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Research Article

Resting-state functional magnetic resonance imaging: features of statistical processing of ROI-analysis data

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ABSTRACT

BACKGROUND: In many works, to study intra- and inter-network connections, a method for constructing networks is used — ROI-analysis (region of interest analysis). The conflicting results obtained when assessing brain connectivity using ROI-analysis can be explained by methodological differences associated with the statistical processing of fMRI data. In this regard, it is relevant to conduct a study with a comparative assessment of various statistical methods of ROI-analysis in processing resting state fMRI data.

AIM: to assess the functional connectivity of the main resting state networks of the brain using ROI-analysis using various statistical approaches.

MATERIALS AND METHODS: We analyzed data from 15 resting-state fMRI studies of the brain of patients without neurological and mental pathology. fMRI scanning was performed on a Phillips Ingenia 1.5 T scanner using a gradient echo-planar imaging (EPI-BOLD) sequence. ROI-analysis was used to build networks. Statistical data processing was performed using methods: functional network connectivity, randomization/permutation spatial pairwise clustering statistics, and threshold-free cluster enhancement.

RESULTS: The number of connections between the structures of brain networks recorded using the method of functional network connectivity is 280, spatial pairwise clustering — 186, threshold-free cluster enhancement — 182. An interesting fact is that negative connections were identified only when using parametric statistics.

CONCLUSION: A comparative assessment of methods for statistical processing of fMRI data during ROI-analysis was carried out. The functional network connectivity method based on multivariate parametric statistics turned out to be more informative than randomization/permutation spatial pairwise clustering statistics and the method based on threshold-free cluster enhancement. Despite the growing popularity in recent years of resting-state fMRI in the study of functional activity and connectivity of the brain, there are no standardized algorithms for constructing networks of the brain.

Keywords: resting-state networks; resting-state fMRI; ROI-analysis; statistics.

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Научная статья

Функциональная магнитно-резонансная томография в состоянии покоя: особенности статистической обработки данных ROI-анализа

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АННОТАЦИЯ

Актуальность. Во многих работах для исследования внутри- и межсетевых связей используют метод построения нейросетей — анализ на основе зон интереса. Противоречивые результаты, получаемые при оценке коннективности головного мозга с использованием анализа на основе зон интереса, можно объяснить методологическими различиями, связанными со статистической обработкой данных функциональной магнитно-резонансной томографии. В связи с этим актуально проведение исследования со сравнительной оценкой различных статистических методов анализа на основе зон интереса в обработке данных функциональной магнитно-резонансной томографии в состоянии покоя.

Цель исследования: оценить функциональную связность основных нейросетей покоя головного мозга при анализе на основе зон интереса с применением различных статистических подходов.

Материалы и методы. Проведен анализ данных 15 функциональных магнитно-резонансных томографий в состоянии покоя головного мозга пациентов без неврологической и психической патологии. Функциональное магнитно-резонансное исследование выполнялось на сканере Phillips Ingenia 1,5 Тл с использованием последовательности градиентной эхо-планарной визуализации. Для построения нейросетей применен анализ на основе зон интереса. Статистическую обработку данных выполняли с помощью методов функциональной сетевой коннективности, пространственной парной кластеризации, основанной на анализе рандомизации/перестановки, и улучшения беспорогового кластера.

Результаты. Количество связей между структурами сетей головного мозга, зафиксированных при использовании метода функциональной сетевой коннективности, равно 280, пространственной парной кластеризации — 186, улучшения беспорогового кластера — 182. Интересный факт заключается в том, что отрицательные связи выявлены только при использовании параметрической статистики.

Заключение. Выполнена сравнительная оценка методов статистической обработки данных функциональной магнитно-резонансной томографии при проведении анализа на основе зон интереса. Метод функциональной сетевой коннективности на основе многомерной параметрической статистики оказался информативнее, чем пространственная парная кластеризация, основанная на анализе перестановок/рандомизации, и метод, основанный на улучшении беспорогового кластера. Несмотря на возрастающую в последние годы популярность функциональной магнитно-резонансной томографии в состоянии покоя в исследовании функциональной активности и коннективности головного мозга, нет стандартизированных алгоритмов построения его нейросетей.

