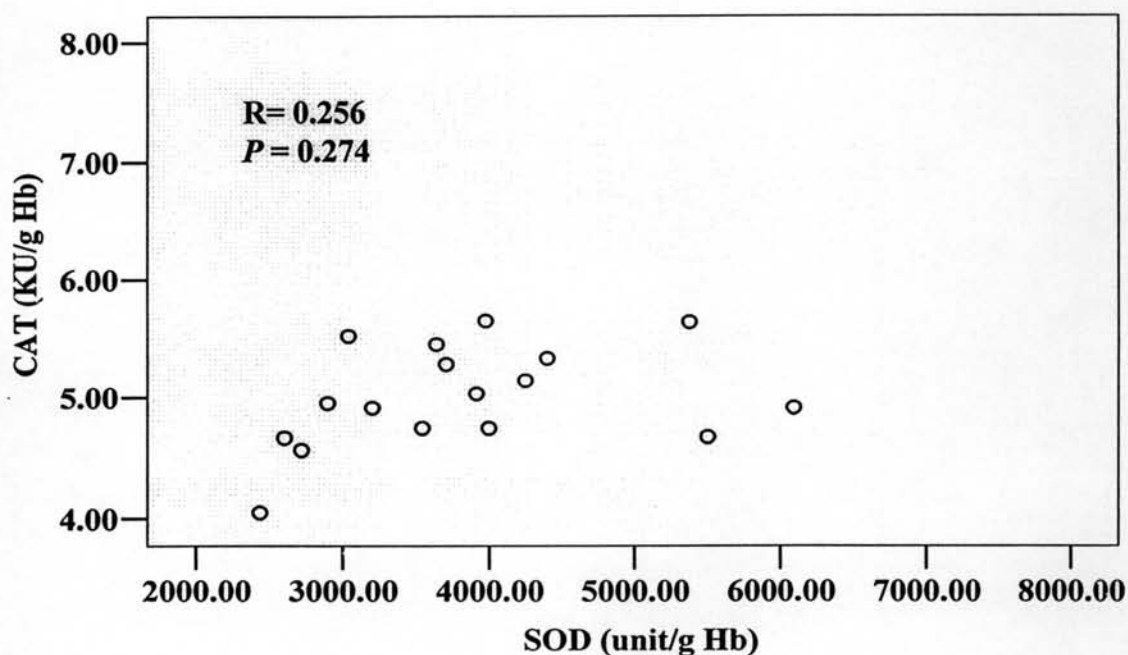
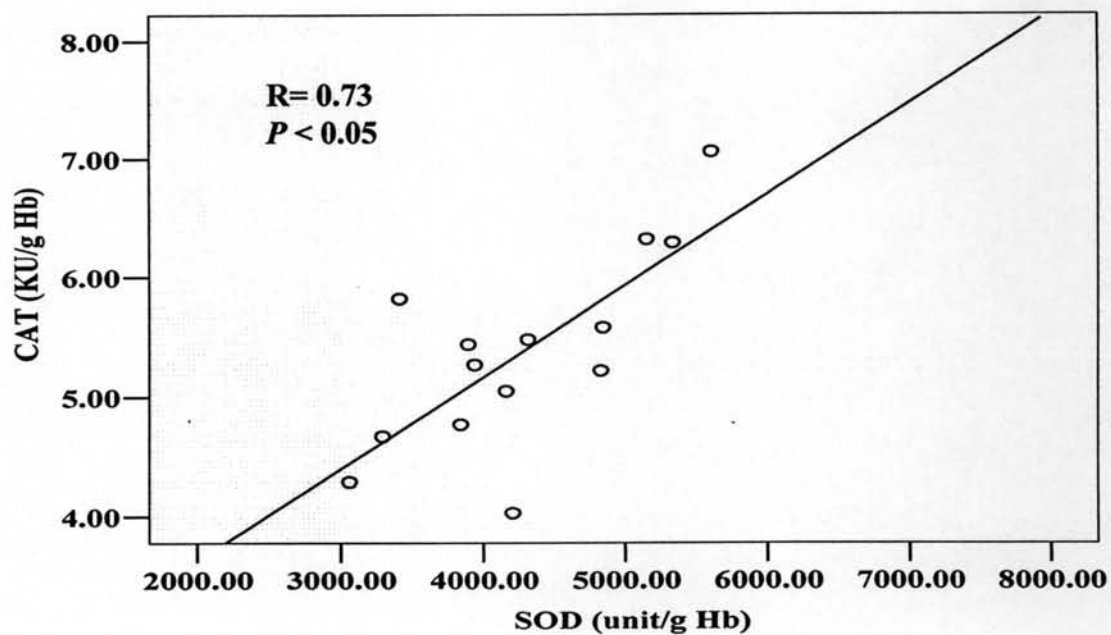
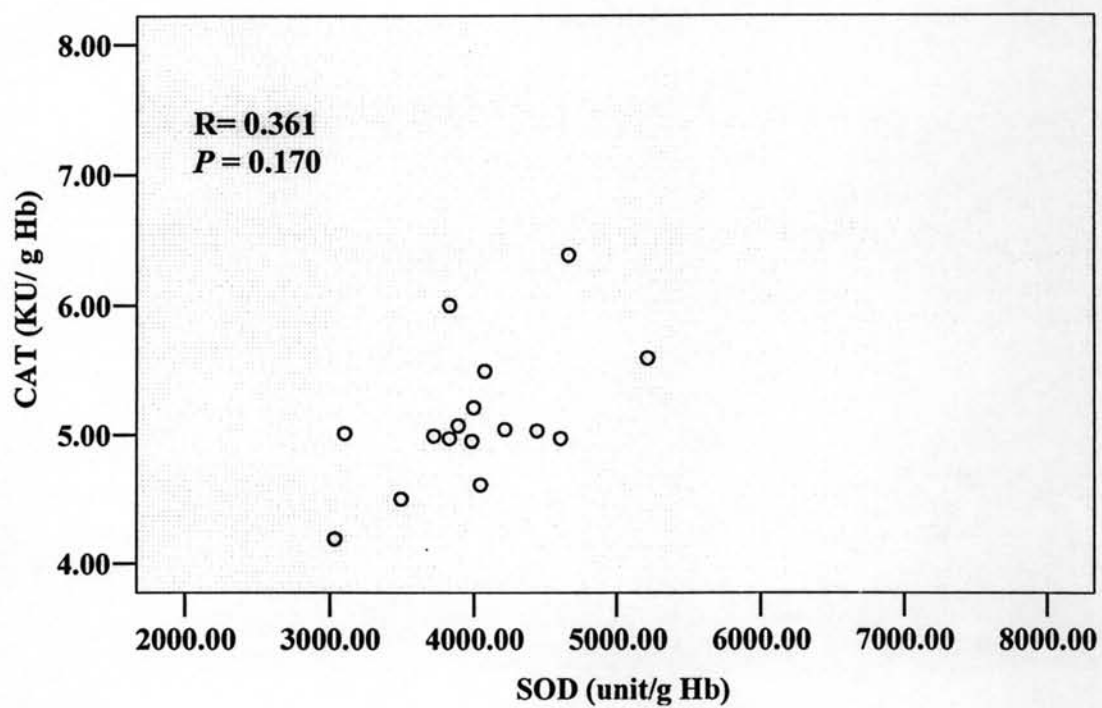


