

WINE QUALITY TESTING BY USING TERAHERTZ SPECTROSCOPY

A THESIS SUBMITTED TO
THE GRADUATE SCHOOL OF NATURAL SCIENCES
OF
MIDDLE EAST TECHNICAL UNIVERSITY

BY

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IN PARTIAL FULFILMENT OF THE REQUIREMENTS
FOR
DEGREE OF MASTER OF SCIENCE
IN
CHEMISTRY

JULY 2018

Approval of the thesis:

WINE QUALITY TESTING BY USING TERAHERTZ SPECTROSCOPY

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ABSTRACT

WINE QUALITY TESTING BY USING TERAHERTZ SPECTROSCOPY

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July 2018, 96 pages

In this study, a recently developing technique called Terahertz (THz) Spectroscopy is being tested for its use in wine analysis. The technique is cost effective, reproducible and harmless to the sample. Thus, its applications in pharmacy, security and food analysis have increased recently. THz spectroscopy is a powerful technique since it is able to measure the electric field and, therefore, provides information about frequency dependent absorption coefficient and refractive index. In this thesis, a preliminary study on wine chemicals and on wine quality analysis were carried out by both THz-TDS and FT-THz spectrometers. The steps were i) determination of an optimum sample cell parameters for both THz-TDS and FT-THz measurements for ~10-15% (v/v) ethanol and water mixtures, ii) determination of chemicals that are most effective in wine quality in terms of color, scent and especially the taste of wines, iii) collection of time domain and frequency domain spectra of all the chemicals and water ethanol mixtures, iv) collection of time domain spectra of all the wine samples and v) application of Principle Component Analysis (PCA) to the collected data. The spectra of wine chemicals were collected in pellet form, then in two solution forms (solid solution with polyethylene (PE) as solvent and saturated liquid solution with pure ethanol) in order to determine the main spectral features of the chemicals. Distinct and well resolved absorption features were observed and most of these bands appear to be associated with intramolecular motions. On the other hand, the results of PCA analyses of the absorbance, refractive index and dielectric properties of wines obtained from THz-TDS spectra were used on classification of wines according to their alcohol content, grape types, and more.

Though the results are very promising, they show that a controlled sample set and many more wine samples are needed for a better classification.

Keywords: Terahertz, Terahertz Time Domain Spectroscopy, Wine Analysis, Principal Component Analysis

ÖZ

ŞARAP KALİTE ANALİZİNDE TERAHERTZ SPEKTROSKOPİ İNCELEMESİ

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Temmuz 2018, 96 sayfa

Bu çalışmada son zamanlarda gelişmekte olan Terahertz (THz) spektroskopi tekniğinin şarap analizi üzerine uygunluğu test edilmiştir. Teknik, örneğe zarar vermeyen, tekrarlanabilir ve uygun maliyetli olmasıyla gıda analizi, ilaç endüstrisi ve güvenlik alanlarında kendine hızla yer bulmaktadır. THz spektroskopi ölçümlerini direkt olarak elektrik alanını ölçerek gerçekleştirebilmesi sebebiyle hem frekansa bağlı soğurma katsayısı hem de kırıcılık indisi analizleri yapabilmektedir. Bu tezde, şarap kimyasallarının ve şarap örneklerinin analizi üzerine ön çalışmalar gerçekleştirilmiştir. Çalışmadaki adımlar, i) 10-15% (h/h) etanol çözeltileri kullanılarak Zamana Dayalı Terahertz Spektrometre (ZDTS) ve FT-THz ölçümleri için örnek haznelerinin optimize edilmesi, ii) şarapta tadı, kokuyu ve rengi diğerlerine kıyasla daha fazla etkileyen şarap kimyasallarının belirlenmesi, iii) şarap kimyasallarının ve su-etanol karışımlarının hem zaman alanında hem de frekansa bağlı spektrumlarının toplanması, iv) şarap örneklerinin zaman alanında spektrumlarının alınması ve v) Temel Bileşenler Analizinin (TBA) veriler üzerinde uygulanmasıdır. Kimyasalların temel soğurma özelliklerinin belirlenmesi adına saf pelet spektrumlarına ek olarak derişik sıvı formunda ve polietilenli (PE) katı pelet formlarında çözeltileri hazırlanmış ve spektrumları toplanmıştır. Belirgin ve ayrışık soğurma bantları bu spektrumlarda gözlemlenmiş ve bu sinyallerin birçoğunun molekülün kendi hareketleri sonucu olarak gözlemlenen bantlar olduğu belirlenmiştir. Bununla birlikte, örneklerin soğurma katsayıları, kırıcılık indisleri ve dielektrik özellikleri kullanılarak yapılmış TBA analizi şarap örneklerinin yapıldığı

üzüm cinsi, örneklerin alkol oranı ve diğer çeşitli bilgiler kullanılarak sınıflandırma yapılabilmesine olanak sağlamıştır. Sonuçlar umut verici olsa da daha kesin bir sınıflandırmanın yapılabilmesi için daha kontrollü bir örnek setinin kullanılmasına ve daha fazla şarap örneğinin ölçülmesine ihtiyaç olduğu görülmektedir.

Anahtar Kelimeler: Terahertz, Zamana Dayalı Terahertz Spektroskopisi, Şarap Analizi, Temel Bileşenler Analizi

To my lovely sister, mother and father...

ACKNOWLEDGEMENTS

I would like thank to Dr. Okan Esentürk for his supervision throughout my studies and especially for the patience that he showed to me during both my undergraduate research studies and this thesis.

I would like to thank to my colleagues Dr. Emine Kaya, Yusuf Samet Aytekin, Alim Yolalmaz, Zühra Çınar, Pelin Yıldız, Bahar Atik, Orçun Tolga Şengöz and Zeynep Suvacı for their kind friendship and support.

I would like to thank to my thesis committee members for honouring my thesis defence and broadening my horizon with their valuable comments and questions.

