Investigation of the multiple comparisons problem in the analysis of the wave train electrical activity of muscles in Parkinson’s disease patients

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Abstract. A new method has been developed for the analysis of the wave train electrical activity of muscles based on the wavelet analysis and ROC analysis that enables to study the time-frequency characteristics of electromyograms (EMG) and acceleration (ACC) signals in patients with Parkinson’s disease (PD). The idea of the method is to find local maxima (that correspond to the wave trains) in the wavelet spectrogram and to calculate various characteristics describing these maxima: the leading frequency, the duration of the wave trains in periods, the bandwidth of the wave trains, the number of wave trains per second. The degree of difference between a group of patients and a control group of volunteers in the space of these parameters is analyzed. ROC analysis is used for this purpose. The functional dependence of AUC (the area under the ROC curve) on the values of the boundaries of parameters’ ranges under consideration is investigated. The developed method involves investigation of a big number of ranges of selected characteristics; therefore a multiple comparisons problem appears during statistical hypothesis testing. It is necessary to find a compromise between the degree of detail of the studied characteristics and the magnitude of the Bonferroni correction. The paper describes the statistical hypothesis testing on the data of early Parkinson’s disease patients.

1. Introduction
The investigation of the multiple comparisons problem [1–5] is currently an urgent task of biomedicine. A big number of methods have been developed to solve the multiple comparisons problem. These methods are based on the analysis of family-wise error rate (FWE), the false discovery rate (FDR), the application of random field theory (RFT) [5–7], the permutation method [3], etc. Unfortunately, the overwhelming majority of these methods do not consider the connection between the multiple comparisons problem and the problem of multiscale analysis of biomedical data. Meanwhile, these two problems are very closely related to each other, both from the point of view of the mathematical apparatus and from the point of view of the practical application of methods of statistical data analysis. The problem of multiscale analysis is that biomedical data may contain patterns that appear on various, previously unknown scales of images or signals. It is necessary to investigate data in a certain space of scales to detect such patterns. However, different scales of images and signals correspond to a different number of
multiple comparisons and, therefore, suggest a different level of statistical correction for the number of multiple comparisons. Moreover, the investigation of data on multiple scales is a case of the multiple comparisons and, therefore, may require the application of an additional correction for the multiple comparisons. This problem, in particular, was considered in the random field theory and an approach to analyzing data based on the space of scales (the scale space approach) was proposed [8]. However, in this paper, we will use the standard Bonferroni correction [1] to concentrate on the problem of multiscale analysis.

Earlier, we have developed the method of analysis of the wave train electrical activity of cerebral cortex based on the wavelet analysis and the ROC analysis [9, 10]. The idea of this method is that we consider the electroencephalogram (EEG) as a combination of wave trains [11]. Unlike works dedicated to the detection of the electrical activity of specific types, such as alpha-spindles [12] and sleep spindles [13–18], we analyze all types of wave trains electrical activity in the cerebral cortex in a wide range of frequencies. In addition, we consider the wave train as a typical component of EEG, but not as a special kind of EEG signals [19, 20]. The analysis method considered in this paper is based on the statistical analysis of wavelet spectrograms, the new method of visualization of the statistical analysis results, and an improved wave train detection algorithm.

To analyze electromyograms (EMG) and signals of an accelerometer (ACC), the developed algorithm for signal analysis [9, 10, 21–24] has been modified. An additional step of smoothing the wavelet spectrograms of signals was added. Smoothing is required because the standard fast wavelet calculation algorithms have the following drawback: wavelet spectrograms are inevitably contaminated by artifacts (outliers and high-frequency oscillations) when processing signals of complex shape. These artifacts can be mistakenly recognized as the wave trains. To solve this problem, the smoothing of the spectrograms by the Gaussian window with specially selected parameters developed by the authors was applied to remove the “jags” in the wavelet spectrograms. Parameters of the smoothing window (time width and bandwidth) are changed adaptively depending on the wavelet scaling parameter. Moreover, the threshold used for initial wave train selection has been reduced; currently, the duration of the wave trains must be more or equal to 1 period. The wave train duration is measured at a height equal to the square root of $1/2$ from the peak maximum.