Figure 4.10 The percentage of oxidized glutathione (GSSG) with respect to total glutathione (GSH) at 0, 3, 6 and 12 month. Each bar represents the mean value and the vertical lines show the SE.

Part IV: The correlation**1. The correlation between SOD activities and CAT activities**

The positive correlation between level of SOD activities and CAT activities in placebo group at 3, 6 and 12 month were found with the r -value of 0.74, 0.73 and 0.79, respectively ($p < 0.05$). In curcuminoid group, these correlations were not found (Figure 4.11).

(A) 1. Placebo group 3 month**(A) 2. Curcuminoid group 3 month**

(B) 1. Placebo group 6 month.**(B) 2. Curcuminoid group 6 month.**

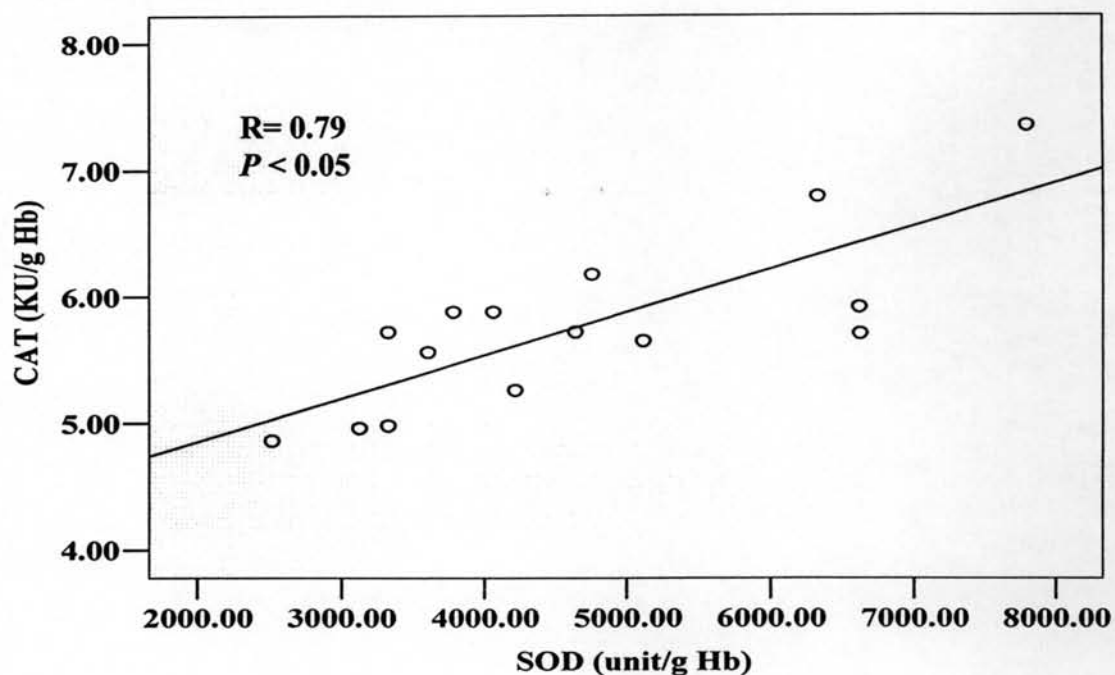
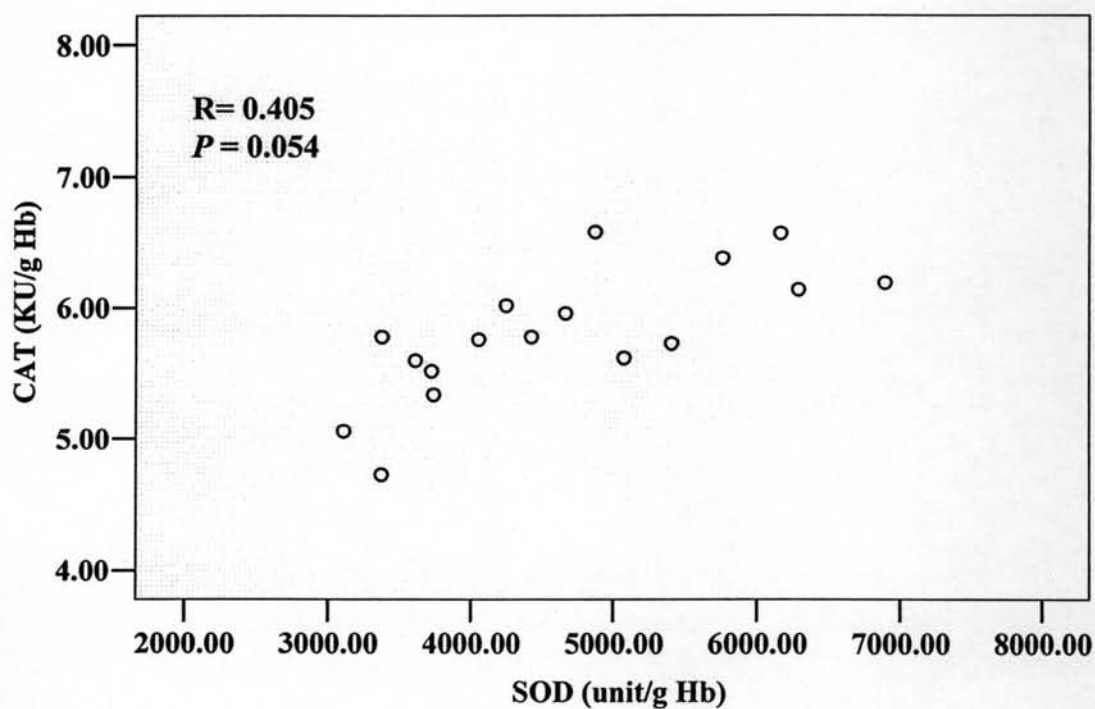
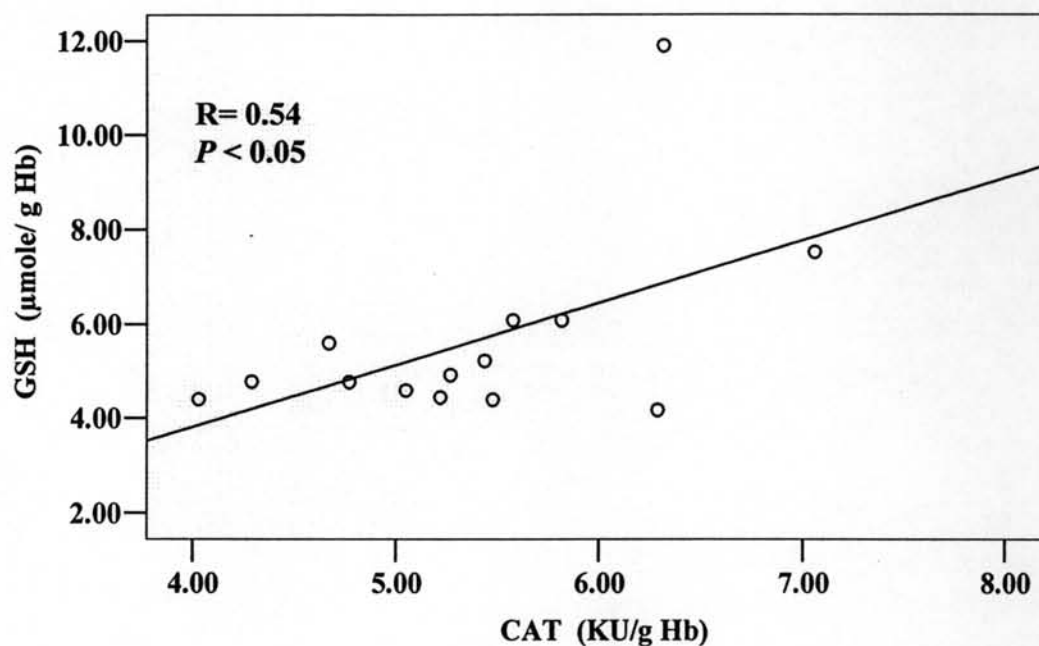
(C) 1. Placebo group 12 month.**(C) 2. Curcuminoid group 12 month.**

Figure 4.11 The correlation between activities of SOD and CAT at (A) 3, (B) 6 and (C) 12 month, respectively. The data analyses from fifteen LHON patients in placebo group and seventeen LHON patients in curcuminoid group.