I would like to acknowledge TÜBİTAK (215Z449) and METU-BAP for the financial support.

Finally I sincerely thank to my family for their endless support.

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LIST OF SYMBOLS AND ABBREVIATIONS

CW	: Continuous Waveform
FFT	: Fast Fourier Transform
fs	: Femtosecond
FT-THz	: Fourier Transform Terahertz Spectroscopy
GaAs	: Gallium Arsenide
GaP	: Gallium Phosphide
GaSe	: Gallium Selenide
HDPE	: High Density Polyethylene
InAs	: Indium Arsenide
iPCA	: Interdigital Photoconductive Antenna
LiNbO ₃	: Lithium Niobate
mW	: Milliwatt
OAPM	: Off-axis Parabolic Mirror
OR	: Optical Rectification
PC	: Principal Component
PCA*	: Photoconductive Antenna
PCA	: Principal Component Analysis
PE	: Polyethylene
ps	: Picoseconds
SNR	: Signal-to-noise ratio
TBA	: Temel Bileşenler Analizi
THz	: Terahertz
THz-TDS	: Terahertz Time-Domain Spectroscopy
Ti:Sapphire	: Titanium Sapphire
TPX	: Polymethylpentene
TRTS	: Time Resolved Terahertz Spectroscopy
ZnTe	: Zinc Telluride
ZDTS	: Zamana Dayalı Terahertz Spektroskopisi

CHAPTER 1

INTRODUCTION

Spectroscopy is a technique in which light is used to characterize physical and chemical properties of materials. The changes in the characteristics of an electromagnetic radiation before and after its interaction with a matter is monitored by spectroscopy. There are several types of spectroscopic techniques depending on the frequency of the light that is used. Those lie on a range in the electromagnetic spectrum from low energy radio waves to high energy gamma-rays, including T-rays that is used in this thesis.

1.1 Terahertz Radiation

Molecules show several types of reactions when they interact with a light beam with different frequencies. For the Infrared and Microwave Spectroscopy, quantification or qualification is achieved via observation of motion of molecules. Terahertz rays lie between microwave and infrared regions on the electromagnetic spectrum [Figure 1]. The energy of rotational motions of a molecule matches with the energy of light waves in the microwave region level. On the other hand, vibrational motions have the energy in the infrared region level. Thus, the energy of light waves in the terahertz region matches with the low energy vibrational motions, motions of hydrogen bonded molecules and the rotational motions of molecules¹

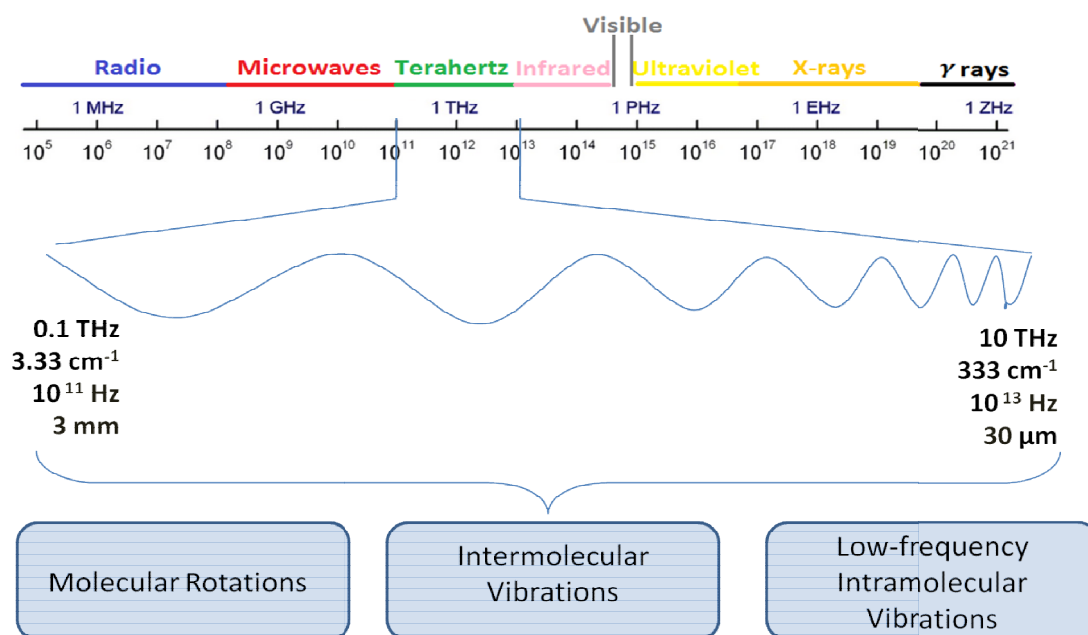


Figure 1. Electromagnetic spectrum

1.2 Development of Terahertz Spectroscopy

Terahertz region in the electromagnetic spectrum was called as “Terahertz Gap” for many years because of lack of technology to generate and detect THz waves. Even today, the technology is still limited in generation and detection of the THz waves, which limits the application of THz spectroscopy in many fields. As the technology has advanced and higher frequencies can be achieved in microwave region using electronic components, it becomes possible to produce very low energy terahertz waves over the years. On the other hand, with the development of high energy, ultra-broadband lasers, it become feasible to produce broad THz waves from the optical side. By combining both techniques, Terahertz science started to show itself in 1970s. Yang et al. produced the first Terahertz radiation in 1971 by using an intense laser and a crystal². With the fast development of ultrafast lasers, intense lasers became more and more widespread in science and engineering. In 1980s, it was realized that photoconductive switching method could also be used for generation of

higher frequency microwave pulses and a draft of THz-TDS was able to reveal itself with set of experiments conducted in late 1980s³⁻⁸. Auston, Smith and Nuss published an article in 1988 that reports the generation and detection of THz pulses successfully in the range of 0.1-2.0 THz with the pulse durations less than 1 picosecond by using 50 μm photoconductive antennas⁹. The first commercial Terahertz spectrometer was produced in 2000¹⁰. Even though there are many companies that produce built-in spectrometers nowadays, still it is more common to construct and use them in laboratories.