Ключевые слова: нейросети покоя; ROI-анализ; статистика; фМРТ в состоянии покоя.

Как цитировать

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研究文章

静息状态态下的功能磁共振成像：ROI分析数据统计处理的特点

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简评

现实意义。在许多研究中，使用构建神经网络的方法来研究网络内和网络间的联系——基于感兴趣区域的分析。使用兴趣区分析法评估大脑连通性时获得的结果相互矛盾，这可以用功能磁共振成像数据统计处理方法上的差异来解释。因此，在处理静息状态下的功能磁共振成像数据时，有必要对基于兴趣区的不同统计分析方法进行比较评估。

本研究的目的是评估主要静息状态下大脑神经网络的功能连接性，根据感兴趣的区域，采用不同的统计方法进行分析。

材料和方法。我们对15名无神经和精神疾病患者的大脑静息状态下的功能磁共振成像数据进行了分析。功能性磁共振成像是在1.5 TL Phillips Ingenia扫描仪上使用梯度回波平面成像序列进行的。在构建神经网络时采用了基于兴趣区的分析方法。统计数据处理采用了功能网络连接、基于随机化/重排分析的空间成对聚类和无角聚类增强等方法。

结果。使用功能网络连接法记录的大脑网络结构之间的连接数量为280个，空间成对聚类为186个，无角聚类增强为182个。一个有趣的事实是，只有在使用参数统计时负联系才会被识别出来。

结论。对基于兴趣区分析中功能磁共振成像数据的统计处理方法进行了比较评估。事实证明，基于多元参数统计的功能网络连接方法比基于置换/随机分析的空间成对聚类方法和基于无角聚类增强的方法信息量更大。尽管近年来静息状态下的功能磁共振成像在大脑功能活动和连接性研究中越来越受欢迎，但目前还没有构建其神经网络的标准化算法。

关键词：静息神经网络；ROI分析；统计学；静息态fMRI。

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BACKGROUND

Resting-state functional magnetic resonance imaging (fMRI) is used to study the pathophysiology of diseases associated with impaired functional brain activity [1–3] by showing the degree of blood oxygen saturation in a local area of the brain (BOLD signal). According to recent research, pathological connectivity between resting neural networks may underlie various neurological and psychiatric diseases [4, 5]. Resting neural networks refer to spatially independent brain structures that are capable of coordinated activations without specific tasks or stimuli and involved in performing various cognitive functions; detection, processing, and integration of internal and external stimuli; and mental processes [6, 7]. Modern neurobiological studies described eight main resting neural networks [8–10]:

1. Passive mode network (DefaultMode): medial prefrontal cortex, posterior cingulate cortex/preclinical, and inferior parietal lobe on both sides
2. Sensorimotor network (SensoriMotor): superior (supplementary motor cortex of medial sections of frontal lobes) and bilateral lateral (motor and sensory cortex on both sides)
3. Visual network: medial, occipital, and lateral subnetworks
4. Significance network (Salience): anterior cingulate cortex, insula, rostral prefrontal cortex, and supramarginal gyrus
5. Attention network (DorsalAttention): frontal visual field and intraparietal sulcus
6. Executive control network (FrontoParietal): dorsolateral prefrontal and posterior parietal cortex on both sides
7. Language network: inferior frontal gyrus and posterior superior temporal gyrus on both sides
8. Cerebellar network: anterior and posterior cerebellar lobes

Several studies used the neural network construction method for region-of-interest-based analysis (ROI-analysis) to examine intra- and inter-network connectivity. Inconsistent findings in assessing brain connectivity using ROI-analysis can be attributed to methodological differences in statistical processing of fMRI data. To obtain valid scientific data, reproducibility of results should be established. The primary distinctions are evident in selecting the statistical analysis method for neural networks.

The most commonly used methods for statistical data processing in ROI-analysis are parametric statistics based on functional network connectivity, nonparametric statistics based on randomization/rearrangement analysis (i.e., spatial pairwise clustering), and nonparametric statistics based on threshold-free cluster enhancement [11].

