STUDYING THE BIOCHEMICAL STATUS OF VARIOUS TISSUES IN RATS ON A MODEL OF PARKINSON'S DISEASE UNDER THE INFLUENCE OF MELANIN

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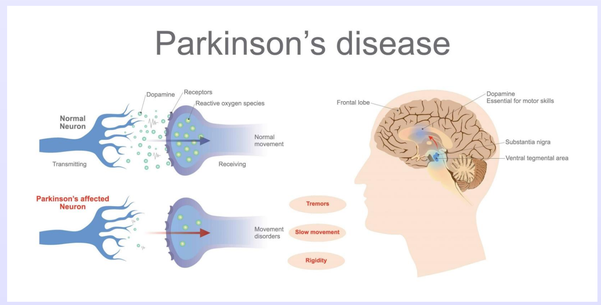
Introduction

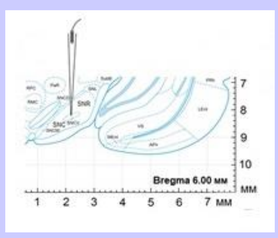
Parkinson's disease is a common movement disorder in a wide range of neurodegenerative diseases, often associated with gradual personality degradation. The main cause of Parkinson's disease (PD) is the progressive destruction and death of dopaminergic neurons in the substantia nigra pars compacta (SNc), which is accelerated by oxidative stress. There is a decrease in the activity of the antioxidant system in the brain; the antioxidant therapy is a promising direction in the treatment of PD.



In the present study, bacterial melanin (BM) was used, which has a high biological activity and a biostimulating effect. The aim of the study was to determine the quantitative and qualitative changes in the total fractions of associates from membranes (cell membranes and intracellular membranes) of rat tissues in rotenone-induced PD and under BM exposure.

Material and Methods

The study was carried out on sexually mature white rats weighing 200–250 g. Rotenone was administered under anesthesia (pentobarbital, 40 mg / kg, i.p.) at a dose of 12 μg rotenone in 0.5 μl DMSO (at a rate of 1 μl / min) into the medial forebrain bundle according to stereotaxic atlas coordinates (PD group). The BM solution was administered to the experimental animals the next day after the injection of rotenone at a concentration of 6 mg/ml (i.p.) (PD+BM group). The animals were kept under the same conditions during the entire postoperative period (4 weeks) prior to the acute experiment, and isolation and purification of the total fractions of the isoforms of NLP-Nox associates from these groups of rat tissues were performed.



Scheme of the experiment on the intracerebral administration of rotenone into the medial forebrain bundle

Then the following biochemical analyzes from these groups of rat tissues were carried out:

1. *Isolation and purification of a total fraction (O2— producing associates NLP- Nox) from cells membranes and membranes of intracellular formations of brain, lung and small intestine of the rats.*
2. *Determination of NADPH and a lipid componenin the composition of isoforms of the total fraction of NLP-Nox.*
3. *Determination of the stationary concentration ofO2−, produced by NLP-Nox isoforms presented above.*

The statistical processing: variation-statistical method of Student-Fisher, reliability criterion "p", m±M, n=6.

Results

The optical absorption spectra of the total fractions of the isoforms of NLP-Nox from the brain, lung, and small intestine membrane formations in C, PD, and PD+BM groups in oxidized and reduced states at pH 9,5 the characteristic absorbance at 412 nm, 530 nm, and 560 nm (Fig.1).

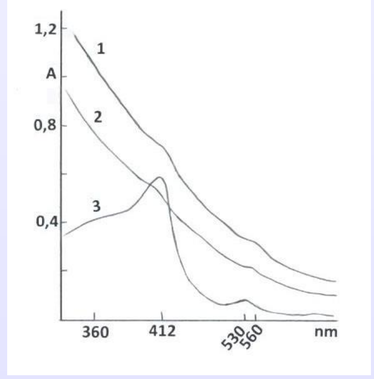


Fig 1. The optical absorbtion spectra of the weak opalescent water solutions of total fraction of isoforms of the NLP-Nox from brain (1), small intestine (2) and lung (3) tissues.

The content of the lipid component (malondialdehyde) in the presented associates of NLP-Nox from groups of PD rats is higher to 20-22%, than that in control rats. In the oxidized state, the characteristic maxima of the optical absorption spectra of the isolated NLP-Nox at 412, 530, and 560 nm were observed. In the reduced state (by potassium dithionite), optical absorption maxima were observed at 418, 540, and 558 nm. The form of these spectra did not differ for NLP-Nox from the all groups and was not observed. In the UW regions, the maximal optical absorbance of the presented associates at 260 nm, 275 nm, and 280 nm (the characteristic absorptions for the proteins) and the forms of the optical absorption spectra of isoforms of the total fraction of NLP-Nox from tissues of rats in the PD groups were similar to the spectra of control and PD+BM groups and were not present.

Tissue	C	PD	PD+BM
Brain	82,6±4,3	90,7± 5,4(p<0,001)	84.1± 3,3(p<0,005)
Small intestine	21,9 ±1,1	81,6±4,5 (p>0,001)	30,1± 3,1(p<0,001)
Lung	10,32±0,3	14,2±0,05 (p>0,005)	11,4±0,3 (p<0,001)

Table 1. Specific contents of the isoforms of NLP-Nox (mg) in 1 g tissues (mg/g) in the control (C), PD and PD+BM groups.

The increase in the amount of these associates from tissues in the PD group can be conditioned by the increase in lipid peroxidation of the membranes, and with decay of the band for the binding of the associate to the membranes. As a result, the release of NCL-Nox from the membranes to the solution in the PD groups was observed. This phenomenon could be used as a new diagnostic test for PD.

Tissue	C	PD	PD+BM
Brain	4,3±0,2	3.8±1,1 (p<0,01)	5,1±0,2 (p<0,001)
Small intestine	83,2±4,3	70,5±2,4 (p<0,005)	78,6±2,2(p<0,005)
Lung	21,8±2,1	13,5±0,4 (p<0,001)	17,7±1,6 (p<0,001)

Table 2. The stationary concentration (x 10-4 M) of produced O2− by 1 mg total fraction of the isoform of NLP-Nox was associates from rat tissues at 20 °C (M/mg) in the control (C), PD and PD+BM groups.

Conclusion

In comparison with the indices of the control group, the specific content of the total fractions of the isoforms of NLP-Nox associate from PD groups was higher, especially in the small intestine. This change may be a new mechanism for rotenone-induced PD.

The antioxidant bacterial melanin was shown to have a regulating effect on the membrane formations in the brain, lung, and small intestine by inhibiting the release of new membrane-bound thermostable formations (NLP-Nox associates) from these membranes while also regulating the stationary concentration of produced O_2^-.