1.3 Generation and Detection Methods

Two main generation and detection techniques are going to be mentioned in this study. Photoconductive antennas (PCAs*) and birefringent materials can be used for both generation and detection of THz pulses.

There are two main parts in PCAs*: semiconductor material and metal rods. Semiconductor material needs to have bandgap energy less than the energy of laser pulse. Also, they should have high charge carrier mobilities, because it leads to fast rise times¹¹. For fabrication of PCAs*, GaAs wafer is usually chosen as the semiconductor material. In this technique, incident laser pulse creates electron-hole pairs in the semiconductor and then they are accelerated with the application of bias voltage through metal rods on itself. Finally, acceleration leads to the emission of radiation in Terahertz frequency¹¹.

Optical rectification (OR) is another technique that is used for Terahertz generation. Hu and Xu were able to generate pulses with Terahertz frequency by using femtosecond optical pulses and electro-optic materials^{12,13}. This technique can produce very broadband THz radiation. In the technique, incident optical laser pulses interact with an electro-optic material such as ZnTe. The different frequencies present in this ultrashort and broadband pulse mix with each other and result a broadband THz radiation generation. Even up to 100 THz (3300cm^{-1}) range radiation generation was observed in 2005¹⁴. One of the disadvantages of using OR method

was the energy of Terahertz pulses generated per frequency because they were relatively lower than the ones generated with PCAs*; however, high energy Terahertz generation up to 1.5 μJ with 3 THz range was observed in 2007¹⁵.

Terahertz detection with PCAs* relies on the similar procedure with the generation with PCAs*. When the optical laser pulse arrives to PCA*, it creates electron-hole pairs in the semiconductor material; however, there is no external bias voltage applied. Instead, acceleration is achieved via incoming THz pulse. After acceleration of electron-hole pairs, a current is generated that can be read on computer. Lock-in amplifier can also be used in that case in order to increase SNR. Finally, an exact image of THz pulse can be observed on the computer.

For the electro-optical detection technique, a birefringent crystal is used. While GaP, InAs and GaSe were used in order to detect THz pulses¹⁶, the commonly used one is ZnTe crystal¹⁷. Without terahertz beam, there is no amplitude difference between s and p components of the pulses coming from the detection arm. As the terahertz beam interacts with the detection crystal, refractive index of the crystal changes in magnitude proportional to the THz amplitude. Therefore, pulses coming from detection arm interact with the crystal whose refractive index is changing by the terahertz pulse with the help of the delay of the terahertz pulse in time. Then, by using Wollaston prism and balanced photodiode, this amplitude difference can be read and shown on computer¹¹.

1.4 Terahertz Spectroscopy Techniques

There are two commonly used Terahertz spectroscopy techniques introduced here: Time-Resolved Terahertz Spectroscopy (TRTS) and Terahertz Time-Domain Spectroscopy (THz-TDS).

TRTS briefly gives information about dynamic properties of the samples. Optical pulses generated from an ultrafast laser is used both to pump and probe the sample and used as well as Terahertz generation. After photo-excitation of the sample, time dependent Terahertz spectrum is obtained¹¹.

THz-TDS is the most common Terahertz spectroscopy technique. In this technique, briefly, laser light is splitted into two arm by using a beamsplitter. One arm is called the generation arm and other one is called the detection arm. The light in the generation arm hits to THz wave generator (PCA*, electro-optic crystal etc). Then, generated THz pulse travels through the sample and arrives to detection component (PCA*, electro-optic crystal etc.). On the other hand, optical pulses coming from detection arm also arrives to detection component. By using other optical components, electric fields in the presence and absence of sample are detected manually. With application of Fourier transform, absorbance information can be obtained after conversion of time domain data to frequency domain data. At the same time, information about refractive index and dielectric properties of samples can be investigated by using THz-TDS¹⁷.

1.5 Applications of Terahertz Spectroscopy

Today, Terahertz technology finds a growing interest both in scientific application areas and in industry as a tool for characterization of both the static and dynamic properties of the materials. Terahertz spectroscopy has many advantages that make it either a complementary technique or a competitive technique in many fields. THz waves have longer wavelength and lower energy, compared to UV-Vis, Near-IR and Mid-IR regions of the electromagnetic spectrum; therefore, the technique is non-destructive, in other words it is safe for many applications such as characterization of explosives¹⁸ or biological materials¹⁹. In addition, most of non-metal materials are transparent to Terahertz waves, hence technique can analyze samples that are behind the non-metal barriers. One of the most important features of Terahertz spectroscopy is its ability to make measurements in time domain or time dependent. Therefore, analyses are faster compared to other spectroscopic techniques. In addition the technique measures electric field from both absorbance and index, hence the dielectric information can be deduced. With such preferred properties, technique has the opportunity to reveal itself day by day. Up to now, Terahertz spectroscopy was applied in pharmaceutical industry^{20,21}, petroleum industry^{22–24}, art conservation²⁵,

and medicine^{26,27}. Additionally, it is widely used for analyses of large molecules^{28,29} and DNA³⁰. Terahertz spectroscopy technique is highly sensitive to water, which draws attention to studies related to analyses of samples containing water^{31,32}.

With its non-destructive property along with being able to penetrate through the non-metal materials, THz spectroscopy is becoming a viable technique for food analysis also³³. Baek et al. studied melamine in different food matrices by using THz-TDS and Terahertz imaging and strong absorption features observed between 0.1-3.0 THz frequencies provided the information for both qualitative and quantitative analysis of the compound³⁴. Additionally, research on analysis of antibiotic residues in different food matrices was conducted and results show potential usage of THz-TDS even in the production line of food products³⁵.