Currently, there are no scientific studies that apply several statistical processing methods to one dataset. Therefore, a study that includes a comparative evaluation of different statistical methods for ROI-analysis in the processing of resting-state fMRI data should be conducted.

The present study aimed to assess the functional connectivity of the primary resting brain neural networks in ROI-analysis using various statistical methods.

MATERIALS AND METHODS

Fifteen resting-state fMRI studies of the brain from the Department of X-ray and Radiology (with a course of ultrasonic diagnostics) of the Military Medical Academy were retrospectively analyzed. MR scans of patients without neurologic and psychiatric pathology were selected based on analysis results. Functional MR scanning was performed using a Phillips Ingenia 1.5 Tesla scanner in the MRI department. Resting-state fMRI images were acquired using EPI-BOLD, with a repetition time (TR) of 3000 ms, an echo time (TE) of 50 ms, a field of view of 250 mm, a flip angle (FA) of 90°, a matrix of 128 × 128, a slice thickness of 4 mm, a slice gap of 0.6 mm, and 280 volumes. High-resolution T1-weighted structural images were obtained using a 3D-TFE sequence with a TR of 7.5 ms, a TE of 3.5 ms, an FA of 8°, a matrix of 256 × 256, and a slice thickness of 1.2 mm for volume 1.

The data were analyzed using neuroimaging software, including MATLAB, CONN21a, and SPM 12 [12]. Preprocessing involved functional alignment and unfolding, slice synchronization correction, direct functional segmentation, and normalization in the Montreal Neurological Institute space coordinate system. Further, functional spatial smoothing was performed using an 8-mm-wide Gaussian kernel. Outliers, which are BOLD signal artifacts, were identified using the ART toolkit if they differed by more than 3 standard deviations from the mean image intensity. To exclude false noise sources (e.g., physiological), an anatomical component-based noise reduction strategy (aCompCor) was employed [13]. Finally, a band-pass filter with a frequency window of 0.01–0.1 Hz was applied.

Following all preprocessing procedures, ROI-analysis was conducted to establish the functional connectivity between neural networks in the brain. This was evaluated by calculating correlations (using the Pearson correlation coefficient) between the time series of BOLD signal changes throughout the brain.

The following were used for statistical processing of ROI-analysis data:

1. The functional network connectivity (FNC) method is based on multivariate parametric statistics [14]. The process begins by identifying the networks of interest. Then, FNC analyzes all links between pairs of ROIs

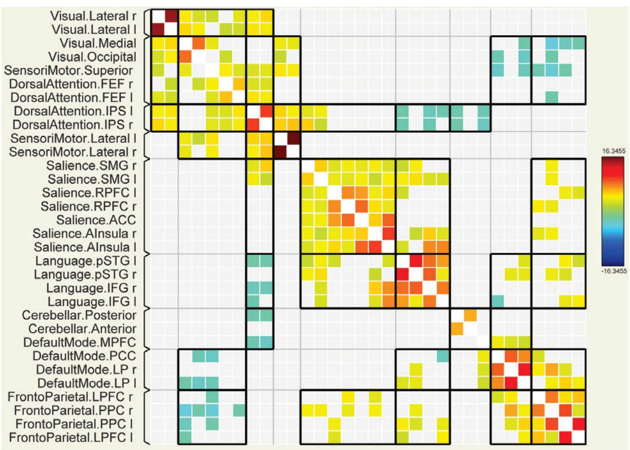
within and between networks by performing a multivariate parametric analysis of the general linear model of all connections included in each of these sets (clusters) of links. To evaluate individual clusters, an adjusted p-value corrected for the expected false discovery rate (FDR; Benjamini–Hochberg coefficient) at the cluster level ($p < 0.05$) was used to select significant interconnect sets. Additionally, an unadjusted p-value for the height (connection level) threshold ($p < 0.05$) was utilized to characterize the structure of individual connections within each significant set.

