A Study of Contingent Negative Variation and Post-imperative Negative Variation: Search for State and Trait Electrophysiological Markers in Schizophrenia

伴隨性負變化和指令訊號後負變化的研究：精神分裂症的狀態和性向電生理學指标

J Simlai, SH Nizamie, CRJ Khess

Abstract

Objective: To compare event-related potential measures, contingent negative variation and post-imperative negative variation in drug-naïve or drug-free schizophrenic patients and normal healthy controls, and to study the effect of antipsychotic medication on the above measures.

Methods: A hospital-based prospective study was conducted at Central Institute of Psychiatry, Ranchi, India. The sample constituted 31 drug-naïve or drug-free patients with schizophrenia and 31 normal healthy individuals, matched for age and gender. An S1-S2 paradigm was used, in which the warning stimulus (S1) was auditory and the target stimulus or imperative stimulus (S2) was visual. The early contingent negative variation was marked at 500 milliseconds after S1, and late contingent negative variation was marked as the negative wave just prior to S2. The post-imperative negative variation was measured as the continued negativity after S2.

Results: Drug-naïve and drug-free patients significantly differed from the controls on amplitudes of early and late contingent negative variations, as well as on latency of late contingent negative variation. The rate for correct classification in 80% of cases (Wilks’ lambda = 0.76) was observed in measuring amplitude of late contingent negative variation only. After exposure to antipsychotic drugs, the late contingent negative variation amplitude was found to normalise in the patient group.

Conclusion: The late contingent negative variation could be considered a state marker for schizophrenia.

Key words: Contingent Negative Variation; Evoked potentials; Schizophrenia

摘要

目的：比較無藥物治療組(即從未接受藥物治療)/停用藥物組(即停用藥物至少6個月)的精神分裂症患者和對照組的與事件相關電位：伴隨性負變化和指令訊號後負變化，並分析它們對抗精神病藥療程的影響。

方法：這項於印度蘭契市中央精神科醫院進行的前瞻研究包括年齡和性別匹配的31名無藥物治療組/停用藥物組患者和31名對照組成員。研究使用S1-S2範例，S1為聽覺性警告刺激，S2為視覺性目標或命令式刺激，並於S1後500毫秒記下伴隨性負變化早成分，而當負波在S2出現前就液度伴隨性負變化晚成分。指令訊號後負變化則為S2後的持續性負波度。

結果：不論伴隨性負變化早成分跟晚成分的幅度，或者伴隨性負變化在成分的昏迷期，無藥物治療組/停用藥物組在大部分電極位置都跟對照組顯著分別。只量度伴隨性負變化成分的時候，能把病例分類的正確度為80%（Wilks’ lambda = 0.76）。在服用抗精神病藥后，患者的伴隨性負變化晚成分的幅度回复正常水平。

结论：伴隨性負變化晚成分可被考慮為精神分裂症的狀態指标。

关键词：伴隨性負變化，誘发电位，精神分裂症

Dr Jayati Simlai, MD, Department of Psychiatry, Ranchi Institute of Neuropsychiatry and Allied Sciences, Ranchi, Jharkhand, India.
Dr S. Haque Nizamie, DPM, MD, Central Institute of Psychiatry, Ranchi, Jharkhand, India.
Dr Christoday R. J. Khess, MD, Department of Psychiatry, Central Institute of Psychiatry, Ranchi, Jharkhand, India.

Address for correspondence: Dr Jayati Simlai, Department of Psychiatry, RINPAS, Ranchi 834006, Jharkhand, India.
Tel: (91) 9431105602; Email: jnou@rediffmail.com

Submitted: 30 November 2009; Accepted: 11 March 2010
Introduction

Among the novel tools for experimental analysis of cognitive function, averaged time-locked event-related potentials (ERPs) have been widely studied. The slow potentials (SPs) are event-related electromagnetic variations, which can be recorded during time intervals in milliseconds that precede expected stimuli.1

The interest in slow waves and their neurophysiological basis began with the discovery of contingent negative variation (CNV) by Walter et al.2 The CNV is a surface-negative slow cortical potential elicited between the warning and the imperative stimuli when a response to the imperative stimuli is required. Pressing a button is an example. It is a slow negativity in the electroencephalography that appears during the anticipation period between a warning stimulus and a target response. There are 2 components of a CNV. The first is related to an orienting response to the warning stimulus and the imperative stimuli when a response to the imperative stimuli is required. Pressing a button is an example. It is a slow negativity in the electroencephalography that appears during the anticipation period between a warning stimulus and a target response. There are 2 components of a CNV. The first is related to an orienting response to the warning stimulus (‘O’ wave) and the second component (‘E’ wave) is thought to be analogous to readiness potential which immediately precedes voluntary self-paced movement.3