Wine is a final product of the fermentation between sugar and yeast in grape juice. It includes approximately 14-15% ethanol that is produced after the fermentation. It is one of the most famous and consumed alcoholic beverages in the world. Therefore, wine analysis is seen as an emerging field between researchers among the world. Besides the research related to its anti-oxidant activity³⁶, the factors that affect the mouthfeel of wine are also topic of interest such as aging, type of the barrel that is used³⁷ and pH of the product itself³⁸. Acidity is one of the most important factors that affect the taste of wine and residual sugar affect the sweetness. Additionally, phenolic compounds are responsible for the color of wine³⁹. According to the studies by Rossouw and Marais, classification of wines from different cultivars could be achieved by using total amounts of polyphenolic compounds in wines⁴⁰.

Up to now, different kind of techniques were applied for wine analysis including HPLC⁴¹, FTIR⁴² and UV-Vis⁴³. Among the studies, Liu et al. studied on classification of wines with respect to their geographic origin by using visible and Near-IR spectroscopy techniques and PCA statistical method⁴⁴. Additionally, similar

work is performed by Teixiera Dos Santos et al. with 97 different wine samples by using vibrational spectroscopy⁴⁵. In another study by Lee et al. 45 different samples from Thai wines were classified with respect to their geographic origins by using FTIR⁴⁶.

Even though wine study with Terahertz spectroscopy is yet an unexplored area compared to other techniques that are used, still there have been a number of studies. Cork enclosures are studied with Terahertz spectroscopy by Yew Li Hor and Anthony J. Teti et al^{47,48}. In those studies THz images of cork samples were mapped. Since cork enclosures have large cells with low humidity inside, it provides high THz transmission. When the technique is compared to other imaging techniques, Terahertz imaging gave better results because of the matching THz wavelength and cell dimensions⁴⁷. In another study conducted by J. Teti et al, diffusion of water to the cork cells and cracks were mapped by using Terahertz imaging with the help of the sensitivity of Terahertz waves to water⁴⁸. Additionally, Terahertz spectra of the chemicals that affect taste, odor and color of wines were individually taken⁴⁹⁻⁵⁷. In this study, we applied Terahertz spectroscopy for wine analysis including the analyses of wine chemicals and ethanol measurements with different concentrations toward development of a method for a classification of wine samples from different cultivars by utilizing the differences in the absorption, refractive index and dielectric properties of the samples with the help of Principle Component Analysis (PCA). One another important aspect of this thesis is its possible contribution to the wine industry in Turkey. Turkey is one of the leading countries for grape production in the world⁵⁸; however, only 2.5-3% of the produced grapes are used for wine production and only 25% of produced wines are exported to other countries⁵⁹. As a result, Turkey exports less than 1% of the produced wines in the world despite having the world's fourth largest vineyards⁶⁰. Therefore, this study has a potential to be a pioneering work for the development of a technique for quality control of wines that can be used in industry also. This way, Turkish ones can be compared with well-known wines in the world and the study may help the establishment of Turkish wine quality.

CHAPTER 2

EXPERIMENTAL

In this study two main instruments have been utilized in order to collect the spectra of the wine samples and wine chemicals in the terahertz region of the electromagnetic spectrum. The following parts will detail the instruments used along with the techniques including the statistical techniques employed in order to classify the wine samples according to their qualities.

2.1 Components of THz-TDS Setup

In this study, a home-built Terahertz time domain spectrometer (THz-TDS) was used to collect the Terahertz spectra of the wine samples and its chemicals. **Figure 2** presents the basic diagram of the optical components of the spectrometer built by a group member, Yusuf Samet Aytekin as a part of his Master thesis study. The main components of the spectrometer are listed in **Table 1** with their model number and the name of the manufacturer companies.

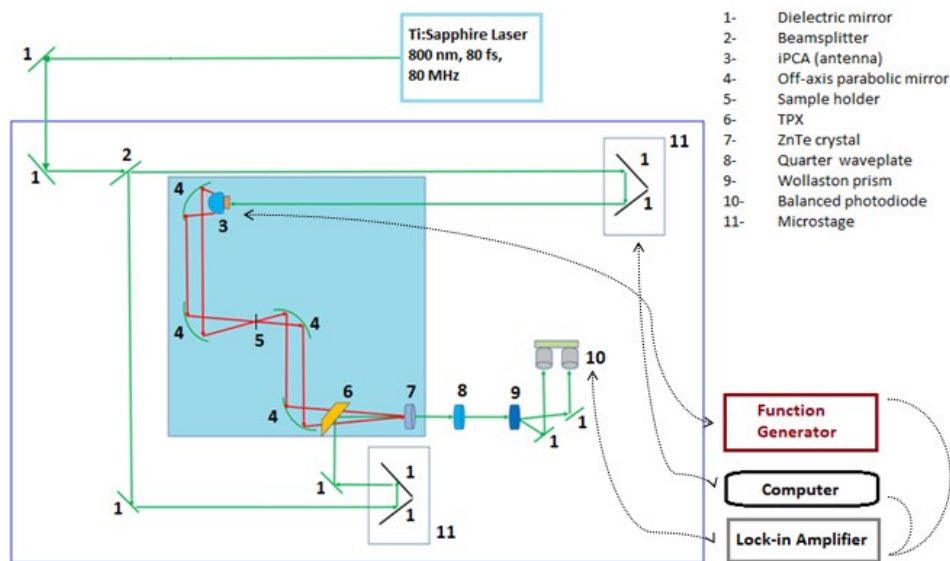


Figure 2. THz-TDS setup used in this study

Table 1. Items used in this system

Component	Manufacturer Company	Model
Ti:Sapphire Mode-Locked laser	Coherent	Mantis-5
iPCA antenna	BATOP	iPCA-21-05-1000-800-h
Dielectric mirrors	Thorlabs	BB1-E03
Beam splitter	Thorlabs	BSF10-B
Lenses	Thorlabs	LA1908-B and LA1172-B
Motorised microstage and its controller	Standa	8MT167-100 and 8SMC1-USBHBHF-B2-2MC
OAPMs	Thorlabs	
TPX plate		
ZnTe crystal	MTI corp	
Wollaston prism	Thorlabs	WP10
Quarter waveplate	Thorlabs	WPQ10M-808
Large area balanced photodiode	Thorlabs	PDB-201-A
Function generator	HM8030-6	HM8030-6
Voltage amplifier	BATOP	
Lock-in amplifier	Stanford Research Systems	45347A
Multifunction Data Acquisition	National Instruments	NI USB 6251 and USB X SERIES 6361
Oscilloscope	Tektronix	DPO2014

