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# Medico-Legal Update

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# Quinone Oxidoreductase 2 Serum Level that affecting the Psychotic Disorders of Methamphetamine Abusers

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## Abstract

**Background:** Some evidence suggests that genetic factors contribute to susceptibility to drug abuse such as methamphetamin and that dopamine-quinone produced by dopamine might be involved in the mechanism of symptoms associated with psychotic methamphetamine (MAP). Quinone detoxification is catalyzed by a group of proteins, namely quinone oxidoreductase (NQO). In this study we analyzed serum NQO2 levels by examining ELISA Sandwich in clients who used methamphetamine. **Methods:** This research is a Case-Control to determine the contribution of NQO2 to the emergence of psychotic disorders due to methamphetamine abuse. The case group is residents who consume methamphetamine with psychotic characteristics, while the control group is clients who consume methamphetamine without psychotic characteristics **Results:** The mean value of NQO2 gene protein in the Elisa Sandwich examination for the case group was lower than the value of the control group (1,246 < 3,657). The t test showed the value of sig (Equal variances not assumed) = 0,000 that value is less than 0.05 (<0.05), this shows the difference in the value of content protein NQO2 between the case group and the control group. Then the cut-off point of the Receiver Operating Characteristic (ROC) curve in this study is recommended statistically between 1,960 and 2,165, meaning that if the expression value of protein content > 2.165 then the potential for psychotic suffering is great, on the other hand if < 1.960, the potential is small for suffering from psychotics. **Conclusion:** The protein content of the NQO2 gene in the Elisa Sandwich examination between the case and control groups is different, the case group is lower than the control. While the CURVE ROC (Receiver Operating Characteristic) Cut-Off value is recommended statistically between 1,960 and 2,165.

**Keywords:** Methamphetamine, Dopamine, NQO2 serum level, Methamphetamine Psychotic.

## Introduction

Recently, methamphetamine abuse has become a major problem and developed into an international problem<sup>20</sup>. According to the *United Nations Office on Drugs and Crime* (UNODC), methamphetamine is a dangerous substances and the 2<sup>nd</sup> ranks after hashish/cannabis in the 1<sup>st</sup> rank of the 20 types of substances that are often misused<sup>19</sup>. The prevalence of narcotics abuse in the world from 2006 to 2013 has increased<sup>19</sup>. In the last five years, the trend of amphetamine type has declined by around 15% in various countries, while the use of ecstasy

has been reported to be stable (12). However, there has been a drastic increase (158%) in the last five years namely the consumption of methamphetamine<sup>14,19,20</sup>. Methamphetamine abusers can also cause psychotic symptoms similar to schizophrenia (Schizophrenia-like psychosis) but there are also methamphetamine abusers who do not experience psychotic symptoms<sup>24,25</sup>. As a reference from several previous studies that contributed to our research, there are two sources of reference that we can point out, namely:

a. Functional polymorphisms of the NQO2 gene is associated with methamphetamine psychosis. (Ohgake, S. at all,<sup>14,17</sup>.

b. Cognitive impairment is related to oxidative stress and chemokine levels in first psychotic episodes. (Martínez-Cengotitabengoa M at all, Schizophr Res 2012, 137:66–72).

**Material and Method**

**Study Design and Subject**

This research is a case-control to determine the effect of NQO2 gene on the onset of psychotic symptoms due to methamphetamine abuse<sup>1,22</sup>. The Selection Criteria for clients who only consume methamphetamine with a diagnosis of Behavioral Mental Disorders due to the use of methamphetamine addictive substances with psychotic characteristics for the case group, while those who do not have psychotic characteristics as a control group and are currently undergoing inpatient rehabilitation programs at the BNN Baddoka Rehabilitation Center. Inclusion Criteria: Methamphetamine abusers, aged 20 - 50 years old, qualify as methamphetamine abuse. Exclusion Criteria: Clients suffering from Mental Disorders and Behavior due to addictive substances other

than methamphetamine. There is a history of hereditary disorders in the Psychotic group, suffering from Severe Physical Disease and consumption of polysubstance during the past year<sup>9,13</sup>.

**Population of Research:** a. Case group: Methamphetamine users have Psychotic disorders, after being examined with the criteria of the Brief Psychiatric Rating Scale (BPRS) and the Diagnostic And Statistical Manual of Mental Disorder (DSM V).

b. Control group: Methamphetamine type who do not experience Psychotic disorders (examination instruments also with BPRS and DSM V).

