INFLUENCE OF OXYGEN O_2 ON MICROWAVE SATURATION OF EPR LINES OF PLANTS CARBONIZED AT 650°C AND POTENTIAL APPLICATION IN MEDICINE

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Abstract

Electron paramagnetic resonance spectroscopy (EPR) was used to examination of spin-lattice relaxation in vascular plants carbonized at 650°C. Application of EPR method of continuous microwave signal saturation in medicine was proposed. The first derivative EPR spectra of pyrolysed bamboo and yucca were measured for samples in air and in vacuum. Influence of microwave power in the range 0.7-100 mW on amplitudes of EPR lines were evaluated. It was stated that amplitudes of the studied carbon materials increase with increasing of microwave power in the studied range. Saturation of EPR lines was not observed, so fast spin-lattice relaxation processes exist in the analyzed materials. Changes of EPR amplitudes for samples in air were slower than for evacuated samples. This effect may be used for determination of oxygen contents in the biological cells cultures and for analysis of optimal parameters of photodynamic therapy of cancer.

Keywords: *carbonized plants, electron paramagnetic resonance, EPR, microwave saturation, paramagnetic centers, oximetry*

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Introduction

Samples sensitive to oxygen contents in the environment are used to examination biochemical processes and conditions of medicinal therapies [1-6]. Application of carbon materials in oximetry is known [2-4]. Carbon materials, especially fusinite [2-3] are ambient for cells. It is difficult to obtain fusinite so the new carbon probes for oximetry were searching in this work. Microwave saturation of EPR spectra depends on the sample environment [7-8]. Paramagnetic coal samples physically interact with oxygen molecules [9-10].

In this work we continue EPR studies of carbonized plant materials [11-12]. Characteristic of paramagnetic centers in yucca and bamboo carbonized at 550 and 950°C for samples in contact with air was presented earlier [11-12]. In this work we search EPR parameter dependent on oxygen content in the sample environment and which is useful to oximetry. Correlations between microwave saturation of EPR lines of the paramagnetic carbonized materials and O_2 contents in the tubes with sample were examined.

Materials and methods

Bamboo (Bambusa vulgaris) and yucca (Yucca flaccid) pyrolysed at 650°C in the neutral atmosphere were studied. Microscopic images of bamboo and yucca carbonized at 650°C at different cross-sections are shown in FIG.1-2 (a,b,c).

The spectroscopic measurements for samples in air and in different degrees of vacuum were done by the electron paramagnetic resonance spectrometer at X-band (9.3 GHz) with magnetic modulation of 100 kHz. Microwave frequency was recorded. EPR measurements were done for samples in air and in vacuum (900-10^{-5} mbar). Changes of amplitudes and linewidths of EPR lines of the studied materials with increasing of microwave power from 0.7mW to 100mW were measured. g-Values were calculated from resonance condition by the use of resonance magnetic field and microwave frequency.

Results and discussions

No EPR lines were measured for original plant samples. Asymmetrical EPR spectra were recorded for bamboo and yucca carbonized at 650°C. After bamboo and yucca heating at the mentioned temperature paramagnetic centers appear in these samples and strong EPR spectra were recorded. Dependencies of these spectra on microwave power and paramagnetic oxygen molecules content in sample environment were observed. Exemplary EPR spectra of carbonized bamboo and yucca for samples in air are presented in FIGURES 3 and 4, respectively. Exemplary EPR spectra of carbonized bamboo and yucca for evacuated samples (10^{-5} mbar) are shown in FIGURES 5 and 6, respectively. EPR spectra recorded with attenuations from 15 dB to 0.5 dB were compared (FIGs.3-6). Decrease of attenuation from 15 dB to 0.5 dB led to increase of microwave power (FIGs.3-6). All effects discussed in this paper increase with increasing of vacuum degree from 900 mbar to 10^{-5} mbar. In this work we shown representative results for the highest degree of vacuum (10^{-5} mbar).