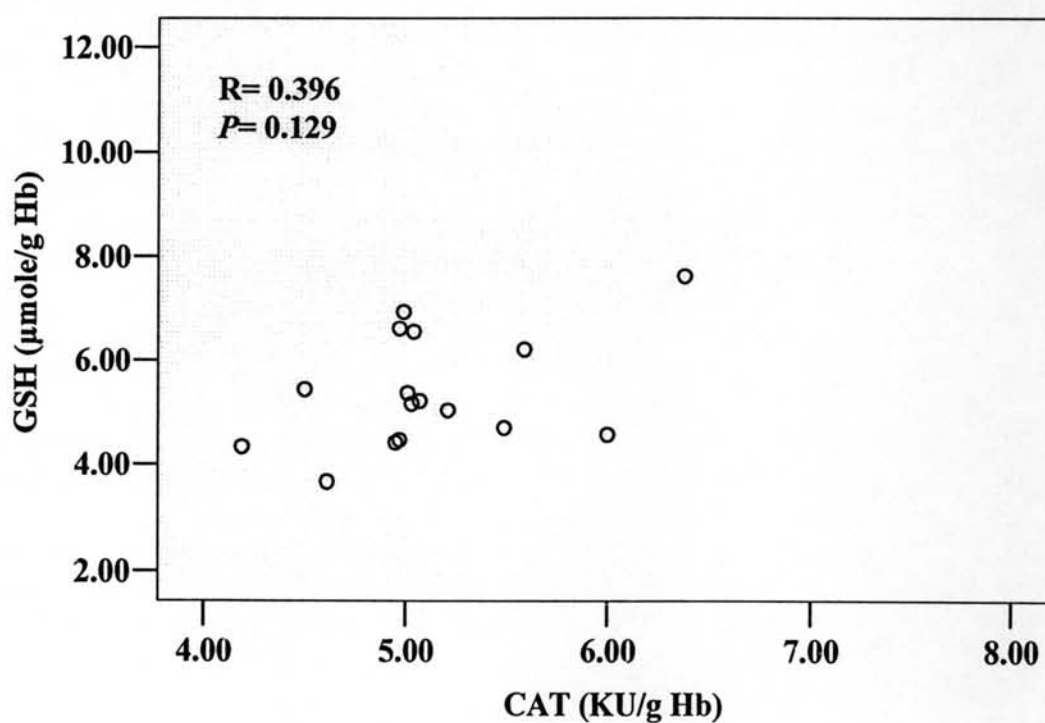
2. The correlation between CAT activities and GSH level

The positive correlation between level of GSH level and CAT activities in placebo group at 6 and 12 month were found with the r -value of 0.54, and 0.75, respectively ($p < 0.05$). In curcuminoid group, these correlations were not found (Figure 4.12).