The CNV represents summation of at least 2 components. There is stimulus-related frontally predominant negativity related to the orienting response and a centrally predominant negativity thought to be related to the premotor potential associated with motor response.4-8 The first portion and part of the second portion of CNV are independent of the motor response.4,9-11

The most consistent finding concerning the ERP of schizophrenics is reduced amplitude of CNV.12-14 Further, the CNV often does not return to baseline immediately. This is known as post-imperative negative variation (PINV).12-15

This is usually seen in acute schizophrenia. Among the SPs, CNV and PINV are well established as altered in amplitude and topography, and the former may be treated as a stable marker for schizophrenia.16 Earlier, SPs were suspected to originate from the prefrontal cortex.17

Event-related potentials give us the knowledge about cortical physiological processes during information processing. Depolarisation in the apical dendritic trees of cortical neurons results in surface-negative potentials.18

The difference observed in ERP between psychotic patients and normal subjects has been consistent and is thought to be related to cognitive processes.12,19 The reduction of the ERP amplitude has been interpreted as a sign of impaired information processing, inability to concentrate, increased distractibility or increased trial-to-trial variability of responses.19,20 The CNV is associated with focused attention and may serve as a state-dependent predictor of conscious awareness. Further, CNV has been proposed to reflect central dopaminergic activity.21,22

Unlike attenuation of most ERP components, there is an enhanced surface-negative potential (PINV) following a voluntary motor response.23 The PINV has been attributed to delayed CNV resolution,24,25 increased preparation for further stimulus and response evaluation,18,26 inability to resolve stimulus ambiguity,27 subjective ambivalence and difficulties in fitting the response into a symbolic scheme,28 and uncertainty about the results of actions.19,20

Methods

Subjects

A total of 31 schizophrenic patients fulfilling the Diagnostic Criteria for Research (DCR) accompanying the 10th revision of the International Classification of Diseases (ICD-10) criteria29 were recruited from the psychiatry inpatient department of the Central Institute of Psychiatry Ranchi, India. The inclusion criteria for the patients were: (1) either drug-naïve (never treated) or drug-free (off-medICATIONS for the last 6 months at least); and (2) aged between 20 and 50 years. The patient sample consisted of only males, of which 8 were drug-naïve and 23 were drug-free. A control group matched for age and gender was recruited from normal healthy volunteers. The volunteers were assessed using the General Health Questionnaire-5 (GHQ-5).30 Those scoring 1 or more were excluded from the study. The aims, objectives and procedure of the study were explained, and informed consent was obtained from all subjects. The protocol of this study was approved by the academic and ethics committee of the institute.

After the subjects were enrolled for the study, they were assessed by Mandal et al’s Handedness Preference Schedule,31 a standardised Indian version of Annett’s handedness inventory.32 All subjects were found to be right-handed. Within 24 hours the patients were rated on the Brief Psychiatric Rating Scale (BPRS)33 and Positive and Negative Symptom Scale (PANSS) for schizophrenia34.

At baseline (drug-naïve / drug-free state) and after 1 month of antipsychotic treatment (post-drug state), CNV and PINV recordings were performed once for the control group and twice for the patients. After the baseline investigation, the patients were started on antipsychotics. They received 400 to 600 mg of chlorpromazine equivalent per day. Parkinsonian side-effects were assessed clinically and those who developed them were given trihexyphenidyl (2-4 mg per day). None of the patients were given benzodiazepines.

A Neuropack Sigma 8 software (Nihon Kohden, Japan) was used to record CNV and PINV. The CNV and PINV recordings were undertaken within 1 to 2 hours of food intake (10 am-12 noon, 2-4 pm). All the patients were afebrile at the time of recording. The patient’s scalp and hair were thoroughly washed with soap containing minimum glycerine. The subjects were allowed to dry their hair before the recording procedure. They were made to lie on a cot in the supine position keeping their eyes open and fixed at some specific spot and avoid blinking. They were also asked to remain immobile and stay relaxed. The hair was parted at the site of electrode placement and the skin cleaned by an absorbent cotton pad moistened in acetone. A small amount of skinpure (sand and jelly paste) was rubbed onto the cleaned spot so that the skin impedance was lowered. The electrode was pressed onto the cleaned spot using a small amount of electrode conductance jelly. Electrode resistance
was kept to less than 2000 ohms.

