Mapping of fluorescent compounds in lyophilized blackcurrant (*Ribes nigrum L.*) fruits using spectroscopy and nonlinear microscopy

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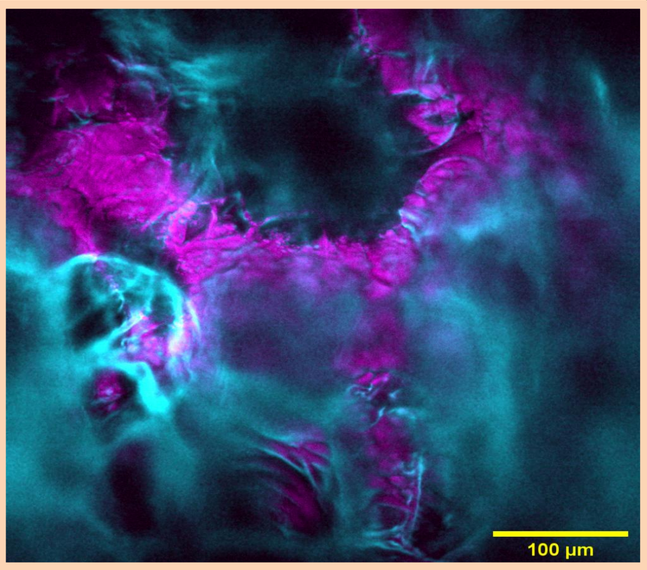
Blackcurrant (*Ribes nigrum L.*) belongs to the important medicinal plants that act preventively and therapeutically on the organism [1, 2]. Bioactive components in fruits and leaves of blackcurrant could have beneficial effects on the skin fibroblasts that produce collagen [3]. The influence of parts and extracts of this plant on erythrocyte membranes has been the subject of research in recent years [4]. Blackcurrants (*Ribes nigrum L.*) contain high levels of polyphenol anthocyanins in fruits and flavonoids in leaves that have beneficial effects on health, owing to antioxidant and anticarcinogenic properties. These compounds are responsible for the coloring of many plants, flowers and fruits. Cyanidin-3-O-glucoside (C3G) is one of the principal types of anthocyanidins and is the most common and abundant one in fruits blackcurrant [5]. Anthocyanidins/anthocyanins can be employed as probes for oxidation processes in biomedical experiments. Their advantages include biocompatibility and the lack of toxicity [6].

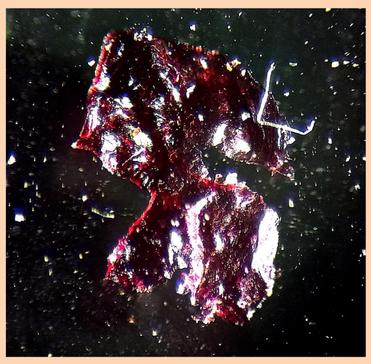
Bright field microscopy image of lyophilized black currant berry
Microscope objective 20X 0.8

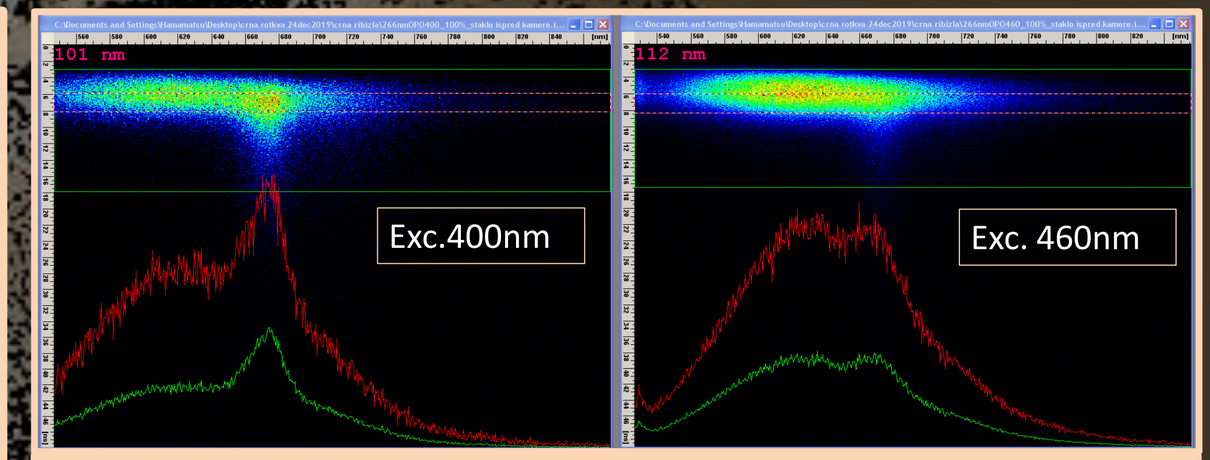


Nonlinear microscopy:

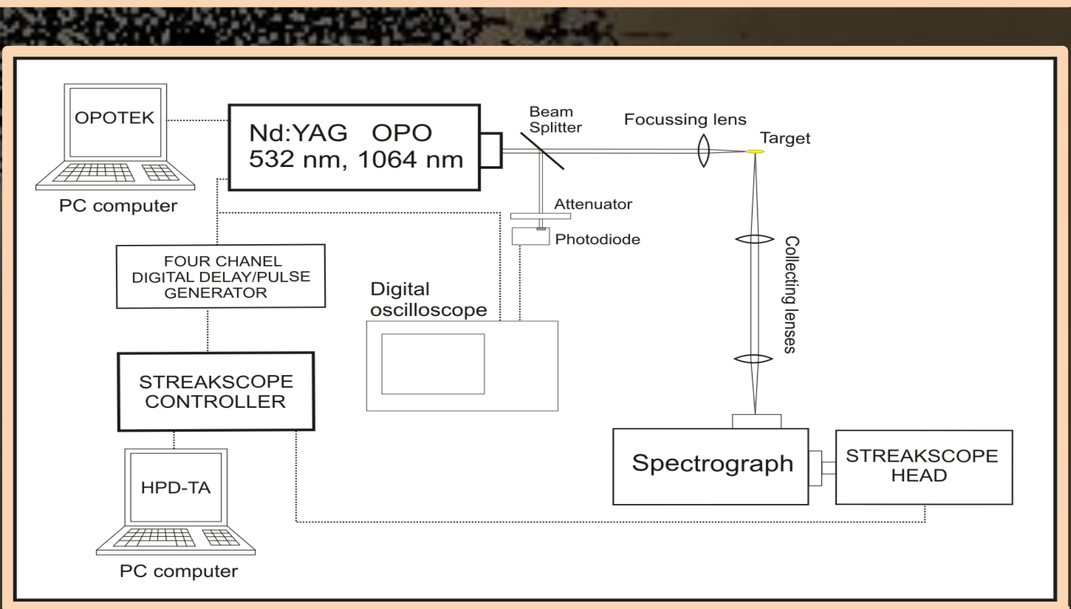
Magenta-Autofluorescence image
Two photon exc. 730nm,
Emission filter 530/40 (510-550)
Laser power 8 mW
Pulse duration 160fs
Repetition rate 76 MHz

Cyan - Upconversion image
Continuous wave exc. 730nm,
Emission filter (570 – 700)
Laser power 8mW

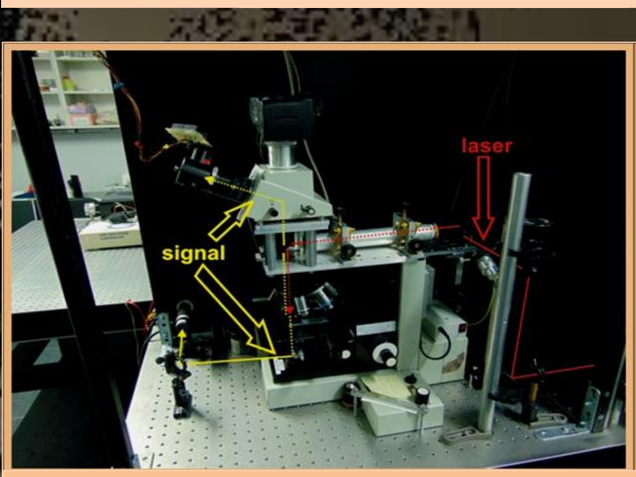




Streak images of lyophilized black currant berry
- Optical Parametric Oscillator was used for single photon excitation, tunable over a spectral range (320nm - 475nm)
- The image has been acquired in photon counting mode operation using HPD-TA software.
- The fundamental advantage of the streak scope is its two dim. nature (wavelength – horizontal axis, time- vertical axis).
- That is especially important in measuring ultra fast light events that most commonly occur in biological samples (~ps resolution).



Time resolved spectroscopy setup



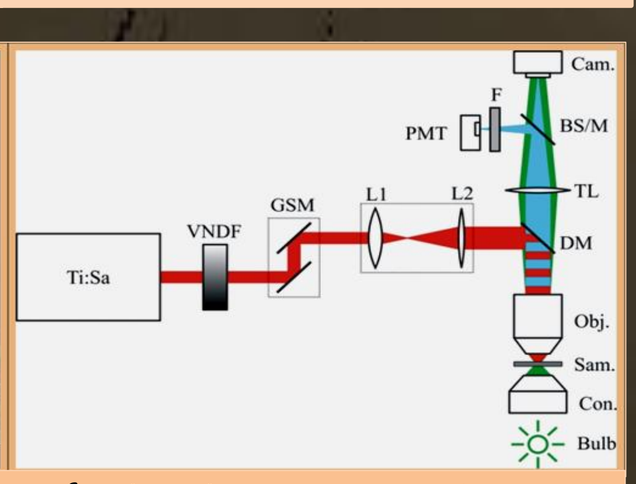


Photo and experimental scheme for imaging mesurements

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The aim of this study is to present the analysis and mapping of the Blackcurrants (*Ribes nigrum L.*) components using spectroscopy and imaging measurements [1, 7]. Time resolved optical characteristics were analyzed by using TRLS (Time Resolved Laser Spectroscopy) experimental setup [1]. Nonlinear optical properties of the plant have been studied using two-photon excited autofluorescence (TPEF), and upconversion luminescence (UCL) simultaneously [7]. The benefits of using UCL for biological applications are in reducing the photobleaching and providing photostability. Upconversion emission is also more efficient than the TPEF and SHG. Moreover, UCL could be achieved with a low power continuous wave (CW) laser.