(A) 1. Placebo group 6 month



(A) 2. Curcuminoid group 6 month



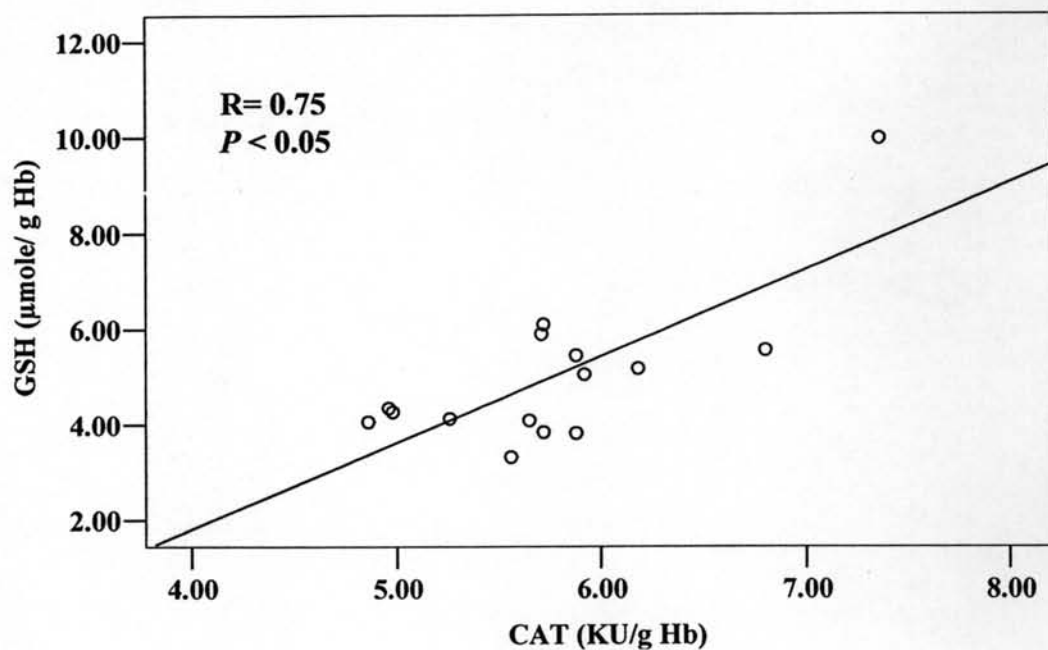
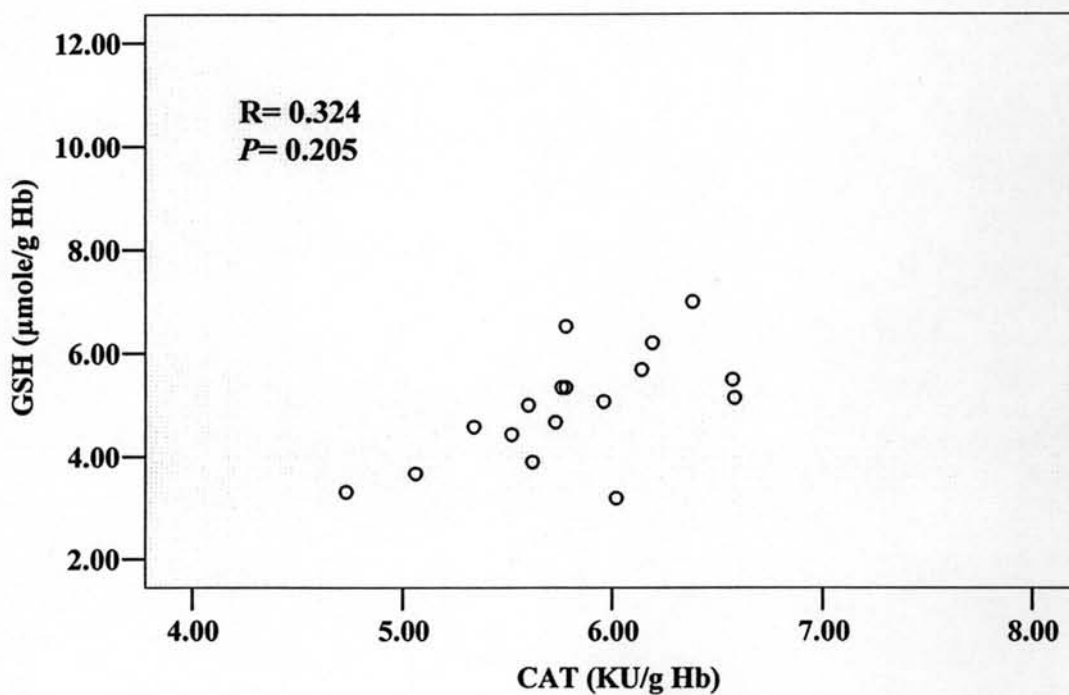
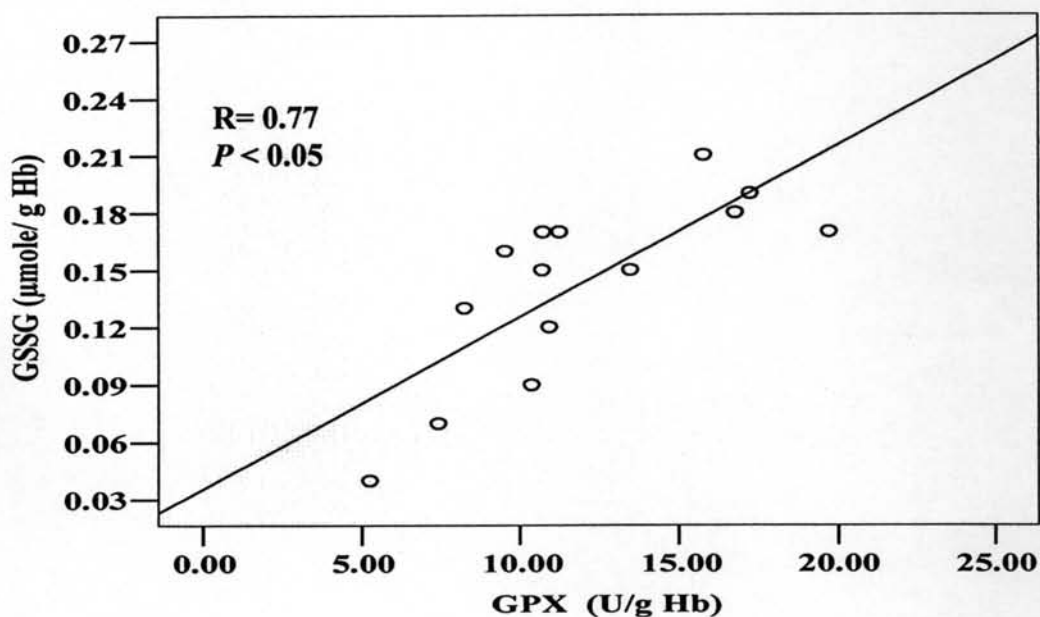
(B) 1. Placebo group 12 month**(B) 2. Curcuminoid group 12 month**

Figure 4.12 The correlation between activities of CAT and GSH at (A) 6 and (B) 12 month, respectively. The data analyses from fifteen LHON patients in placebo group and seventeen LHON patients in curcuminoid group.