2.2 Working Principle of THz-TDS Setup

2.2.1 Generation of THz Waves

Two components in the setup play the main role in THz wave generation. Basically, light is produced via a Ti:Sapphire laser and iPCA antenna is used in order to generate THz pulses under an applied voltage upon excitation of the antennas with the laser light produced with the Ti-Sapphire laser.

2.2.1.1 Laser

A femtosecond Ti:Sapphire mode-locked laser is used in our system as a light source for both generation and detection of THz waves [**Figure 3**]. Those kind of lasers can produce light as in the form of pulses each have durations in femtosecond level. Before mode-locking, laser produces ca. 800 nm CW light having 230-400 mW output power depending on the current setting of the diode lasers inside. After mode-locking, approximately 480-550 mW average output power can be achieved for a pulsed laser light at 80 MHz repetition rate with ca. 800 nm center wavelength.

Those kind of ultrafast lasers are so sensitive that they can be easily affected by room climate changes such as temperature and humidity. In order to keep the temperature constant, an auto-set air-conditioning system is used in our lab through a HEPA filter to keep the working bench clean. Temperature is currently set to 20.5°C on the air-conditioning system. In addition, a chiller is used in order to keep the temperature inside the laser constant during the operations. Otherwise, laser would easily be heated up and stop operating. In our chiller, 5% ethanol solution is used as the circulating liquid and that solution is renewed with monthly periods. Temperature of this solution is set to 21°C.

Average temperature and humidity in the lab deviate during seasonal changes. Therefore, the laser system requires alignment of its optical beam path in the laser

cavity to compensate the beam path changes by these humidity and temperature changes for a minimum of 2-3 times a year.



Figure 3. Laser that is used in this study

2.2.1.2 iPCA Antenna

The generated laser light is directed to a beam splitter by using dielectric mirrors and the light is splitted into two arms with a ratio of 90:10. Then, 90% of generated light (by power) is directed to the iPCA antenna which can be seen in **Figure 4**.

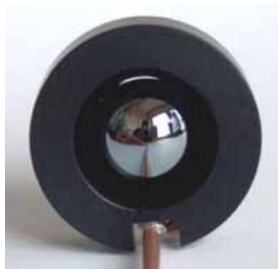


Figure 4. iPCA antenna

Antenna has gold (Au) electrodes on a GaAs chip with LT GaAs fast absorbing layer [Figure 5]. Electron-hole pairs are generated between valence and conduction bands when the photons of focused laser light drops on the gaps between electrodes. This is only possible if the energy of photons coming from laser is higher than the band gap energy of the semi-conductor. Then electron-hole pairs are accelerated with applied bias voltage and finally THz pulses are produced¹¹. In our iPCA antenna, applied bias voltage is 14 V and power of incoming laser light is approximately 480 mW.

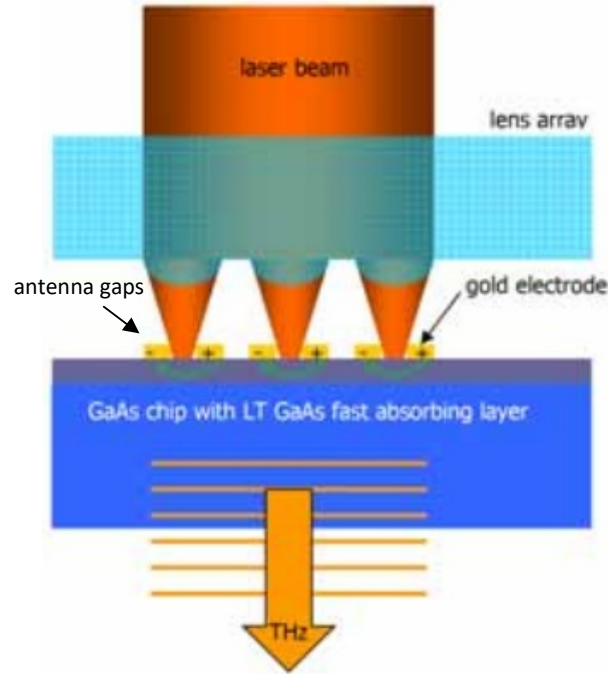


Figure 5. Structure of iPCA

2.2.2 Detection

THz wave detection is achieved via electro-optic sampling in our system. In this technique, a birefringent crystal, ZnTe by MTI corporation, is used in this setup. Birefringence is defined as the change of the refractive index in a material (typically crystalline material) between the different planes, where the polarization directions of the light beam travelling through⁶¹. After its generation in the antenna the THz