The use of the method of analysis of wave train electrical activity allowed us to identify new neurophysiological regularities in patients with Parkinson’s disease (PD) compared with healthy volunteers in the range of 3–7 Hz in the EMG and ACC signals. It was found that wave trains in the range of 3–7 Hz have the following characteristics: 50 $\mu$V$^2$/Hz and higher, 1–5 periods, 1–5 Hz frequency band [25]. Differences between groups were revealed both on healthy hands and on the tremor hands of patients (with a tremor on the left hands and with a tremor on the right hands). While, the differences in the healthy left hands of right-hand-tremor patients are more pronounced on EMG, and the differences in the healthy right hands of left-hand-tremor patients are more pronounced on the signals of the accelerometer.

2. Experimental settings

Untreated (that is, previously not taking special medicines) PD patients at the early stages were compared with healthy volunteers. Note that the group of patients with PD included both left-hand-tremor patients (15 people) and right-hand-tremor patients (18 people), the total is 33 patients. The number of healthy volunteers was 18 people. All patients and healthy volunteers were right-handed. No statistically significant differences between the ages of patients and healthy volunteers were found. EMG electrodes were located on the outer sides of the arms, on the extensor muscles, accelerometers were placed on the backs of the palms. EMG and ACC signals were recorded in a special position of the subject – the subject was sitting in a chair, his hands were on the armrests of the chair, his palms were straightened and turned
Figure 1. The diagrams of AUC values based on EMG signals with detected statistically significant areas without corrections for multiple comparisons. The first column corresponds to the left-hand-tremor patients. The second column corresponds to the right-hand-tremor patients. The first line corresponds to the left hands. The second line corresponds to the right hands. The abscissa is the lower bound of the frequency range and the ordinate is the upper bound of the frequency range.

edge to the floor, his legs were put on the heels. The eyes were closed during all recordings. The 41-channel multifunctional digital system for neurophysiological studies Neuron-Spectrum-5 (Neurosoft Ltd.) was used for EMG recording. The accelerometer developed by E. M. Timanin in the IAP RAS [26] was used to record ACC. The sampling rate of EMG was 500 Hz and the accelerometer sampling rate was 1378.125 Hz. The 0.5 Hz high-pass filter and the 50 Hz notch filter were used during EMG and ACC recording. For EMG signals, the 60–240 Hz bandpass Butterworth filter and Hilbert transform for envelope detection were used. The duration of every record was about 2 minutes. The record was analyzed as is, without a selection of areas in the signal.

3. The method
To detect statistically significant differences in the studied characteristics of wave trains of electrical muscle activity in groups of patients and control subjects, the Mann-Whitney non-parametric test is used. On Figure 1 and Figure 2 special diagrams of AUC values (further, AUC diagrams) with detected statistically significant areas in the frequency range from 3 to 7 Hz (without correction for multiple comparisons) is presented. Statistically significant areas in almost all images can be seen, except the image where the number of wave trains in EMG signals on the right hands of control subjects and the number of wave trains on the right (healthy) hands of left-hand-tremor patients are compared (Figure 1).

The developed method involves the change of a big number of ranges of selected characteristics, therefore, when checking statistical hypotheses, the problem of multiple comparisons appears. The simplest solution is using Bonferroni correction. The alpha level remains constant, 0.05. The amount $K$ of cells on the diagram varies. We will call the number of columns (or rows) in the diagram as the resolution $R$ of the method. The key problem is what resolution should be chosen? Large resolution allows detecting areas of small size in the
Figure 2. The diagrams of AUC values based on the ACC signals with detected statistically significant areas without corrections for multiple comparisons. The first column corresponds to the left-hand-tremor patients. The second column corresponds to the right-hand-tremor patients. The first line corresponds to the left hands. The second line corresponds to the right hands. The abscissa is the lower bound of the frequency range and the ordinate is the upper bound of the frequency range.

diagram, however, the Bonferroni correction may turn out to be too big and, therefore, the statistical test will not detect any statistically significant differences. It is necessary to ensure that small parts are visible in the diagram and, at the same time, the correction to the multiple comparisons is not too big. That is, it is necessary to find a compromise between the degree of detail of the investigated characteristics and the Bonferroni correction magnitude.

