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Short communication

Modified Richardson's method versus the box-counting method in neuroscience

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HIGHLIGHTS

- Lewis Fry Richardson was a Quaker and physicist.
- Richardson's method measures the fractal dimensions of open and closed lines.
- The fractal dimension of neuronal arborization can be measured by this method.
- Modified Richardson's method gives more precise results than box-counting method.

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ABSTRACT

Background: The morphology of dendrites, including apical dendrites of pyramidal neurons, is already well-known. However, the quantification of their complexity still remains open. Fractal analysis has proven to be a valuable method of analyzing the degree of complexity of dendrite morphology.
New method: Richardson's method is a technique of measuring the fractal dimension of open and closed lines of objects. This method was modified in order to measure the fractal dimension of neuronal arborization. The focus of this experiment was on the apical dendrites of superficial and deep pyramidal neurons in the rat cerebral cortex.
Results: Apical dendrites of superficial cortical pyramidal neurons have a higher mean value of the fractal dimension as compared to deep pyramidal neurons.
Comparison with existing method: Using the modified Richardson's method we showed that the mean value of the fractal dimension of apical dendrites in superficial pyramidal neurons is highly statistically significant as compared to the value of the fractal dimension in deep pyramidal neurons. On the other hand, the mean values of the fractal dimension between the same groups of apical dendrites measured by the most popular box-counting method showed merely a statistically significant difference.
Conclusion: The modified Richardson's method of fractal analysis is an efficient mathematical method for calculating the fractal dimension of dendrites and could be used in order to calculate the complexity of dendrite arborization.

1. Introduction

Lewis F. Richardson (1881–1953) was a Quaker and physicist who stated an interesting hypothesis: more wars are fought between nations that share a longer common border (Richardson, 1961; Mandelbrot, 1967). Addressing this issue required measuring the lengths of national borders. Therefore, Richardson, from his point of view as a scientist, using a divider with sharp points at the ends of both arms setting it to a prescribed opening: r. He moved the divider along the border of two nations on maps from a 1950s atlas. Each new step started where the previous step left off (Richardson, 1961). Richardson also measured the length (perimeter) of the border (coastline) of Great Britain. Since a typical border (such as a coastline) is irregular and winding, Richardson noticed that as the divider's opening became smaller and smaller, the observed approximate length $L(r)$ tended to

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increase without limit. The diagrams shown in Richardson's paper lead to the conclusion that the approximate length of a border can be fitted by a function (with two parameters *F* and *D*).

$$L(r) = F \cdot r^{1-D} \quad (1)$$

If *N* is the number of the divider's steps, the length of the border is $L(r) = Nr$ and *N* is

$$N = F \cdot r^{-D} \quad (2)$$

According to Richardson, *D* was a simple fitting exponent of no particular significance. But according to Benoît B. Mandelbrot, Richardson's exceptional work deserved serious attention since the exponent *D* should be interpreted as the "first category of the fractal dimension" (Mandelbrot, 2004).

After Richardson's time, computers emerged. Richardson's (segment-counting, ruler-counting, trace, yardstick) method of measuring the fractal dimension of an object was valuable but, at the same time, tedious and time-consuming. Therefore, the need for more sophisticated methods for this measurement emerged. Applying the methodology from Mandelbrot's "The Fractal Geometry of Nature" (Mandelbrot, 2004), many new computer-based methods and different types of software for measuring the fractal dimension appeared (Smith et al., 1989; Morigiwa et al., 1989). One of the most popular techniques is the box-counting method (Takeda et al., 1992; Panico and Sterling, 1995; Fernández and Jelinek, 2001; Milošević and Ristanović, 2006; Ristanović and Milošević, 2012). The main difference between Richardson's and box-counting method is that the fractal dimension obtained by Richardson's method measures the waviness or ruggedness of the objects border, whereas the fractal dimension D_b obtained by the box-counting method includes the space-filling value in addition. Hence, the Richardson's fractal dimension should be somewhat smaller than the corresponding box-counting dimension of the same geometrical object or pattern (Ristanović et al., 2013). However, precise comparison of these methods has not been done so far.