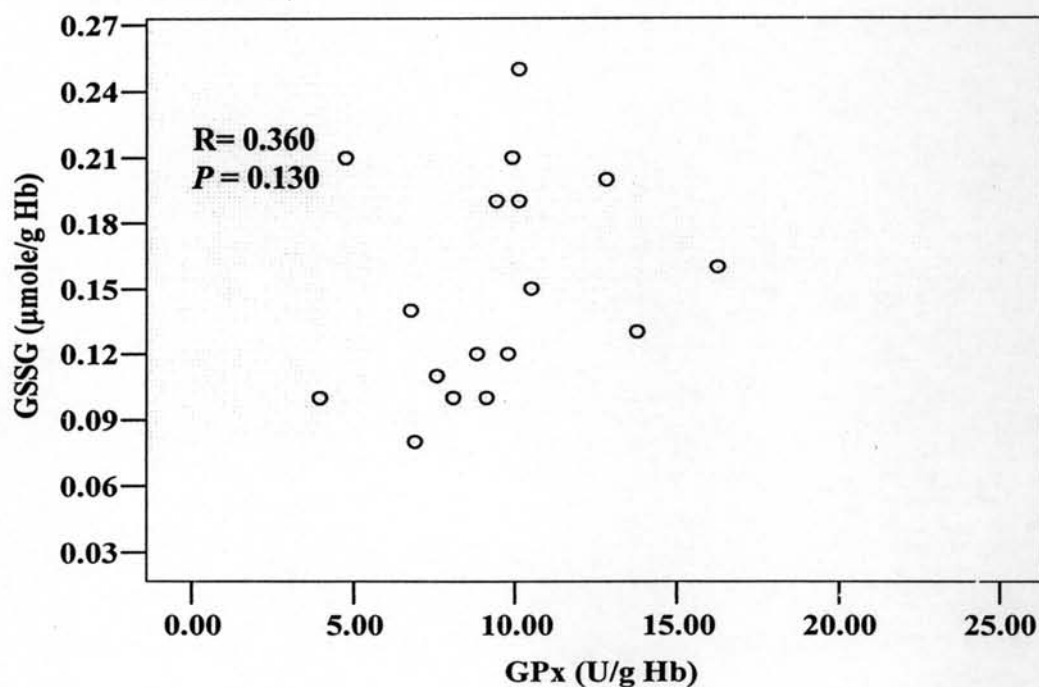
3. The correlation between GPx activities and GSSG level

The positive correlation between level of GPx activities and GSSG level in placebo group at 3, 12 month were found with the r -value of 0.77 and 0.54, respectively ($p < 0.05$). In curcuminoid, these correlations group were not found (Figure 4.13).