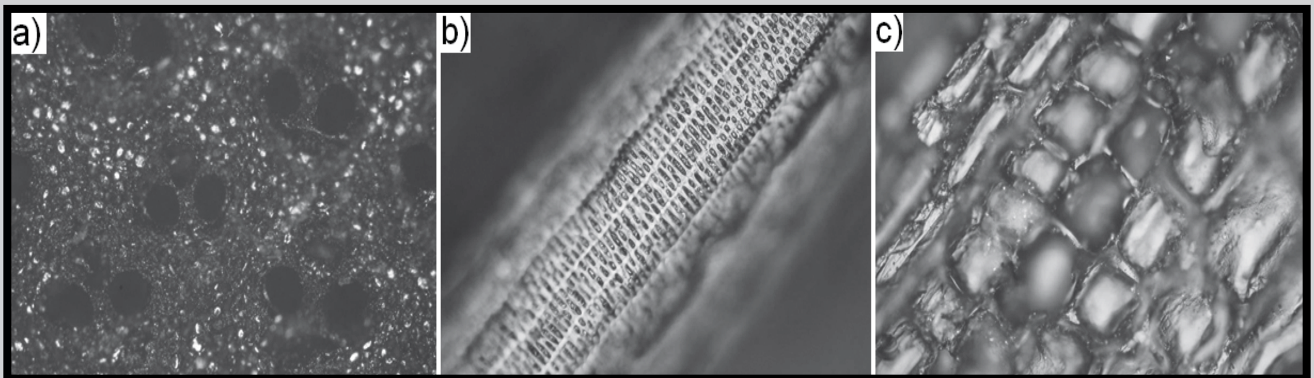

FIG.1. Various sections of bamboo carbonized at 650°C: a) cross-section of the stem – internal structure; magn. 80x, in air; b) intersection along the stem – internal structure of single fibre; magn. 500x, in oil; c) intersection along the stem – the structure of interfibrous area; magn. 500x, in oil.

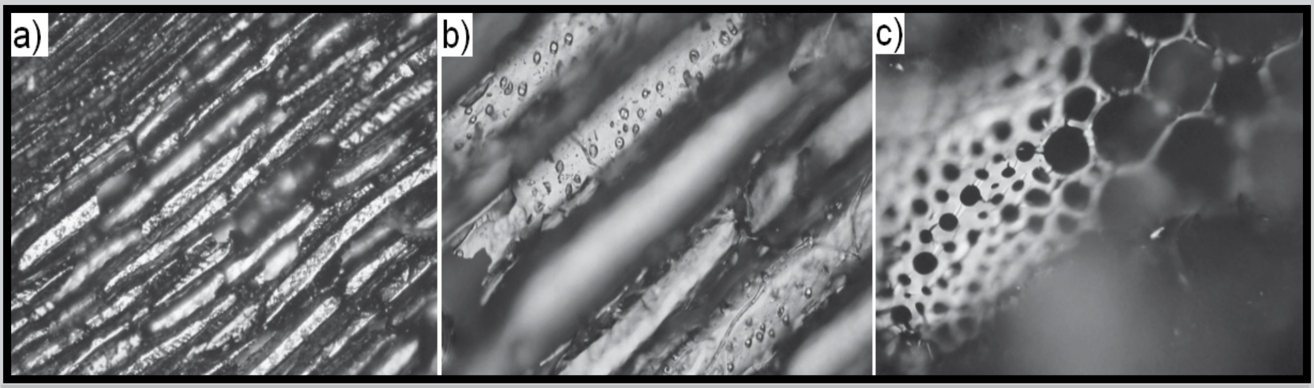

FIG.2. Various sections of yucca carbonized at 650°C: a) Intersection along the stem. Fibrous structure; magn. 160x, in air; b) Intersection along the stem. Fibrous structure. Pores in walls of fibres; magn. 500x; c) Cross –section of the stem. Various diameters of fibres and different thickness of walls; magn. 500x, in oil.

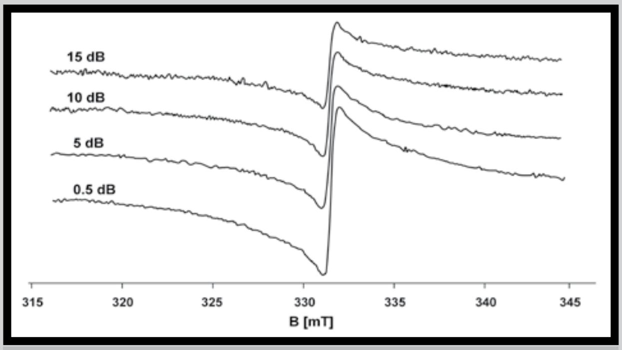

FIG.3. EPR spectra of bamboo carbonized at 650°C for sample in air. The lines were recorded with attenuations (dB): 15, 10, 5 and 0.5.