waves are propagated through the sample by using off-axis parabolic mirrors and directed to the ZnTe crystal. Unfortunately, detection of THz waves directly is not feasible thus ZnTe crystal is used in order to measure the THz field indirectly through electro-optic sampling. The 10% of the laser beam separated on the beam splitter is delivered to the ZnTe crystal through detection arm with the help of dielectric mirrors. As the THz wave changes the optical properties of the crystal while it is passing through, the polarization of detection beam also changes as it propagates through ZnTe at the same time and space. The linearly polarized laser beam becomes circularly polarized as it exits the crystal. A quarter wave plate on the setup is used to control the polarization of the detection laser light. The angle of the quarter wave plate is set such that the detection laser light exits the wave-plate when the THz beam is blocked, both polarization components (S and P) have equal power resulting a power ratio of 1. However, when the THz beam is on, this ratio deviates from 1 significantly and the deviation amount depends directly on the field intensity of the THz beam. Afterwards, detection beam arrives to Wollaston prism and it is used for separation of the two polarization components (s and p). Later it is delivered to two balanced photodiode where the intensity of the laser light is measured. Without THz beam, intensities of s and p polarizations of light will be the same since both polarizations will have the same power. Therefore, when the intensities of signal of two polarizations on the photodiodes are subtracted, a net signal of zero is measured. As the THz field interacts with the crystal, this balance is changed in one way or another thus a record of THz field in time is collected on a computer with help of signal lock-in amplifier and analog digital signal converter [Figure 6].

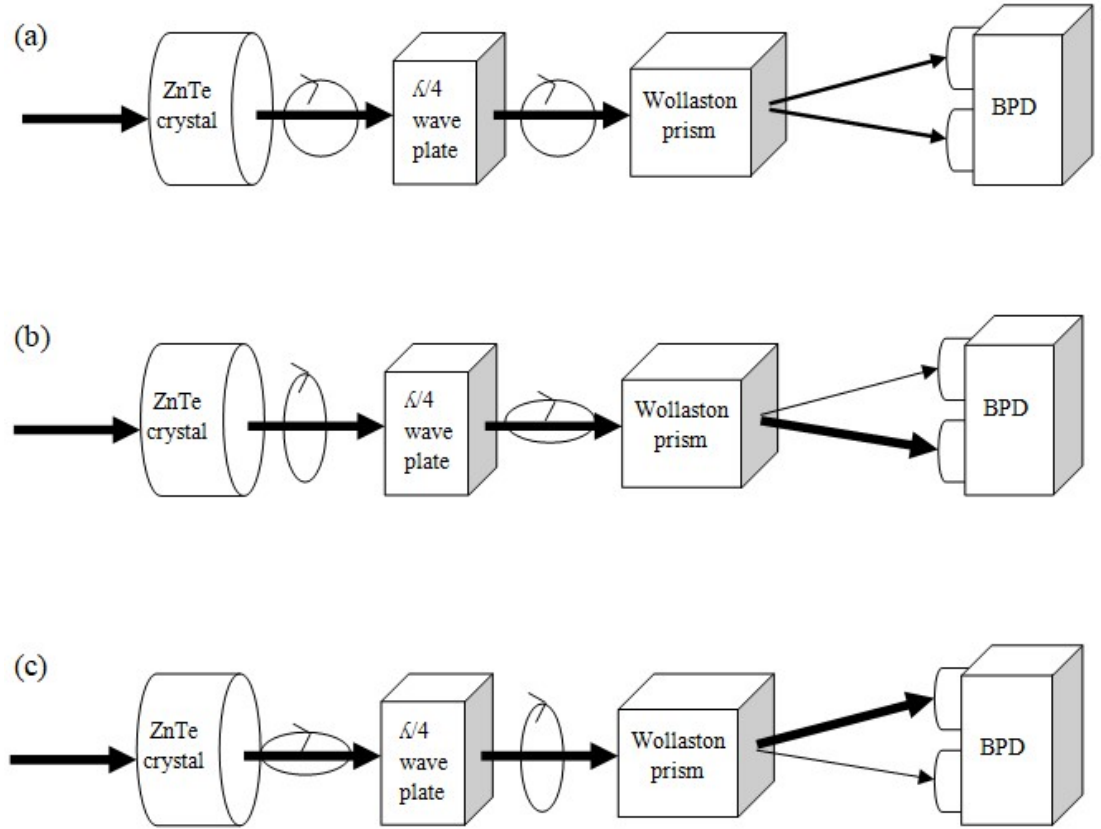


Figure 6. Scheme of electro-optic detection is shown here. (a) Intensities of s and p polarizations of detection laser pulse are equal to each other if there is no overlap with THz pulse in the ZnTe crystal; however, (b) if positive side of the THz beam is overlapped with detection laser pulse, then balanced photodiode is able to read the difference, also (c) it can read the difference when the negative side of THz beam is overlapped.

In order to get the THz signal profile in time, a delay system is integrated into the setup by using motorized stages. These motorized stages either in generation or in detection arm are used to delay THz beam in time relative to the detection beam with less than picosecond resolution in time. An example of a THz time profile collected is given in **Figure 7**.