It is known that there are several different methods to calculate the fractal dimension that may give slightly different values from the same experimental material (Smith et al., 1996; Jelinek and Fernandez, 1998). The present study has shown that the fractal dimension D_b obtained via the box-counting method significantly differs from the fractal dimension *D* obtained via Richardson's method. The apical dendrites of superficial and deep pyramidal neurons from the rat cerebral neocortex were convenient histological substrates for applying and comparing both methods.

2. Materials and methods

2.1. The experimental method

The classical Golgi–Kopsch technique was applied on brain samples of male Wistar rats in order to impregnate neurons of the cerebral cortex. A total of 20 cortical pyramidal neurons from coronal sections were selected: 10 superficial and 10 deep pyramidal neurons. The two main criteria for the adequate selection of pyramidal neurons were: the cell body position and the projection path of the apical dendrite. According to these criteria, the superficial neurons are those whose cell bodies are positioned in lamina II–III, and whose apical dendrites project to lamina I, while deep neurons are those whose cell bodies are positioned in lamina V–VI, and whose apical dendrites project to lamina III–IV.

Pictures of selected neurons were taken at multiple focal planes, and pasted together. Cell bodies, spines and basal dendrites

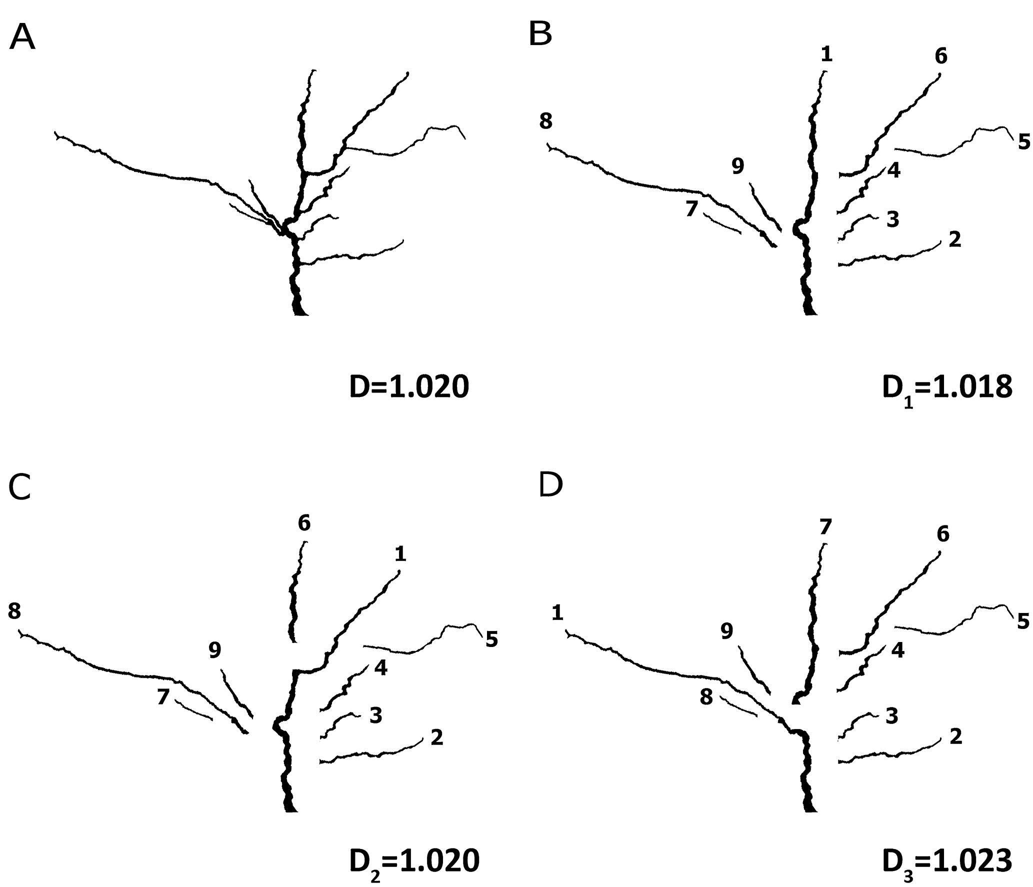

Fig. 1. Modified Richardson's method. (A) An apical dendrite. (B–D) Disassembling the apical dendrite in (A) into parts. Fractal dimension *D* is the mean of D_1, D_2 and D_3 (see text).