**2.3 Statistical Analysis**

Analysis of the data in this study (Elisa’s protein content) using statistical application assistance, IBM SPSS version 24. To see the difference between the two samples, namely the case group and the control group, an independent t test was performed. Then to determine the cut-off point between psychotic and non-psychotic (Specificity and Sensitivity), by Receiver Operational Characteristics Curve (ROC) test and then performed AUC test (Area Under Curve) so that cut-off from data obtained in both groups.

**Results**

**Table 1. Characteristic of subject based on age and gender**

Distribution of case subjects (n = 30)			Distribution of control subjects (n = 30)		
Ages	n	%	Ages	n	%
20-35 yrs	27	90	20-35 yrs	27	90
36-50 yrs	3	10	36-50 yrs	3	10
Gender			Gender		
Men	29	96,7	Men	30	100
Women	1	3	Women	0	0

Table 1 shows that the age of the subjects in the case group ranged from 20-35 years (90%), most of the men were 29 (96.7%), and the rest were women.

**T-Test**

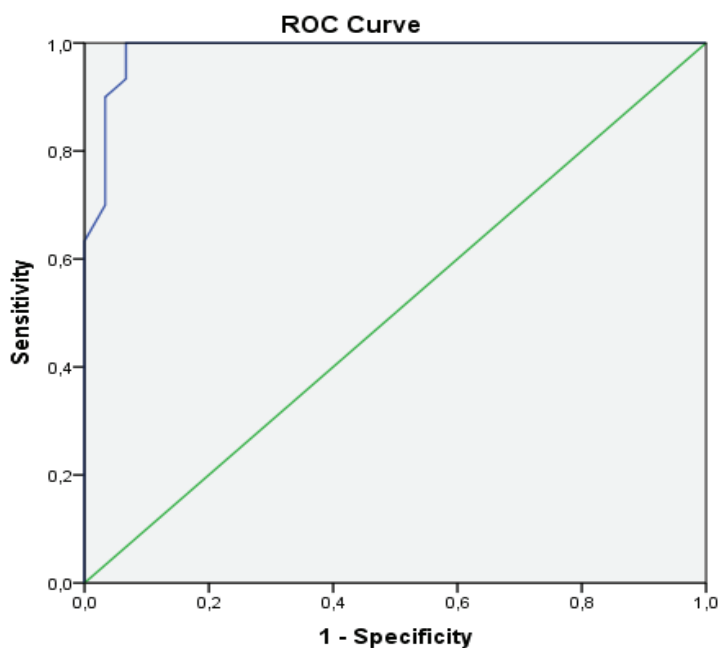
Independent t test is a test to see the difference between the two sample groups, namely cases and controls.

The mean value of Elisa protein levels in the case group (1,246) is smaller than the control group (3,657).

Table 2. Independent Samples Test										
		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Content Sandwich Elisa	Equal variances assumed	10,668	,002	12,144	58	,000	2,41100	,19853	2,01359	2,80841
	Equal variances not assumed			12,144	49,684	,000	2,41100	,19853	2,01217	2,80983

The result of *t-test* showed the sig. value of 0.000 was less than 0.05 (< 0.05). It shows that  $H_0$  rejected and  $H_a$  accepted, meaning that there were significant differences between case and control groups.

**ROC (Receiver Operating Characteristic) KURVE**



Diagonal segments are produced by ties.

**Figure 1. ROC Curves in Case Control Groups**

The ROC (Receiver Operating Characteristic) curve shows that the value is far from the 50% line and close to 100%.

**Table 3. Area Under the Curve**

**Test Result Variable(s): PROTEIN LEVELS SANDWICH ELISA**

Area	Std. Error <sup>a</sup>	Asymptotic Sig. <sup>b</sup>	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
,986	,012	,000	,962	1,000

The test result variable(s): PROTEIN LEVELS SANDWICH ELISA has at least one tie between the positive actual state group and the negative actual state group. Statistics may be biased.

- a. Under the nonparametric assumption
- b. Null hypothesis: true area = 0.5

The AUC (Area under the Curve) value is 98.6% (IK95%; 96.2% - 100%), meaning that if the ELISA Sandwich protein content / content score is used to diagnose the presence or absence of psychotic symptoms due to methamphetamine abuse in 100 clients, then the right conclusion will be obtained at 98.6 clients.

**Table 4. Coordinates of the Curve**

**Test Result Variable(s): PROTEIN LEVELS SANDWICH ELISA**

No.	Positive if Greater Than or Equal To <sup>a</sup>	Sensitivity	1 - Specificity
25	1,9600	1,000	,100
26	2,1650	1,000	,067

The test result variable(s): PROTEIN LEVELS SANDWICH ELISA has at least one tie between the positive actual state group and the negative actual state group.