The idea of our approach is a multiscale analysis. We change the resolution in the diagrams to search a Bonferroni correction in which statistically significant effects do not disappear. The goal of this procedure is to find the highest resolution where a statistically significant area passed the Bonferroni correction appears.

Thus, the signal analysis method includes the following steps:

(i) The signals are prefiltered and the envelope of EMG is computed.
(ii) The wavelet spectrograms are computed.
(iii) The wave trains are detected in the spectrograms.
(iv) The characteristics of the wave trains and the quantity of the wave trains per second are computed.
(v) The AUC diagrams are created.
(vi) The Mann-Whitney statistical test is applied for each point in the diagram.
(vii) Bonferroni correction is applied for multiple comparisons.
(viii) The statistically significant areas are detected in the diagrams.
(ix) The v–viii steps are repeated for different values of the resolution.
(x) The statistically significant areas passed the Bonferroni correction are detected. In particular, the resolution that corresponds to the largest statistically significant area passed the Bonferroni correction is detected. Also, the highest resolution is detected where a statistically significant area passed the Bonferroni correction appears.
Figure 3. There is the resolution 69 with a statistically significant area. The patients with a tremor of the left tremor hand are considered.

Figure 4. There is an isosurface corresponding to p-value $\leq 0.05$. The patients with a tremor of the left tremor hand are investigated (accelerometer data, frequencies are from 3 to 7 Hz).

Consider the AUC diagrams in the following subspace of wave train parameter values: the frequency range is from 3 to 7 Hz, the amplitude is from 50 $\mu V^2$/Hz and higher, the number of periods is from 1 to 5 periods, and the wave train bandwidth is from 1 to 5 Hz. In this example, accelerometer signals are examined. Patients have a tremor of the left hand; this hand is considered. A statistically significant area appears the first time at the resolution 108 (that is, the number of cells under the consideration is $108 \times 108$). The Figure 3 demonstrates the resolution 69 with a statistically significant area (approximately in the range from 4 to 6 Hz). Note that the correction for multiple comparisons is made only on the number of cells in the AUC diagram. Additional correction for the number of considered scales is not made, therefore, the shape of the obtained areas of statistically significant differences is random. In this paper, the form of areas of statistically significant differences is not investigated. Only the fact of the presence of statistically significant differences in the frequency domains under study is important to us.

The obtained dependences of statistically significant areas on the resolution value can be represented as isosurfaces. The Figure 4 demonstrates the isosurface corresponding to p-value $\leq 0.05$. The patients with a tremor of the left tremor hand are investigated (accelerometer data, frequencies are from 3 to 7 Hz).
In the examined figures, statistically significant areas have the forms of “pears”, that is, they gradually increase from top to bottom and then decrease (approximately at the resolution 3–4). A decrease in statistically significant regions occurs because of a small number of cells under consideration. Regions with regularities of opposite directivity fall into the same cells, as well as regions with no regularities.

Let us consider the top views of isosurfaces based on EMG (Figure 5, Figure 6) and ACC (Figure 7) signals. The statistically significant areas that have passed the Bonferroni correction remained on the EMG diagrams only in tremor hands. They are observed both in the left-hand-tremor patients and in the right-hand-tremor patients. On the accelerometer diagrams, statistically significant areas remained only in the left-hand-tremor patients (only on the tremor hands).

4. Results
A new method of exploratory data analysis was developed. This method involves calculating AUC values and non-parametric statistical hypotheses testing to detect statistically significant differences in the characteristics of the wave trains of the muscles’ electrical activity. A detailed analysis of data from Parkinson’s patients and control subjects was carried out. It is proved that the revealing differences between the groups of patients with PD and healthy subjects
Figure 7. The top views of the isosurfaces based on ACC data of the left-hand-tremor patients (tremor left hand is presented in the figure). The abscissa is the lower bound of the frequency range and the ordinate is the upper bound of the frequency range.

are statistically significant. Areas with the smallest p-value that have passed the Bonferroni correction were found. Statistically significant differences in the EMG data were found both in the left-hand-tremor patients and in the right-hand-tremor patients (only on the tremor hands). In the accelerometer data, statistically significant differences were found only in the left-hand-tremor patients (only on the tremor hands); however, the regularities are more pronounced in ACC than in the EMG data. It has been shown that the analysis of patients with Parkinson’s disease using EMG and ACC gives different results. EMG and accelerometer can complement each other. The regularities found in EMG and accelerometer data can be useful for diagnosing the early stages of Parkinson’s disease.