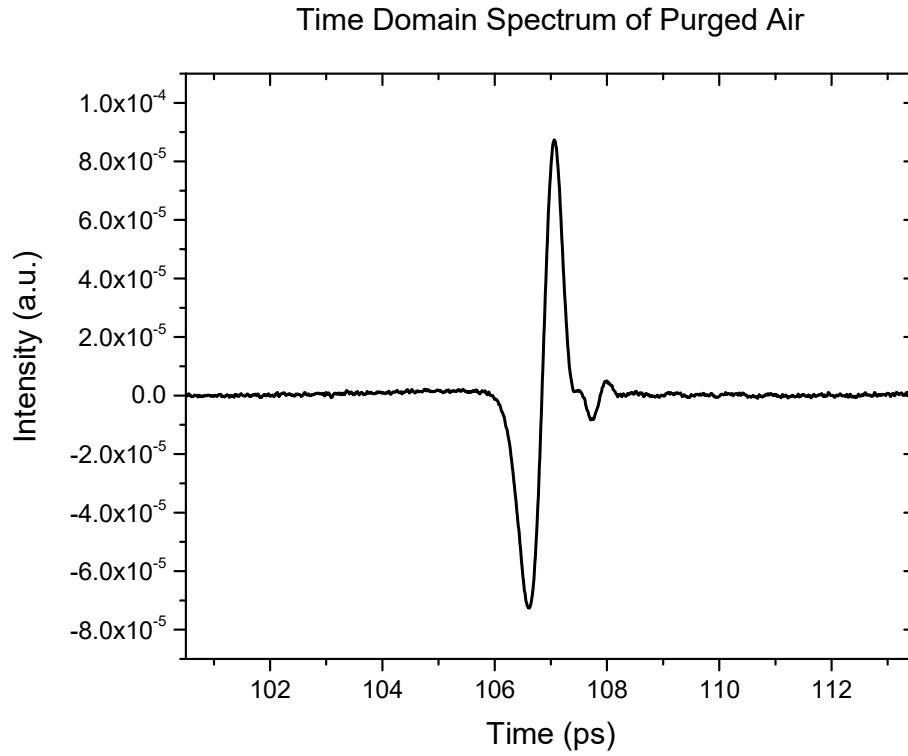


Figure 7. A typical THz waveform

After detection, the intensity of signal is increased with the help of lock-in amplifier. It also decreases the noise level; therefore, S/N ratio is highly enhanced with lock-in amplifier.

2.2.3 Data Collection

The electronic components used in our laboratory for data collection and signal enhancement are lock-in amplifier, function generator, balanced photodiode, a data acquisition card (DAQ) and a computer. As it is mentioned, there needs to be a delay with THz beam in order to get the whole THz wave profile in time. This is achieved with motorized micro-stages which are controlled with LabView software. On the other hand, function generator is connected to both lock-in amplifier and THz wave

generator (iPCA in our case) to apply bias voltage. The information is firstly taken from the balanced photodiode and sent to lock-in amplifier, then it is transferred to LabView software through data acquisition card. Finally, LabView collects all the information and displays the data. Here, the control on the motorized stage is one of the main points for high S/N ratio. In addition, the bandwidth of THz signal, parameters on lock-in amplifier, LabView and function generator settings are also other important points that need to be well controlled before each set of experiment for the best dynamic range. Initially, these are all optimized during the system set-up by our former group member, Yusuf Samet Aytekin⁶². However, before the start of each set of experiments, effects of change in laser settings and angles of ZnTe crystal, quarter wave plate and Wollaston prism were studied in order to get the optimum S/N ratio and bandwidth of THz signal. Those optimized parameters are shown in **Table2**.

Table2. Optimization parameters of the setup

LabView	
Scanning mode	Continuous
Number of points	1950
Scan length	13 ps
Lock-in Amplifier	
Signal input	DC coupled
Sensitivity	10 μ V
Reserve	Normal
Time constant (dB)	24 dB
Time constant	100 ms
Optical Power	
Generation arm	3 mW
Detection arm	500 mW
Function Generator	
Voltage	14 V
Frequency	8 kHz
Laser	
Wavelength (CW)	803 nm
Wavelength (Mode-locked)	820 nm
Bandwidth (CW)	8 nm
Bandwidth (Mode-locked)	110 nm
Pulse width (Mode-locked)	80 fs
Angles	
ZnTe crystal	287°
Quarter waveplate	100°
Wollaston prism	55°

2.2.4 Analysis of Data

Figure 8a presents the raw data collected as a Time Domain Spectrum (TDS). The time domain profile shows the change of the intensity of electric field in time. In order to get a frequency domain spectrum, Fast Fourier Transform (FFT) with Blackman windowing is applied.

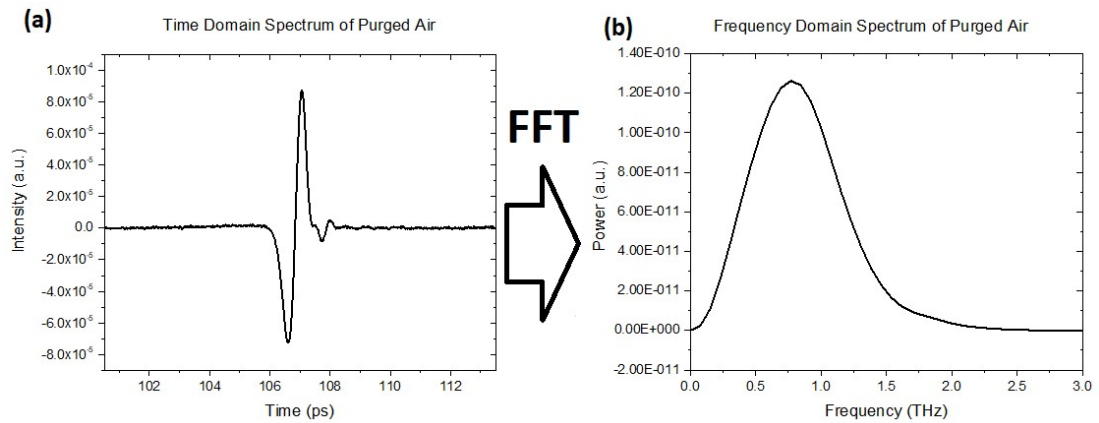


Figure 8. Typical time domain (a) and frequency domain (b) spectra

Frequency domain spectrum also demonstrates the bandwidth of the system for the related sample. **Figure 9** indicates that the effective bandwidth is between ca. 0.2 – 2.5 THz when the samples would be only air or empty sample cell itself (air and TPX); however, with the addition of sample, bandwidth gets narrower and it is ca. 0.2 – 1.85 THz.

