# THE CLINICAL AND LABORATORY STUDIES OF SUPEROXIDE DISMUTASE ACTIVITY IN THE HUMAN WHOLE BLOOD WITH EARLY GASTRIC CANCER

# LAN-KAI-WEI et al.

### Department of Biochemistry, Sichuan Cancer Institute, Chengdu, P. R. China

In this study, we tried to evaluate the usefulness of Superoxide Dismutase (SOD) activity in detecting gastric cancer. SOD was determined according to M. Minami and H. Yoshikawa, a simple and inexpensive assay method. For 11 fibrogastroscopy and pathological proved gastric cancer cases, the average levels of SOD activity was found significantly lower than 30 patients with gastric diseases, 8 patients with atypical hyperplasia and 32 controls.

KEY WORDS: Superoxide Dismutase (SOD); Human whole blood; gastric cancer; gastric diseases; atypical hyperplasia; pyrogallol; nitroblue tetrazolium (NBT).

#### INTRODUCTION

Recent studies indicated that Superoxide Dismutase (SOD) (EC 1, 15.1.1), is an antioxidant defense enzyme.

Some of the data implicating cancer are associated with SOD activation.

In this study, we tried to evaluate the usefulness of SOD activity in detecting gastric cancer.

SOD activity reported here has been detected by pyrogallol autooxidation nitroblue tetrazolium reducing reaction (M. Minami and H. Yoshikawa 1978).

#### SUBJECT AND METHODS

#### Human blood samples

Whole blood samples at random were collected from our hospital patients with different gastric diseases, before fibrogastroscopy.

Whole blood was obtained by venipuncture prevented from coagulating by 0.1% heparin 0.1 ml. 0.1 ml of blood was hemolysed by 0.9 ml of cold (4°C) water. Hemoglobin was removed by 0.25 ml of chloroform and 0.5 ml of ethanol with rigorous mixing. The mixture was centifuged at 18000  $\times$  g for 50-60 min. The clear supernatant was used for SOD assay.

#### Reagents

- 1. Tris-cacodylic buffer (pH 8.20)
- 2. Pyrogallol solution
- 3. The solution of Nitroblue Tetrazolium (NBT)
- 4. Triton X-100 solution
- 5. Standard SOD
- 6. The reaction stopper of 2 M Formic buffer (pH 3.5)



## Procedure

The reaction mixture consisted of 0.25 ml supernatant of the sample, 0.5 ml Triscacodylic buffer pH 8.20, 0.1 ml 16% (v/v) Triton X-100 solution and 0.25 ml NBT (80 g/100 ml of water). The reaction was started by adding 10 microliters of 0.9 mM Pyrogallol solution. Incubation was performed for 5 minutes at 37°C. The reaction stopper with using the reaction was stopped by adding 0.3 ml of 2 M Formic buffer (pH 3.5). The absorbance at 540 nm.

The inhibitory rate was calculated in the following formula:

$$I = \left(1 - \frac{T}{B}\right) \times 100(\%)$$

Where

I = Inhibitory rate

B = Absorbance of the non-inhibitory medium (H<sub>2</sub>O) (instead of sample)

T = Absorbance of the medium with the sample

## **RESULTS AND DISCUSSION**

These results indicated as follows:

	N	$\frac{\text{SOD} (\mu g/\text{ml})}{\text{M} + \text{SD}}$	
			<u> </u>
Control	32	31.7	5.91
Gastritis	30	25.7	3.84
A typical hyper- plasia	8	17.33	3.99
Gastric Cancer	11	12.99	4.55

(1) All data are expressed as the mean  $\pm$  standard deviation. For the control study, 32 healthy male and female research of volunteer aged 25 to 45 years old were tested. Blood samples at random were collected from our hospital.

(2) These results shown that SOD level in gastric cancer group were significantly lower than in normal group. Changes of SOD enzyme levels in various pathological conditions also decreased.

(3) These results indicated SOD as an important enzyme in the action of protecting cell against superoxide (O<sub>2</sub>) free radicals.

(4) SOD levels might be helpful to early detection of the gastric cancer.

(5) The method of SOD assay is a simple and inexpensive for treating a large number of samples.

## References

- 1. J.M. McCord and I. Fridovich (1969) Superoxide Dismutase. An enzymic function for erythrocuprein. Journal of Biological Chemistry, 244, 6049.
- M. Minami and H. Yoshikawa (1979) A simplified assay method of superoxide dismutase activity for clinical use. *Clinica Chimia Acta*, 92, 337-342.
- 3. S.A. Lesko and R. Zneng (1982) In: *Free Radicals, Lipid Peroxidation and Cancer*, edited by D.C.R. McBrien and T.F. Slater. Academic Press, London.

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