The CNV was recorded by making the subject wear a pair of special goggles which emitted a red stroboscopic light produced in an odd-ball paradigm. A continuous masking noise of 75 dB was emitted through the headphones. Initially a warning tone appeared which alerted the subject for the following stroboscopic light stimulus. The moment the red stroboscopic light appeared, the subject was instructed to switch off the light by pressing the button each time as quickly as possible. The type of target stimulus (S2) was visual and the warning stimulus (S1) was auditory. The stimulation rate of S1 was 0.1 Hz and the interval between S1 and S2 was 3 seconds. The tone frequency of S1 was 1.5 KHz with a plateau time of 30 milliseconds; the rise and fall of time being 10 milliseconds. The high-cut filters were at 20 Hz and low-cut at 0.01 Hz for all channels. The preset count was 100. Rejection level was 3 divisions (which was indicated in the machine and its manual) and the paper speed was 25 mm per second. The various parameters measured were: N1, which was the first negative wave; after the S1 stimulus or the ‘O’ wave; N2, which was the early CNV (CNV [i]) marked at a fixed point 500 milliseconds after the stimulus S1, was called the ‘E’ wave; and N3, which was the late CNV (CNV [t]), was marked as a negative wave just prior to the onset of the light stimulus S2. The PINV was measured as the continued negativity after the stimulus S2. The scalp electrode sites used for recording were Fz, Cz, Pz, C3, and C4. The raters were final-year master-degree psychiatry residents who were undergoing postgraduate training. The inter-rater reliability was tested and found to be good (Kappa coefficient, -0.88).

Statistical Analysis
The group comparison of the various measures for drug-naïve / drug-free state versus normal controls and post-drug state versus normal controls was undertaken using one-way analysis of variance. Discriminant analysis was performed to find out whether any of the ERP measures could correctly discriminate between patients and controls. In the patient population, the results obtained in the drug-naïve / drug-free state and the post-drug state were compared to explore the possible effects of antipsychotic medication, using paired t tests.

Results
The mean (SD) age of the patient group was 30 (9) years, and that of the controls was 30 (4) years. The mean (SD) duration of the illness in the patient group was 6 (5) years. The mean (SD) BPRS score in the pre-drug state was 57 (13), and in post-drug state it was 35 (12). The mean (SD) PANSS positive index in the pre-drug state was 2 (8) and 13 (6) in the post-drug state. Corresponding values of the PANSS negative index were 22 (8) and 16 (7), and that of the composite index were 20 (11) and 20 (7), respectively.

The CNV (i) amplitudes between drug-free / drug-naïve schizophrenic patients and normal controls analysed by one-way analysis of variance are shown in Table 1. The results showed a significant difference at Cz electrodes. Table 2 shows the comparison of CNV (t) amplitude, and that there were significant differences at Fz, Cz, and C4 electrodes. A comparison of PINV amplitude between the normal controls and the patients of pre-drug state revealed that there was no significant difference at any of the sites. Table 3 shows the comparison of CNV (t) latency, with significant difference being found at all the 5 electrodes. When comparing PINV latency in the normal controls and pre-drug–state patients, no significant difference was noted at any of the sites. To investigate how correctly the 2 groups were classified as per the various measures described above, a discriminant analysis was performed. The rate for correct classification in 80% of cases (Wilks’ lambda = 0.757) was observed for only the measure of CNV (t) amplitude and hence only this was considered for further analysis. Comparison of CNV (t) amplitude between pre-drug and post-drug state in the patient population was performed using the paired t test, and that Table 4 revealed significant differences at all sites. Finally, comparison of CNV (t) amplitude of the post-drug state in the patient population with normal controls was performed using one-way analysis of variance, but no significant difference was found at any of the sites (Figure).

Discussion
The institute where this study was conducted is a 673-bedded tertiary centre catering to a large, predominantly rural population. Outpatient consultations are free but the patients have to buy their own medicines. This could be a reason why some poor patients were off-medication (drug-free) when seen for consultations. Hence we were able to recruit such individuals along with never-treated (drug-naïve) patients.

Age, gender, and psychoactive substances are known to influence CNV genesis, so the age and gender were matched for the control group, whilst the effects of antipsychotics on ERP measures were studied in the patient group. The earlier studies of ERP in schizophrenia were plagued by methodological constraints, including diagnostic uncertainties due to lack of operationalised diagnostic guidelines and signal averaging techniques. Furthermore, they did not address the influence of medications. Although ERP studies have been conducted on diverse sample population in various clinical laboratories, most of this data come from western countries. This study was conducted on Indian patients (who are ethnically and genetically quite different), in the knowledge that ethnicity may have an effect on CNV. We used the ICD-10 DCR criteria for diagnostic purposes to ensure a reliable and homogenous sample of schizophrenic patients. For the normal controls, who were matched for age and gender, GHQ-5 was used to ensure the absence of any psychopathology. For signal averaging, we used the Neuropack Sigma 8 software. The effect of antipsychotic drugs on CNV was examined by
Table 1. A comparison of early contingent negative variation amplitude.