2. The spatial pairwise clustering (SPC) method [15] is a nonparametric statistic based on randomization and rearrangement. The process begins with a matrix estimated using a general linear model. The networks in this matrix are sorted either manually by the user (e.g., from an atlas) or automatically using a hierarchical clustering procedure [16]. Then, a statistical parametric map is determined using a height threshold ($p < 0.001$). The suprathreshold regions that result define nonoverlapping clusters. Each cluster is characterized by its mass, which is the sum of the square of the F- or T-statistics for all compounds within the cluster. These masses are compared to the distribution of expected cluster mass values under the null hypothesis. The null hypothesis is numerically estimated using multiple iterations of randomization/rearrangement of the original data. To evaluate individual clusters, an unadjusted p-value at the cluster level ($p < 0.01$) and a p-value adjusted for expected FDR at the cluster level ($p < 0.05$) were used to select only the significant clusters.

3. The threshold-free cluster enhancement (TFCE) method [17] is a nonparametric statistic. Similar to SPC analysis, TFCE starts with the entire matrix estimated using a general linear model, with the networks sorted either manually or automatically. Instead of defining a parametric statistical map using a height threshold, the analysis proceeds by computing the corresponding TFCE score map, combining the strength of the statistical effect for each connection with the size of all clusters. The null hypothesis's expected distribution of TFCE values is then numerically estimated using ⩾1000 iterations of randomization or rearrangement of the original data. A cluster level adjusted family-wise error p-value ($p < 0.05$) is applied to select significant clusters.

RESULTS AND DISCUSSION

Based on the results of various statistical processing methods, connectivity matrices that allow for the evaluation of inter- and intra-network connections were obtained (Figs. 1 and 2). The FNC method reveals that the visual and SensoriMotor networks exhibit positive connections with each other and with the DorsalAttention network and negative connections with DefaultMode and FrontoParietal networks (Fig. 1). The DorsalAttention network displays positive connections with the Salience, visual, and SensoriMotor networks and negative connections with the DefaultMode, FrontoParietal, language, and cerebellar networks. The cerebellar network has only one connection. The Salience network interacts positively with the DorsalAttention, FrontoParietal, and language networks. The language network is positively functionally related to the Salience and FrontoParietal networks and negatively related to the DorsalAttention network. Moreover, the language network has connections with both direct and inverse correlation with the DefaultMode network. The DefaultMode and FrontoParietal networks are interconnected with the DorsalAttention, visual, SensoriMotor, and language networks. Additionally, the FrontoParietal network is linked to the Salience network.



Visual: visual network;
SensoriMotor: SensoriMotor network;
DorsalAttention: network of attention;
Salience: network for identifying significance;
Language: speech network;
Cerebellar: cerebellar network;
DefaultMode: passive mode network;
FrontoParietal: executive control network

Fig. 1. Connectivity matrix using the method of multivariate parametric statistics based on functional network connectivity (FCN)
Рис. 1. Матрица коннективности при использовании метода многомерной параметрической статистики на основе функциональной сетевой коннективности (FCN)

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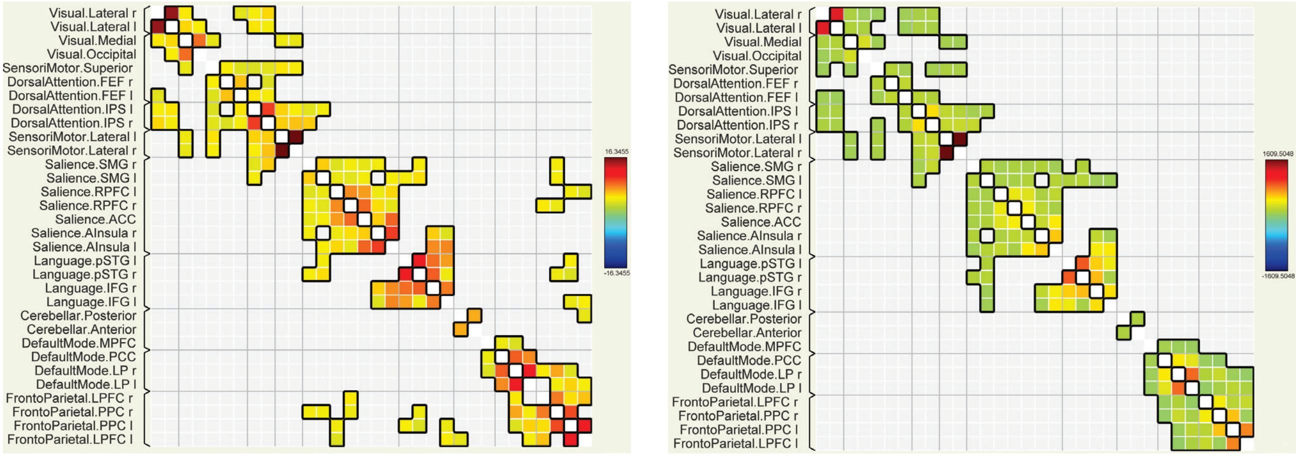