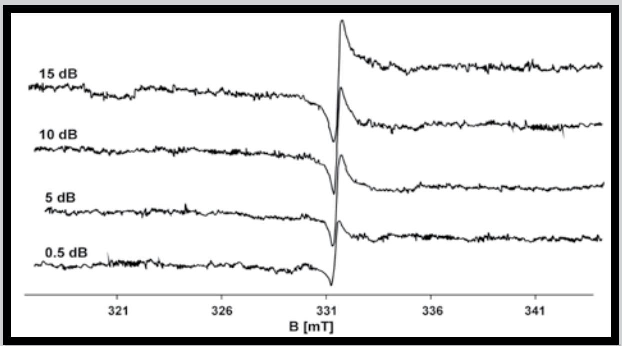

FIG.4. EPR spectra of yucca carbonized at 650°C for sample in air. The lines were recorded with attenuations (dB): 15, 10, 5 and 0.5.

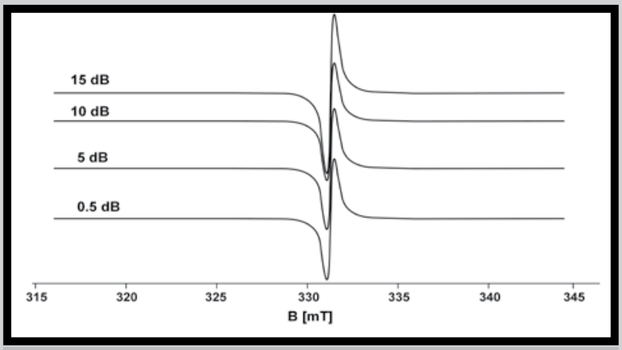

FIG.5. EPR spectra of bamboo carbonized at 650°C for sample in vacuum (10^{-5} mbar). The lines were recorded with attenuations (dB): 15, 10, 5 and 0.5.

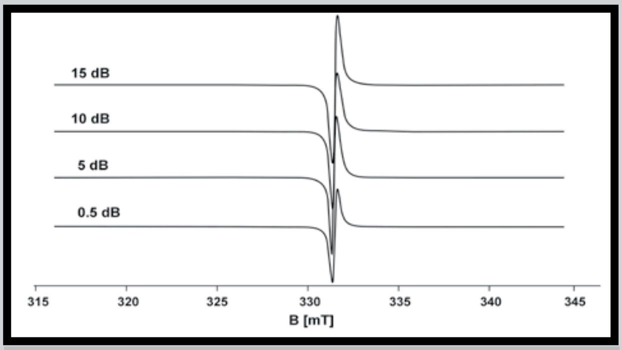

FIG.6. EPR spectra of yucca carbonized at 650°C for sample in vacuum (10^{-5} mbar). The lines were recorded with attenuations (dB): 15, 10, 5 and 0.5.

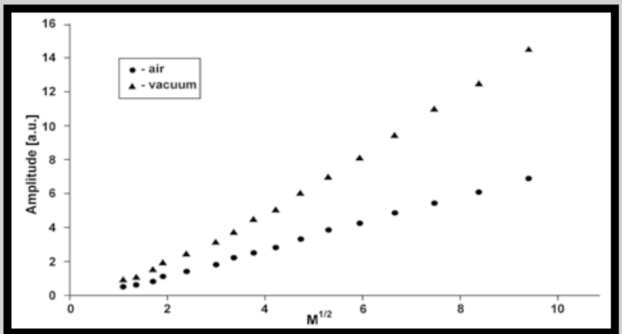

FIG.7. Influence of microwave power (M) on amplitudes of EPR lines of bamboo carbonized at 650°C for samples in air and in vacuum (10^{-5} mbar).