<table>
<thead>
<tr>
<th>Electrode site</th>
<th>Normal Mean</th>
<th>Normal SD</th>
<th>Pre-drug state Mean</th>
<th>Pre-drug state SD</th>
<th>F ratio</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fz</td>
<td>7.5</td>
<td>2.0</td>
<td>6.7</td>
<td>3.4</td>
<td>1.17</td>
<td>NS</td>
</tr>
<tr>
<td>Cz</td>
<td>4.5</td>
<td>1.7</td>
<td>3.8</td>
<td>2.4</td>
<td>4.27</td>
<td>0.04</td>
</tr>
<tr>
<td>Pz</td>
<td>6.3</td>
<td>1.7</td>
<td>5.9</td>
<td>2.2</td>
<td>0.60</td>
<td>NS</td>
</tr>
<tr>
<td>C3</td>
<td>7.1</td>
<td>1.4</td>
<td>6.8</td>
<td>2.5</td>
<td>0.44</td>
<td>NS</td>
</tr>
<tr>
<td>C4</td>
<td>4.3</td>
<td>1.4</td>
<td>3.8</td>
<td>2.0</td>
<td>1.49</td>
<td>NS</td>
</tr>
</tbody>
</table>

Abbreviations: F = frontal electrode; C = central electrode; P = parietal electrode; SD = standard deviation; NS = not significant.

Table 2. A comparison of late contingent negative variation amplitude.

<table>
<thead>
<tr>
<th>Electrode site</th>
<th>Normal Mean</th>
<th>Normal SD</th>
<th>Pre-drug state Mean</th>
<th>Pre-drug state SD</th>
<th>F ratio</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fz</td>
<td>23.0</td>
<td>5.6</td>
<td>19.7</td>
<td>5.8</td>
<td>5.32</td>
<td>0.02</td>
</tr>
<tr>
<td>Cz</td>
<td>17.4</td>
<td>3.0</td>
<td>13.7</td>
<td>4.2</td>
<td>15.72</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Pz</td>
<td>30.4</td>
<td>6.9</td>
<td>28.9</td>
<td>3.9</td>
<td>1.18</td>
<td>NS</td>
</tr>
<tr>
<td>C3</td>
<td>14.3</td>
<td>4.1</td>
<td>12.6</td>
<td>3.7</td>
<td>2.81</td>
<td>NS</td>
</tr>
<tr>
<td>C4</td>
<td>14.7</td>
<td>2.4</td>
<td>12.5</td>
<td>3.8</td>
<td>7.30</td>
<td>0.01</td>
</tr>
</tbody>
</table>

Abbreviations: F = frontal electrode; C = central electrode; P = parietal electrode; SD = standard deviation; NS = not significant.

Table 3. A comparison of late contingent negative variation latency.

<table>
<thead>
<tr>
<th>Electrode site</th>
<th>Normal Mean</th>
<th>Normal SD</th>
<th>Pre-drug state Mean</th>
<th>Pre-drug state SD</th>
<th>F ratio</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fz</td>
<td>3038.4</td>
<td>38.0</td>
<td>3007.7</td>
<td>39.1</td>
<td>9.79</td>
<td>0.003</td>
</tr>
<tr>
<td>Cz</td>
<td>3039.0</td>
<td>38.9</td>
<td>3007.7</td>
<td>39.1</td>
<td>9.98</td>
<td>0.002</td>
</tr>
<tr>
<td>Pz</td>
<td>3039.4</td>
<td>38.1</td>
<td>3007.7</td>
<td>39.1</td>
<td>10.38</td>
<td>0.002</td>
</tr>
<tr>
<td>C3</td>
<td>3038.1</td>
<td>39.4</td>
<td>3007.7</td>
<td>39.1</td>
<td>9.25</td>
<td>0.003</td>
</tr>
<tr>
<td>C4</td>
<td>3041.0</td>
<td>38.6</td>
<td>3007.7</td>
<td>39.1</td>
<td>11.33</td>
<td>&lt; 0.001</td>
</tr>
</tbody>
</table>

Abbreviations: F = frontal electrode; C = central electrode; P = parietal electrode; SD = standard deviation.

Table 4. A comparison of late contingent negative variation amplitude between pre-drug and post-drug state.