(A) 1. Placebo group 3 month



(A) 2. Curcuminoid group 3 month



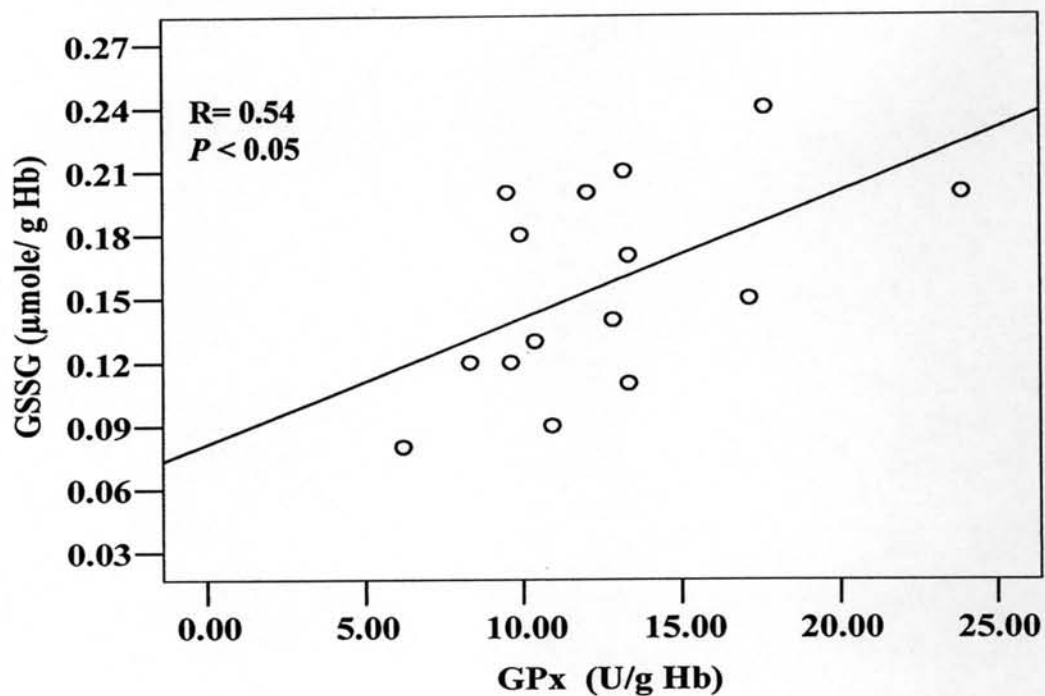
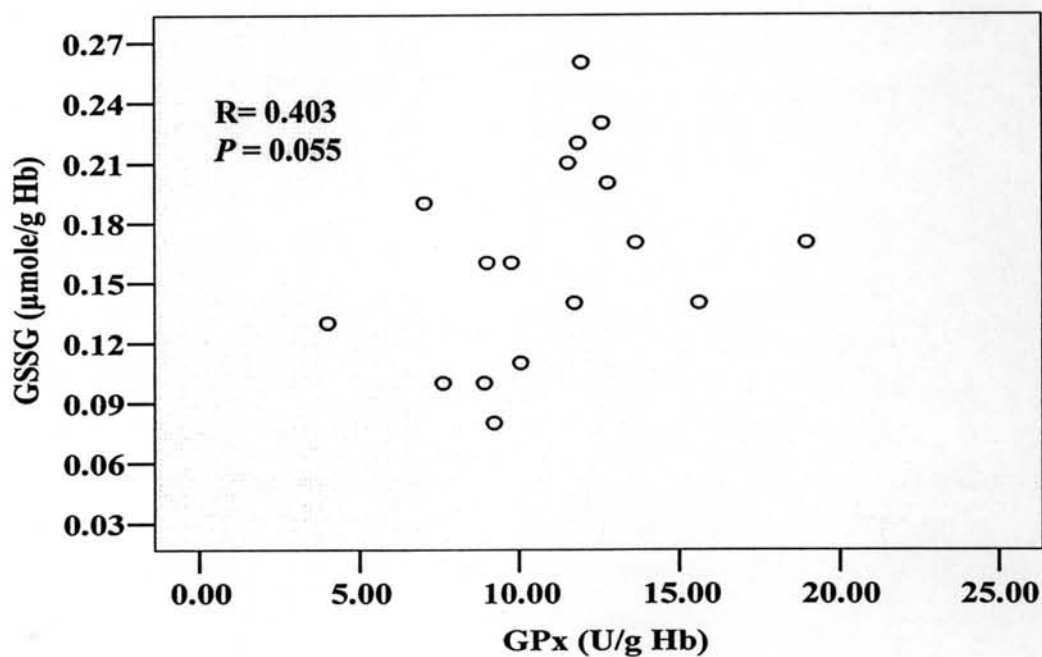
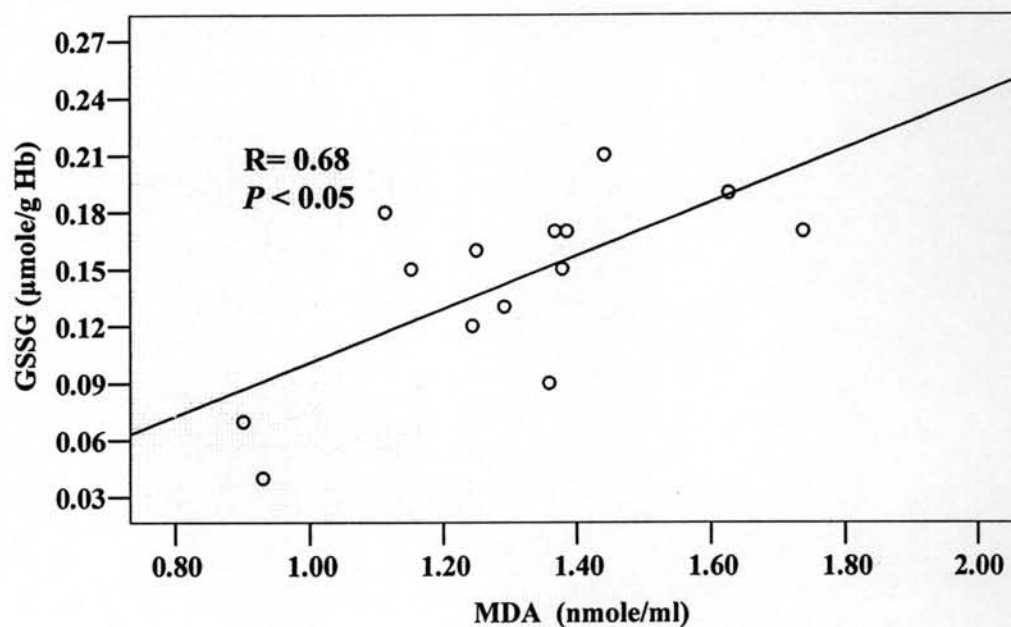
(B) 1. Placebo group 12 month**(C) 2. Curcuminoid group 12 month**

