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***Abstract*— The terahertz absorption spectra of sodium magnesium chlorophyllin (Chl-Mg-Na) and sodium copper chlorophyllin (Cu-Chl), two major members of the chlorophyll derivative family, have been measured in the range 0.2−2.5 THz, at room temperature. The capability of terahertz spectroscopy for quantitative characterization of Chl-Mg-Na intermolecular vibrations was investigated and the sensitivity of transitions with degree of hydration by changes in the molecular environment was examined. For the Cu-Chl derivative, a broad feature was observed around 1.8 THz which currently hinders clear Cu-Chl identification and quantification.**

I. Introduction

The sensitivity of the Terahertz (THz) spectroscopic techniques allows to obtain unique THz vibration modes in the 0.1-3.5 THz frequency range, resulting from the molecular packing interactions within a crystal structure. Consequently, many molecular crystals have been investigated using THz techniques[1-3]. Sodium magnesium chlorophyllin (Chl-Mg-Na) and sodium copper chlorophyllin (Cu-Chl), shown in Fig. 1, are two major members of the semisynthetic derivatives of the natural chlorophyll *a* (Chl-*a*). Previous reports on natural chlorophylls and on semisynthetic derivatives mainly focused the ultraviolet-visible and infrared regions of the electromagnetic spectrum.

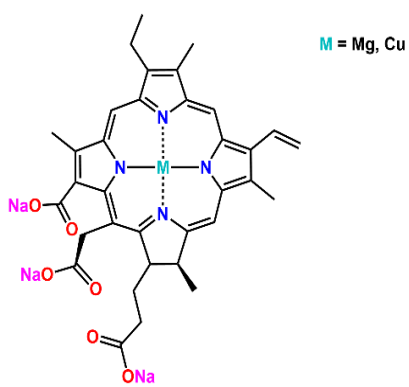

Fig. 1. Structure of the sodium magnesium chlorophyllin ($C_{34}H_{31}MgN_4Na_3O_6$, M=684.92 g.mol^{-1}) or sodium copper chlorophyllin ($C_{34}H_{31}CuN_4Na_3O_6$, M=724.15 g.mol^{-1}).

In this study, we used THz time-domain spectroscopy (THz-TDS) to cover the spectral absorption range from 0.2 to 2.5 THz in order to investigate optical properties of these two major chlorophyll derivatives. Frequency dependent absorption coefficient and refractive index were extracted. In order to acquire further understanding of the observed low-frequency vibrational modes, the THz spectra of the polycrystalline Chl-Mg-Na were analysed as a function of time, under dehydration conditions.

II. Methods

Powdered sodium magnesium chlorophyllin and sodium copper chlorophyllin (commercial grade) were purchased from Catalyons and Alfa Aesar, respectively. The powders were used without further purification. The pure Chl-Mg-Na samples were prepared using as-received pure powder and pressed to pellets. with a 13 mm diameter. In order to record strong absorption features which can be saturated in the pure samples, mixed pellets with high-density polyethylene (HDPE) powder at various dilution ratios were also prepared. For Cu-Chl sample, a recrystallisation in water was done before the fabrication of pure pellets. Table 1 summarizes the different pellet fabrication conditions used. The THz-TDS spectra were acquired using a commercial TeraPulse 4000 instrument (TeraView Ltd, Cambridge, UK) across the spectral range of 0.1-3.5 THz with frequency resolution of 0.002 THz. The sample compartment was evacuated to reduce the effect of the water vapor in the spectra, which also affected the degree of hydration of the sample. All measurements were made with a transmission configuration. The refractive index and absorption coefficient of the pellets (pure or mixed) were extracted by using the Fit@TDS software developed by Peretti et al.[4, 5] This software enables one to model a single uniform layer and add oscillators to take into account peaks from mixed intermolecular and intramolecular vibrational modes and then retrieve the optical parameters from the THz-TDS data.

Table 1. Mixture composition (in weight-percent (wt-%)), thickness and total weight of the pellets containing different relative amounts of HDPE and Chl-Mg-Na, and Cu-Chl. Extracted $n(\omega)$ and $\alpha(\omega)$

Pellet code	Mixture composition	Thickness (µm)	Total weight (mg)	Refraction index (<1.4 THz)	Absorption losses (cm^{-1}) (<1.4 THz)
A	100 wt-% Chl-Mg-Na	574	97.9	1.87	50
B	100 wt-% Chl-Mg-Na	408	77.8	2.12	45
C	50.0 wt-% Chl-Mg-Na	2324	302.6	1.58	25
D	100 wt-% recry-stallized Cu-Chl	560	77.2	1.7	35