The smallest cut-off value is the minimum observed test value minus 1, and the largest cutoff value is the maximum observed test value plus 1. All the other cut-off values are the averages of two consecutive ordered observed test values.

**Discussion**

As is known that methamphetamine is one type of narcotics that are stimulant, if consumed will affect all

parts of the brain, especially in the limbic system, namely the Nucleus Acumbens and Ventral Tegmental Area to activate dopamine (DA), then dopamine is released<sup>16</sup>. from storage. divescular sites to the cytoplasm so that the amount of DA increases in the brain. Furthermore, DA can carry out automatic oxidation to produce DA-Quinones. DA-Kuinon is a very reactive molecule and easily produces semi-quinones that are not stable by the reduction process. Semi-quinones enter the redox cycle together with oxygen molecules to form reactive oxygen species (ROS).

Then, ROS and semi-quinone cause oxidative stress. Therefore, it can be said that this oxidative

stress plays an important role in the mechanism of MAP, where this is evidenced by the involvement of DA-Kuinson in the mechanism of MAP which induces neurotoxicity and psychotic symptoms in humans<sup>14, 15</sup>. DA-quinone is usually detoxified by genes catalyzed by the Quinoneoxidoreductase (NQOs) protein group and also this gene protects brain cell damage from oxidative stress. In humans, genetic evidence indicates that Quinoneoxidoreductase (NQOs) has four gene loci, both of which have been identified as NAD (P) H-quinone oxidoreductase 1 (NQO1) (Ross et al.) and NRH quinoneoxiductase 2 (NQOs) NQO2)<sup>9, 11</sup>.

In a research article such as that conducted in Japan by Ohgake, S., et.al with the title “NQO2 gene polymorphism associated with methamphetamine psychosis”, in his research found a percentage (11.7%) D / D of NQO2 genes in patients with prolonged psychosis had double the control (4.8%). On the other hand, this study shows that the NQO1 polymorphism (Pro187Ser), in which this gene is not associated with MAP abuse. The NQO1 gene is proven to be a risk factor for breast, lung and leukemia<sup>3, 14, 18</sup>. It has been explained that methamphetamine is a type of narcotic that is stimulant, because it affects the release of excessive dopamine neurotransmitters from Nucleus Accumbens (NA) and Ventral Tegmental Area (VTA) in the Limbic System that causes psychotic disorders. Meanwhile, the NQO2 gene serves to detoxify the effects of excessive dopamine, as well as protect brain organs from the effects of excessive dopamine poisoning which can cause permanent brain damage<sup>14</sup>.

This NQO2 gene is “Helper”, so it is normatively assumed that the lack of regulation of the NQO2 gene causes expression of the protein content of the low NQO2 gene, this can cause psychotic disorders in methamphetamine users<sup>14,15</sup>, due to insufficiency. detoxification function and protection of the NQO2 gene. Further studies such as those conducted by Ohgake et al. (regarding the NQO2 gene polymorphism), we then tried to conduct another study, namely how the influence or contribution of protein levels from the NQO2 gene to psychotic disorders in methamphetamine users<sup>6, 11</sup>. Analysis of NQO2 gene protein content through Sandwich Elisa examination for occupants / perpetrators of methamphetamine abuse in the control group (Mental Disorders and Causative Behavior of Amphetamine

Type Non Psychotic Stimulant Type) and case groups (Mental Disorders and Behavior of Addictive Substances of Amphetamine Type of Stimulant with Addictive Substance with Psychotic Type) ) gives different results, where the average value of the protein content in the control group is higher (3,657) than in the case (1,246). Whereas the recommended KURVE ROC (Receiver Operating Characteristic) cut-off is statistically recommended between 1,960 and 2,165, with a sensitivity value of 100% and specificity  $\geq 90\%$ , it means that a cut-off value of 1,960 to 2,165 which can be used as a reference for the presence or absence potential psychotic disorders in methamphetamine abuse

## Conclusion

a. The mean value of the NQO2 gene protein level in the Sandwich Elisa examination for the case group ie MAP users = 1,246, lower than the value of the control group ie non Psychotic MA users = 3,657. b. The t-test results show the value of sig. (2-tailed) = 0,000 (smaller than  $\alpha = 0.05$  ( $<0.05$ )), so it can be assumed that there are differences between the samples in the case and control groups. c. Cut-Off CURVE ROC (Receiver Operating Characteristic) recommended statistically in both groups is between 1.960 and 2.165, meaning that if the expression value of protein content  $> 2.165$  then the potential for psychotic suffering is great, on the other hand if  $< 1.960$ , the potential is small for suffering from psychotics<sup>2, 21</sup>.

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