Figure 4.13 The correlation between GPx activities with GSSG level at (A) 3 and (B) 12 month. The data analyses from fifteen LHON patients in placebo group and seventeen LHON patients in curcuminoid group.

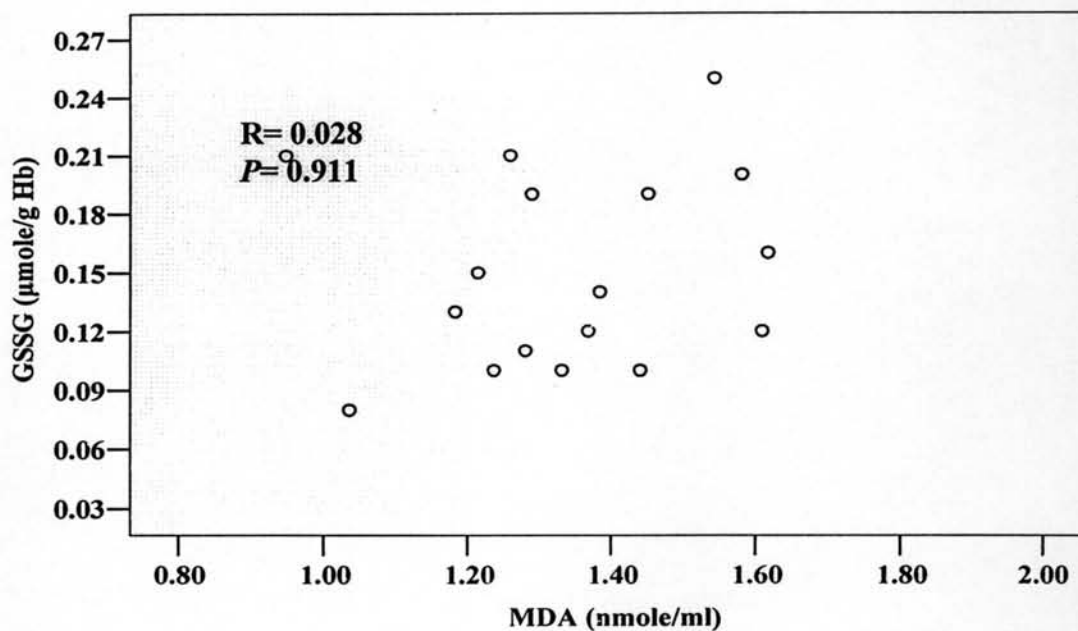
4. The correlation between MDA level and GSSG level

The positive correlation between level of MDA and GSSG level in placebo group at 3 and 6 month were found with the r -value of 0.68 and 0.56, respectively ($p < 0.05$). In curcuminoid, these correlations group were not found (Figure 4.14).