5. References

[1] Shaffer J P 1995 Annual Review of Psychology 46 561-584
[2] Pigeot I 2000 Statistical papers 41 3-36
[3] Nichols T E 2012 Neuroimage 62 811-815
[4] Austin S R, Dialsingh I and Altman N 2014 J. Indian Soc. Of Agricultural Stat 68 303-314
[5] Petersson K M, Nichols T E, Poline J B and Holmes A P 1999 Philosophical Transactions of the Royal Society of London B: Biological Sciences 354 1261-1281
[6] Brett M, Penny W and Kiebel S 2004 Human Brain Function (Burlington: Academic Press) 867-879
[7] Rohani F 2009 Nonparametric Random Fields with Applications in Functional Imaging (Ph.D. thesis McGill University)
[8] Worsley K J, Wolforth M and Evans A C 1997 Proceedings of BrainMap 96
[9] Sushkova O S, Morozov A A and Gabova A V 2016 International Conference on Bioinformatics and Systems Biology (BSB-2016) 1-4
[10] Sushkova O S, Morozov A A and Gabova A V 2017 Advances in Soft Computing. Lecture Notes in Computer Science 10062 403-412
[11] Sushkova O S, Morozov A A, Gabova A V and Karabanov A V 2016 Proceedings of the 12th Russian-German Conference on Biomedical Engineering 80-84
[12] Lawhern V, Kerick S and Robbins K A 2013 BMC Neuroscience 14 101 URL: http://www.biomedcentral.com/1471-2202/14/101
[13] Parekh A, Selesnick I, Rapoport D and Ayappa I 2014 IEEE Signal Processing in Medicine and Biology Symposium (Philadelphia, PA: IEEE) 1-6
[14] O’Reilly C and Nielsen T 2015 Frontiers in Human Neuroscience 9 353 DOI: 10.3389/fnhum.2015.00353
[15] Huupponen E, Clercq W D, G’omez-Herrero G, Saastamoinen A, Egiazarian K, Varri A, Vanrumste B, Vergult A, Huffel S V, Paesschen W V, Hasan J and Himanen S L 2006 Journal of Neuroscience Methods 156 275-283
[16] Nonclercq A, Urbain C, Verheulpen D, Decaestecker C, Bogaert P V and Peigneux P 2013 Journal of Neuroscience Methods 214 192-203
[17] Jaleel A, Ahmed B, Tafreshi R, Boivin D B, Streletz I and Haddad N 2014 Journal of Neuroscience Methods 233 1-12
[18] Camilleri T A, Camilleri K P and Fabri S G 2014 Biomedical Signal Processing and Control 10 117-127
[19] Zhirmunskaya E A 1993 Klinicheskaya elektroentsefalografiya (tsifry, gistogrammy, illyustratsii) (Moscow: Mezhotraslevoy nauchno-issledovatel’skiy inzhenerno-tekhnologicheskiy tsентр “Skan”)
[20] Obukhov Y V, Korolev M S, Gabova A V, Kuznetsova G D and Ugrumov M V 2013 Method of early encephalographic diagnostics of Parkinson disease Patent no. 2484766 Russian Federation
[21] Sushkova O S, Morozov A A and Gabova A V 2016 CEUR Workshop Proceedings 1638 681-690
[22] Sushkova O S, Morozov A A and Gabova A V 2017 13th International Conference on Signal-Image Technology & Internet-Based Systems (SITIS) 168-172
[23] Sushkova O S, Morozov A A, Gabova A V and Karabanov A V 2018 Journal of Neurology and Psychiatry 118 45-48
[24] Sushkova O S, Morozov A A, Gabova A V and Karabanov A V 2018 Journal of Physics: Conference Series 1096 012078
[25] Sushkova O S, Morozov A A, Gabova A V and Karabanov A V 2018 Advances in Artificial Intelligence: 16th Ibero-American Conference on AI (Cham: Springer International Publishing) 253-264
[26] Timanin E M, Gustov A V and Eremin E V Device for complex analysis of different types of human tremor Patent no. 2483676. Russian Federation

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