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Applications of tunable diode lasers and the tunable laser diode spectroscopy in tschachotin's microbeam setups for irradiation of biological tissues, cells and cellular compartments with positional selectivity

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This report provides a historical and technical review of the UV-microbeam technique application for living cell irradiation from the early Tschachotin's works [1-16] up to nowadays and considers the use of the tunable laser diode spectroscopy tools for microbeam manipulations synchronized with in situ and in vivo microspectroscopy, laser microspectrophotometry and laser-excited spectrofluorimetry of living cells and cellular compartments. From the above considerations we propose the possibility of development of a number of tunable laser microbeam technologies, based on the Tschachotin's "UV radiopuncture", "micropuncture" and "ultramicropuncture" concepts.

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Tunable diode laser based spectral chronaximetry, adequatometry and discretometry – three novel methods for complex laser neuroophtalmological and photobiophysical measurements

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Chronaxie is a minimal time required for either muscular or nervous tissue stimulation by a constant electric current of twice the strength of the rheobase. The chronaxie measurement is called chronaximetry. In addition to the chronaximetry of muscles and neural structures there are also methods of chronaximetry for the visual system – the so-called optical chronaxie. This parameter is usually determined from the minimal time interval required for the phosphene appearance, which normally ranges from 0,7 to 2.5 msec. However, under optical stimulation (for "non-electric phosphenes") the electrophysiological effect is known to depend on the spectral range of the stimulating light. Hence, it is necessary to perform spectroscopic measurements during the study of the response at different spectral signals. The most effective transmitter with a tunable wavelength is a tunable diode laser, so it is reasonable to combine laser-optical chronaximetry and TDLS with the response measurements in different spectral ranges. In the latter method retina itself should be both an object of measurements and a light sensor. Thus, tunable diode lasers and «native photobiosensors» allow to study the adaptability, the adequacy of the stimuli and the effects, the dependence of the perception discreteness on the stimulation rhythm, including the EEG rhythm assimilation of photostimulation. All the above parameters will become the novel criteria for bio-TDLS. Optical chronaximetry has been known since 1950-th and was applied at the very first space missions for the control of the astronauts' physiological state. P.O. Makarov was the first to develop the adequate optical chronaximetry methods in his pioneering book. However, he did not suggest a spectral coherent modification of this approach, and hence, not all of his methods of adequatometry and discretometry could be implemented to neuro-ophthalmology. Later he introduced a method of adequatometry of the human olfactory analyzer and developed a general adequate stimulation theory of the sense organ activity.In addition to adequatometry another complementary method known as discretometry was also developed, which allows to study the effect of the stimuli periodicity on the electrophysiological response. We propose here to combine the three above approaches – chronaximetry, adequatometry and discretometry using an adaptive tunable laser diode system with variable frequency of rhythmic exposition and spectral feedback.

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