а *б*

Visual: visual network; SensoriMotor: SensoriMotor network; DorsalAttention: network of attention; Salience: network for identifying significance; Language: speech network; Cerebellar: cerebellar network; DefaultMode: passive mode network; FrontoParietal: executive control network

Fig. 2. Connectivity matrix using non-parametric statistics methods based on: *а* — randomization/permutation spatial pairwise clustering (SPC); *б* — threshold free cluster enhancement (TFCE)

Рис. 2. Матрица коннективности при использовании методов непараметрической статистики на основе: *а* — анализа рандомизации/перестановки — пространственная парная кластеризация (SPC); *б* — улучшения беспорогового кластера (TFCE)

Table. Connectivity of brain networks using nonparametric statistics methods

Таблица. Коннективность нейросетей головного мозга при использовании методов непараметрической статистики

Resting neural networks	SPC	TFCE
Salience	DorsalAttention, Language, FrontoParietal	DorsalAttention, Language
Language	Salience, FrontoParietal	Salience
Cerebellar	FrontoParietal	–
FrontoParietal	Cerebellar, SensoriMotor, DorsalAttention, Language, DefaultMode	DefaultMode

The nonparametric statistical methods applied revealed that the visual, SensoriMotor, and DorsalAttention networks do not have any connections with the DefaultMode and FrontoParietal networks (Fig. 2). Furthermore, the DorsalAttention network does not have any connectivity with the language and cerebellar networks. Finally, the DefaultMode network is only connected with the FrontoParietal network. The networks described above have identical connectivity at SPC and TFCE. However, in the other networks, the results differed (Table 1).

The FNC method recorded 280 connections between brain network structures, whereas SPC and TFCE showed 186 and 182, respectively. Negative correlations were only found when parametric statistics were used (Fig. 3) and are significant as they indicate the suppression of one network while activating another. The negative correlation of networks is supported by the increasing popularity of the triple network model theory [5].

Several studies of brain neural networks did not specify the statistical processing method used. Divergent results of resting-state fMRI data may be related to different methodological approaches. The present study demonstrated that the choice of a statistical method for a single dataset significantly affects study results.

Although parametric statistics reveal a greater number of relationships, whether they have an advantage over nonparametric methods is unclear. Further research is warranted to determine the information these methods provide in intergroup processing, particularly when comparing fMRI data between patients with any pathology and controls.