III. Results

THz-TDS data were used to extract the refractive index and absorption coefficient of the chlorophyll derivatives. The complex transmission coefficient is given by $\tilde{T}(\omega) = \tilde{E}_{sample}/\tilde{E}_{reference}$, where, $\tilde{E}_{sample}$ and $\tilde{E}_{reference}$ are the Fourier transforms of the time-domain signals with and without a sample. ω is the angular frequency given by $\omega = 2\pi f$, where f is the frequency of the radiation in Hertz. $\tilde{T}(\omega)$ is related to the complex refractive index $\tilde{n}(\omega) = n(\omega) - i\,k(\omega)$, where the real part $n(\omega)$ corresponds to a delay in the time-domain and the imaginary part $k(\omega)$ is the extinction coefficient which represents absorption losses in the material. The absorption coefficient $\alpha(\omega)$ is related to the extinction coefficient $\alpha(\omega) = 2\omega\, k(\omega)/c$. Fig. 2 represents the experimental and fitted results of $n(\omega)$ and $\alpha(\omega)$ obtained for the pellet A containing 100% of Chl-Mg-Na. As previously mentioned in Ref. 6, for Chl-Mg-Na, four clear resonances are observed at 1.44 THz, 1.64 THz, 1.82 THz, and 2.03 THz. However, for the Cu-Chl (pellet D)[6], only a very broad feature was observed around 1.8 THz, despite a recrystallization of the product.

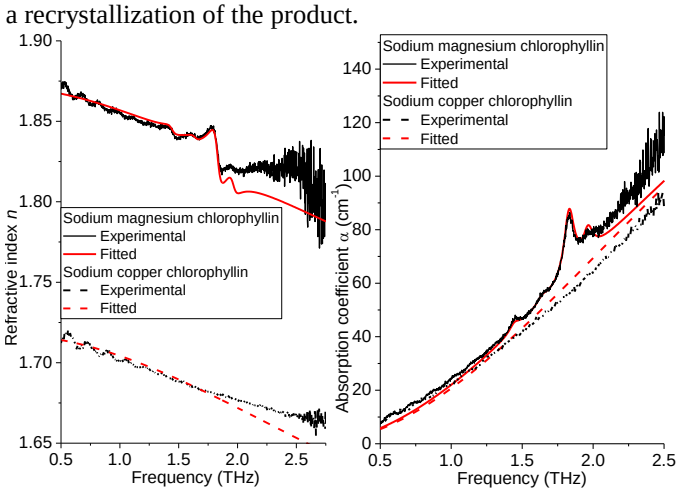

Fig. 2. Comparison between experimental (black) and fitted (red) spectra obtained for a pellet composed with 100% of Chl-Mg-Na (pellet A) and 100% of Cu-Chl (pellet D). a) refractive index $n(\omega)$ and b) absorption coefficient $\alpha(\omega)$.

Table 1 lists the resulting fit parameters extracted from the pellets. Below 1.4 THz, the Chl-Mg-Na material has a constant refractive index $n(\omega)$ of approximately 1.87, whereas for Cu-Chl, a smaller value of ca. 1.70 was observed. The absorption coefficient of Chl-Mg-Na and of Cu-Chl remains <50 cm^{-1} below 1.4 THz.

In polycrystalline materials, the molecular environment can be significantly change using small amounts of solvents. Fig. 3 shows the time-dependent shift of the central frequency of the three well-marked features for the Chl-Mg-Na sample (pellet C) during dehydration. As the sample dries, the feature at 1.82 THz shifts towards lower frequencies and a decrease in intensity is observed. However, no drastic changes were observed for the 1.44 THz and 1.64 THz features. This suggest that the resonance at 1.82 THz relies on important contributions from intermolecular interactions.

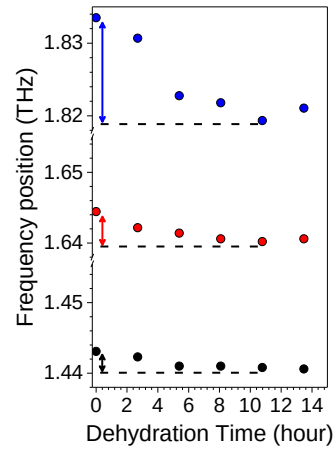

Fig. 3. Time-dependent frequency changes of the well-marked features of Chl-Mg-Na (pellet C) as a function of dehydration time under vacuum. A frequency shifts of 0.015 THz (blue arrow) was observed for the feature at 1.82 THz.

IV. Conclusion

THz spectroscopy allows to probe the frequencies of the low-frequency vibrations involved in chlorophyll derivatives; 1) to determine the optical parameters such as refractive index and absorption coefficient, and 2) to observe their shift and amplitude variation with the degree of hydration. The measured THz spectra of these chlorophyll derivatives are a significant contribution to the collection of biomolecular crystals spectra which are currently accumulated in the literature. Promoting such investigations to chlorophyll in plants offers a rich but challenging field of study[6,7].

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