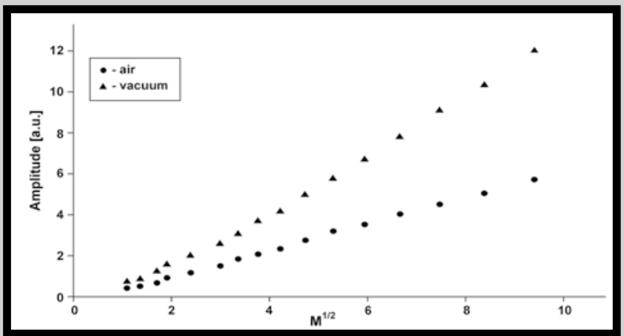

FIG.8. Influence of microwave power (M) on amplitudes of EPR lines of yucca carbonized at 650°C for samples in air and in vacuum (10^{-5} mbar).

Linewidths of EPR spectra strongly changes after removing of paramagnetic O_2 molecules from the tube where samples were located. EPR lines for evacuated samples of both carbonized bamboo and carbonized yucca (FIGs.5-6) were narrower than EPR lines of sample in contact with O_2 (samples in air) (FIG.3-4). Broadening of EPR lines in air results from dipolar interactions of paramagnetic centers of carbonized materials with paramagnetic oxygen molecules.

Linewidths of the recorded EPR spectra increase with increasing of microwave power. This correlation is characteristic for homogeneous broadening of resonance curves [7-8]. It can be concluded that paramagnetic centers are homogeneously distributed in the tested carbonized plant materials. Carbonization process indicates rupturing of chemical bonds in the whole heated plant materials.

g-Values of paramagnetic centers in the carbonized materials were 2.0030. Mainly paramagnetic centers with unpaired electrons localized on carbon or nitrogen atoms exist in the heated plants samples. g-Factor did not change with degree of vacuum and it was the same for evacuated samples as for samples in air.

EPR spectra depend on microwave power used during this experiment. It was stated that amplitudes of EPR lines of the studied carbon materials increase with increasing of microwave power up to 100 mW (FIGs.7-8). Microwave saturation effect was not obtained for the recorded spectra. It means that fast spin-lattice relaxation processes [7-8] exist in the carbonized plant materials. Amplitudes of the EPR lines did not reach the maximal value and the decrease of amplitudes with increasing of microwave power at saturation conditions was not observed. Differences in microwave saturation of EPR spectra of the carbonized samples in air and in vacuum are clearly visible. EPR lines of evacuated samples changes faster with microwave power and the lines of samples in air begin saturate. Additionally amplitudes of EPR lines of the analysed plant materials are lower for samples in air than for evacuated samples (FIGs.7-8). Quasi-chemical bonds between oxygen molecules and samples are responsible for this effect. Similar correlations were observed for coals [9-10].

Results obtained in this work may be developed and used in medicine. It was proved for plant materials that not only amplitudes and linewidths, but also microwave power of microwave saturation of their EPR lines depends on oxygen contents in the sample environment. Correlations between microwave saturation of EPR lines and oxygen contents in the environment may be used in oximetry for photodynamic therapy. Excited singlet oxygen molecules and free radicals damage tumor cells during photodynamic therapy [13-14]. Measurements of microwave saturation of EPR lines of bamboo and yucca carbonized at 650°C located in laser irradiated cells give information about singlet oxygen formation. Increase of diamagnetic singlet oxygen contents in the biological system refers to decrease of paramagnetic triplet oxygen in ground state. Higher formation of singlet oxygen is accompanied by stronger changes in microwave saturation.

Conclusions

Continuous microwave saturation of EPR spectra of bamboo and yucca carbonized at 650°C indicates that:

1. Strong paramagnetism characterizes the studied plant materials carbonized at 650°C for samples in both air and vacuum.

2. Paramagnetic O_2 molecules quench EPR lines of the carbonized materials.

3. Fast spin-lattice relaxation processes exist in the studied plant samples.

4. Paramagnetic oxygen molecules O_2 change spin-lattice relaxation processes in the plant materials carbonized at 650°C.

5. Dependence of microwave saturation of EPR lines of the carbonized plants with degree of vacuum may be used for determination of oxygen contents in biological cells cultures and for analysis of optimal parameters of photodynamic therapy of cancer.

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