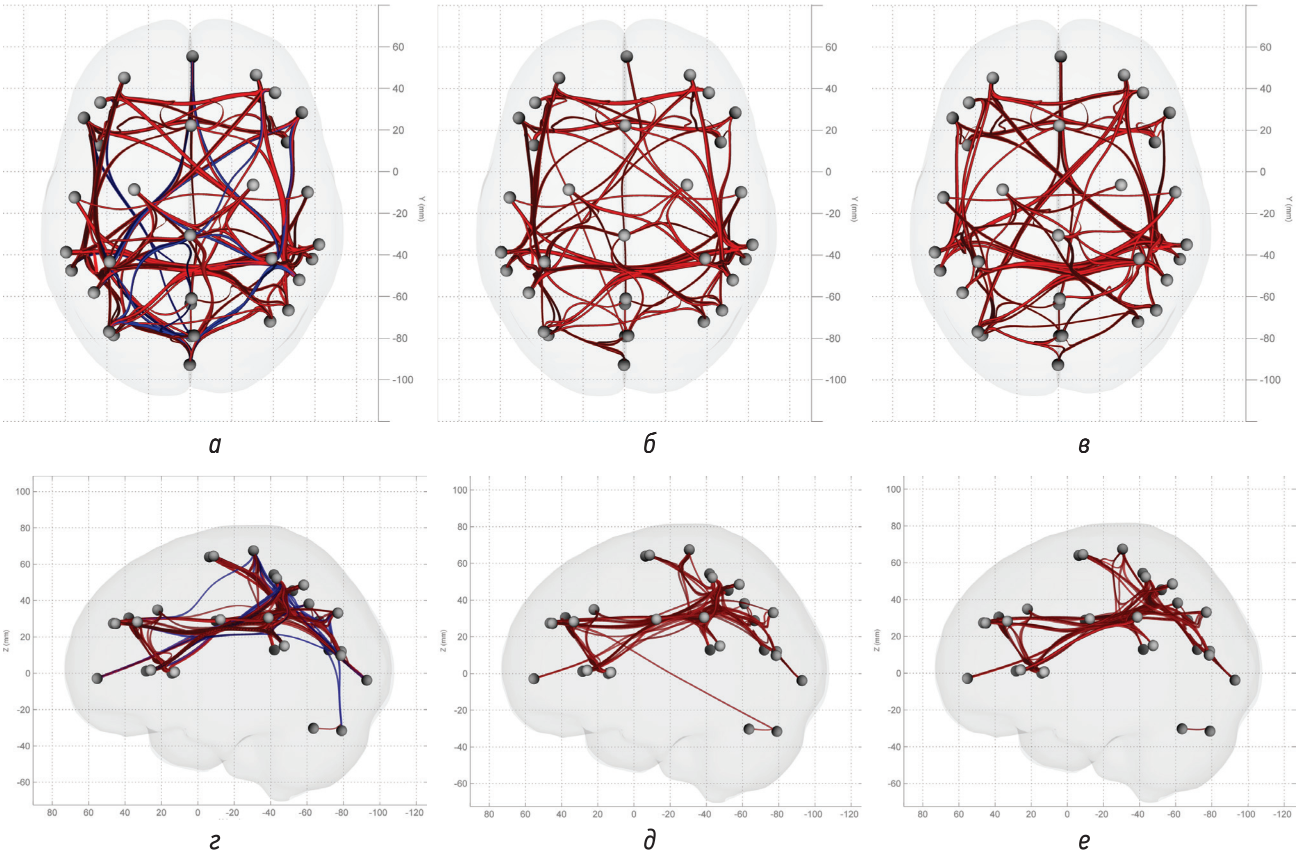

Fig. 3. Spatial image of the functional connections of the resting state networks of the brain using processing statistics methods: *а*, *г* — functional network connectivity (FCN); *б*, *д* — randomization/permutation spatial pairwise clustering (SPC); *в*, *е* — threshold free cluster enhancement (TFCE)

Рис. 3. Пространственное изображение функциональных связей нейросетей покоя головного мозга при использовании методов статистики обработки: *а*, *г* — функциональная сетевая коннективность (FCN); *б*, *д* — анализ рандомизации/перестановки — пространственная парная кластеризация (SPC); *в*, *е* — улучшение беспорогового кластера (TFCE)

CONCLUSIONS

Resting-state fMRI has become increasingly popular for studying the functional activity and connectivity of the brain. However, there are currently no standardized algorithms for constructing neural networks. Various methods are available for analyzing the functional integration and segregation of the brain, each of which provides unique information. Remarkably, each method has its distinctions in statistical data processing.

The use of multivariate parametric statistics in the FNC method enables the identification of a greater number of functional relationships compared to nonparametric methods. This method is particularly useful in identifying links with negative correlation. Our study determined that SPC based on randomization/rearrangement analysis was more informative than the threshold-free enhancement method among nonparametric methods.

ADDITIONAL INFORMATION

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Authors' contributions. All authors contributed substantially to the study and article and read and approved the final version before publication.

Conflict of interest. The authors declare that there are no obvious and potential conflicts of interest related to the publication of this article.

Ethical review. The study was performed as part of a dissertation research and approved by the local ethical committee.

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