(A) 1. Placebo group 3 month



(A) 2. Curcuminoid group 3 month



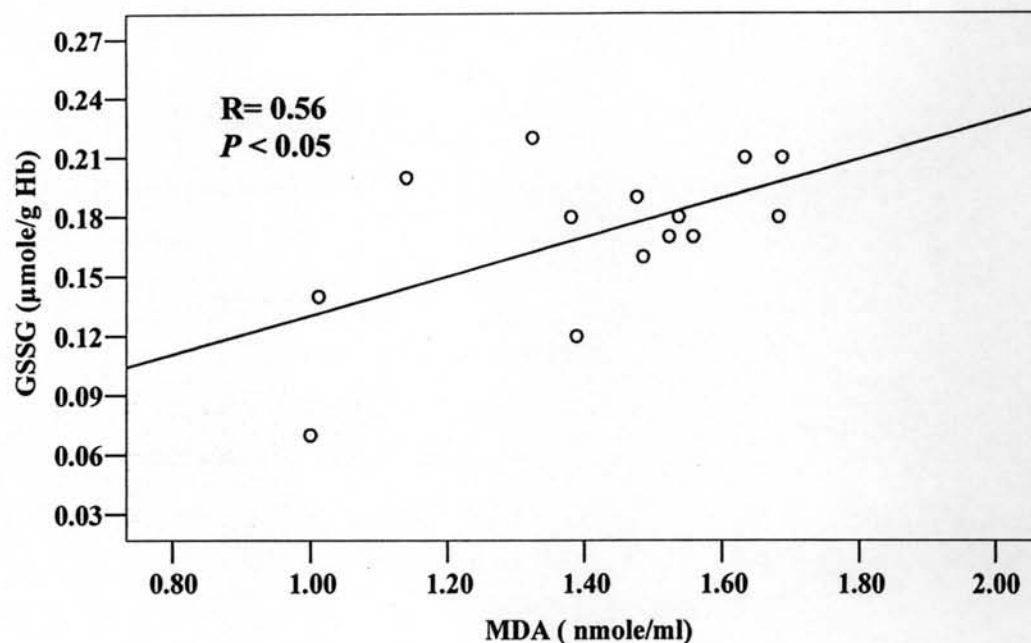
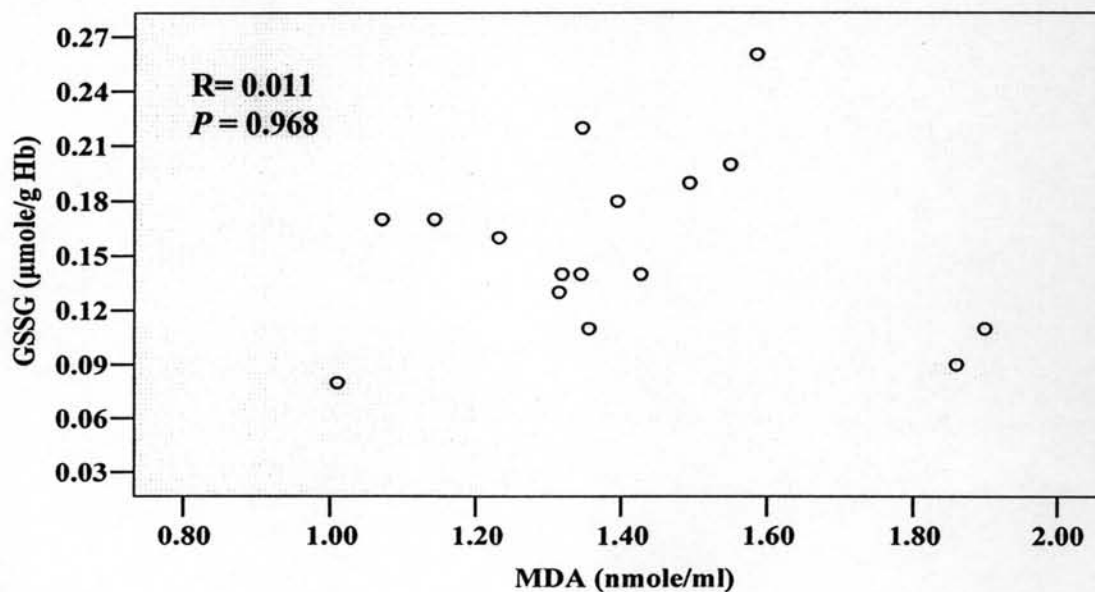
(B) 1. Placebo group 6 month**(B) 2. Curcuminoid group 6 month**

Figure 4.14 The correlation between MDA level with GSSG level at (A) 3 and (B) 6 month. The data analyses from fifteen LHON patients in placebo group and seventeen LHON patients in curcuminoid group.