<table>
<thead>
<tr>
<th>Electrode site</th>
<th>Pre-drug state Mean</th>
<th>Pre-drug state SD</th>
<th>Post-drug state Mean</th>
<th>Post-drug state SD</th>
<th>t</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fz</td>
<td>19.7</td>
<td>5.8</td>
<td>24.7</td>
<td>7.7</td>
<td>-2.78</td>
<td>0.01</td>
</tr>
<tr>
<td>Cz</td>
<td>13.7</td>
<td>4.2</td>
<td>6.9</td>
<td>10.1</td>
<td>-2.12</td>
<td>0.04</td>
</tr>
<tr>
<td>Pz</td>
<td>28.9</td>
<td>3.9</td>
<td>31.3</td>
<td>4.1</td>
<td>-2.11</td>
<td>0.04</td>
</tr>
<tr>
<td>C3</td>
<td>12.6</td>
<td>3.7</td>
<td>14.8</td>
<td>4.2</td>
<td>-2.34</td>
<td>0.02</td>
</tr>
<tr>
<td>C4</td>
<td>12.5</td>
<td>3.8</td>
<td>8.2</td>
<td>3.4</td>
<td>-2.70</td>
<td>&lt; 0.01</td>
</tr>
</tbody>
</table>

Abbreviation: F = frontal electrode; C = central electrode; P = parietal electrode; SD = standard deviation.
studying patients when they were drug-naïve / drug-free and after they had been treated with antipsychotics for 1 month.

In this study, we compared the amplitude and latency of CNV (i), CNV (t), and PINV. Although there were significant differences between the patient group and the controls, we only focused on the amplitude of CNV (t) based on discriminant analysis yielding an overall hit rate of 80%, the reason being that this measure could accurately classify the patients and the normal controls. The CNV (t) amplitude of the patient population was significantly smaller than that in the controls at the Fz, Cz, and C4 sites (in the fronto-central regions). After exposure to antipsychotic drugs, the CNV (t) amplitude was found to normalise in the patient group. Hence, it may be possible that CNV (t) amplitude helps discriminate schizophrenic patients from normal controls. Further, antipsychotic treatment causes normalisation or enhancement of the CNV (t) amplitude.

A study of schizophrenics suggest a negative CNV-dopamine association. Patients, who are presumed to have an overactivity of dopaminergic systems, show consistently low CNV amplitudes. When dopamine antagonists are administered they exhibit enhanced CNV amplitudes, which indicates an inverse relationship between CNV and dopamine. The CNV can be a reliable state marker for schizophrenia, which is characterised by a hyperdopaminergic state in the acute phase of the illness, which is reversed by antipsychotic medication. In recent times, growing evidence indicates that the SPs originate not only from the prefrontal cortex but from other cortical areas including association areas and posterior cortices. Moreover, some SPs may not be related to task performance itself but to preparatory processes such as selective attention or the mere attempt to perform the task. Since ERP objectively records the neural activity associated with perceptual and cognitive processes, these procedures possess enormous face validity in the study of diseases such as schizophrenia. However, to realise their full potential in the study of psychiatric disorders, additional fundamental studies are required. The latter include efforts to relate ERP components to their neuro-anatomical and neurophysiological substrates on one hand, and to psychological processes on the other. When these data become available, it will be possible to use ERP to probe psychological and physiological derangements in schizophrenia. Until then the intriguing ERP differences that have been described cannot be fully integrated with psychological and biological theories of schizophrenia.

The inference that CNV amplitudes normalised after treatment and that this was due to the antipsychotic effects of the medications is unequivocal, but whether this was due to the resolution of symptoms or treatment response cannot be concluded in this study. Although the overall BPRS and PANSS scores showed improvement after treatment (as detailed in the results), it would have been more useful if the patients who responded and those who had not were compared. This remains a major shortcoming of this study.

**Figure.** Contingent negative variation (CNV) recordings of (a) normals, (b) patients in pre-drug state, and (c) patients in post-drug state.

Abbreviations: \( A1 = Fz \) (frontal); \( A2 = Cz \) (central); \( A3 = Pz \) (parietal); \( A4 = C3 \) (left central); \( A5 = C4 \) (right central); \( N1 \) = orienting wave; \( N2 \) = early CNV; \( N3 \) = late CNV; \( PINV \) = post-imperative negative variation.
as it is known that the CNV amplitude may normalise either with antipsychotic treatment or in association with remission of the acute stage of the disease.42-44

Declaration

The authors declared that there was no financial support for this study and that they had no conflicts of interest.

References

39. Basile LF, Ballester G, Castro CC, Gattaz WF. Prefrontal cortex activity assessed by high-resolution EEG and current density reconstruction.

41. Holzman PS. Recent studies of psychophysiology in schizophrenia. Schizophr Bull 1987;13:49-75.