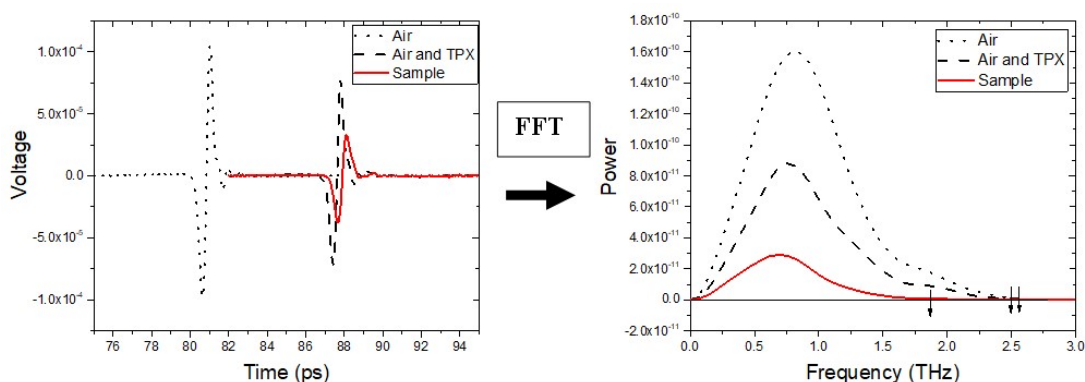


Figure 9. Sample dependency of bandwidth of the system

The frequency domain spectra are obtained by TDS Spectrum Viewer⁶³ and OriginPro 2016 Demo software products. At the end, OriginPro 2016 Demo was chosen for FFT and also further analyses of the spectra of our samples, because some unrealistic oscillating features were observed after data analyses with TDS Spectrum Viewer [Figure 10]. However, it is more compact and easy-to-use software compared to OriginPro and is used for quick investigation and comparison right after the measurements.

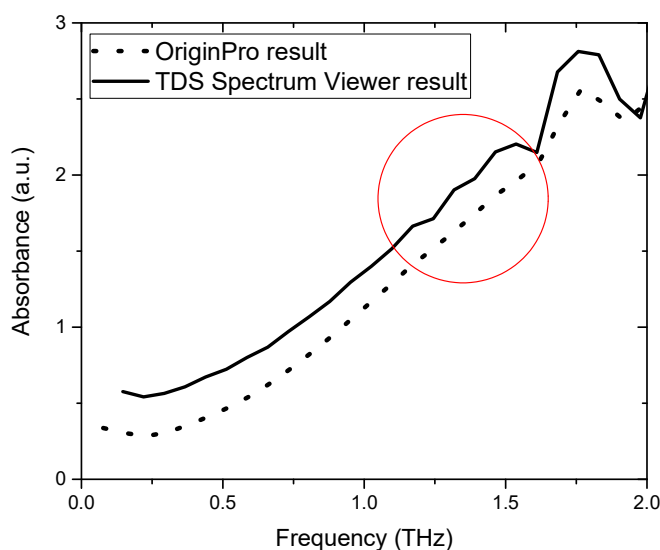


Figure 10. Absorbance spectra of pure Quercetin calculated by different software

FFT of the time domain data produces frequency dependent phase and amplitudes. These are later used to deduce further optical parameter of samples such as absorption coefficient and index by knowing those of the reference materials, which are shown in **Figure 11**. The frequency dependent refractive index of the sample can be calculated by using **Equation 1**, where c is the speed of light in vacuum, d is thickness of sample, ω is angular velocity, $(\phi_{sample} - \phi_{reference})$ is the phase difference between the sample and reference. Absorption coefficients of samples with respect to the frequencies can be calculated via **Equation 2**, where A_{sample} and A_{ref} are intensities of light belong to the sample and the reference respectively. Complex dielectric function can be calculated through **Equation 3** and the real ($\epsilon'(\omega)$) and imaginary parts ($\epsilon''(\omega)$) are dependent on refractive index (n) and extinction coefficient (k) as the **Equation 4 and Equation 5** indicates. Here, extinction coefficient can be calculated through **Equation 6**.

$$n(\omega) = n_{reference} + (\phi_{sample} - \phi_{reference}) \frac{c}{\omega d} \quad (1)$$

$$a(\omega) = -\frac{2}{d} \ln \left(\frac{A_{sample}}{A_{ref}} \right) \quad (2)$$

$$\epsilon(\omega) = \epsilon'(\omega) + i\epsilon''(\omega) \quad (3)$$

$$\epsilon'(\omega) = n^2(\omega) - k^2(\omega) \quad (4)$$

$$\epsilon''(\omega) = 2nk \quad (5)$$

$$k(\omega) = \frac{ca}{2\omega} \quad (6)$$

After calculation of absorbance, refractive indices and dielectric properties of wine samples, Principal Component Analysis (PCA) is applied to each set of data. PCA is a statistical technique that basically identifies the patterns in large sets of data with high number of dimensions and presents the data in a way that it highlights the similarities and/or differences within the sets of data by reducing the number of dimensions. In our study, PCA is used to extract information about correlation between the similar and different properties of wines using their frequency dependent absorbance, dielectric properties or refractive indices of the samples obtain from the set. Therefore, the aim was to be able to classify wine samples according to their properties such as grape type, alcohol content, quality etc. The PCA analyses have been applied on four different data sets [Table 3].

Table 3. Frequency ranges that are used for PCA

	Absorbance	Absorbance (narrow range)	Refractive Index	Dielectric Real Part
Range	0.23 – 1.92 THz	0.46 – 1.07 THz	0.30 – 2.15 THz	0.30 – 2.15 THz

Application of PCA to a set of data results set of scores with eigenvalues. An eigenvalue indicate variance of the data along the corresponding eigenvector. Higher the value means greater the variance; therefore, PC1 is always the eigenvector which indicates the greatest variance in a set of data. Different graphs plotted with principal components with respect to each other give different kinds of information about correlations among the samples. However, eigenvectors with small eigenvalues do not successfully explain the variance in the data. Therefore, only first 3 principal components were chosen for analysis in our study. Eigenvalues of all principal components in four different PCA are plotted in **Figure 12**.

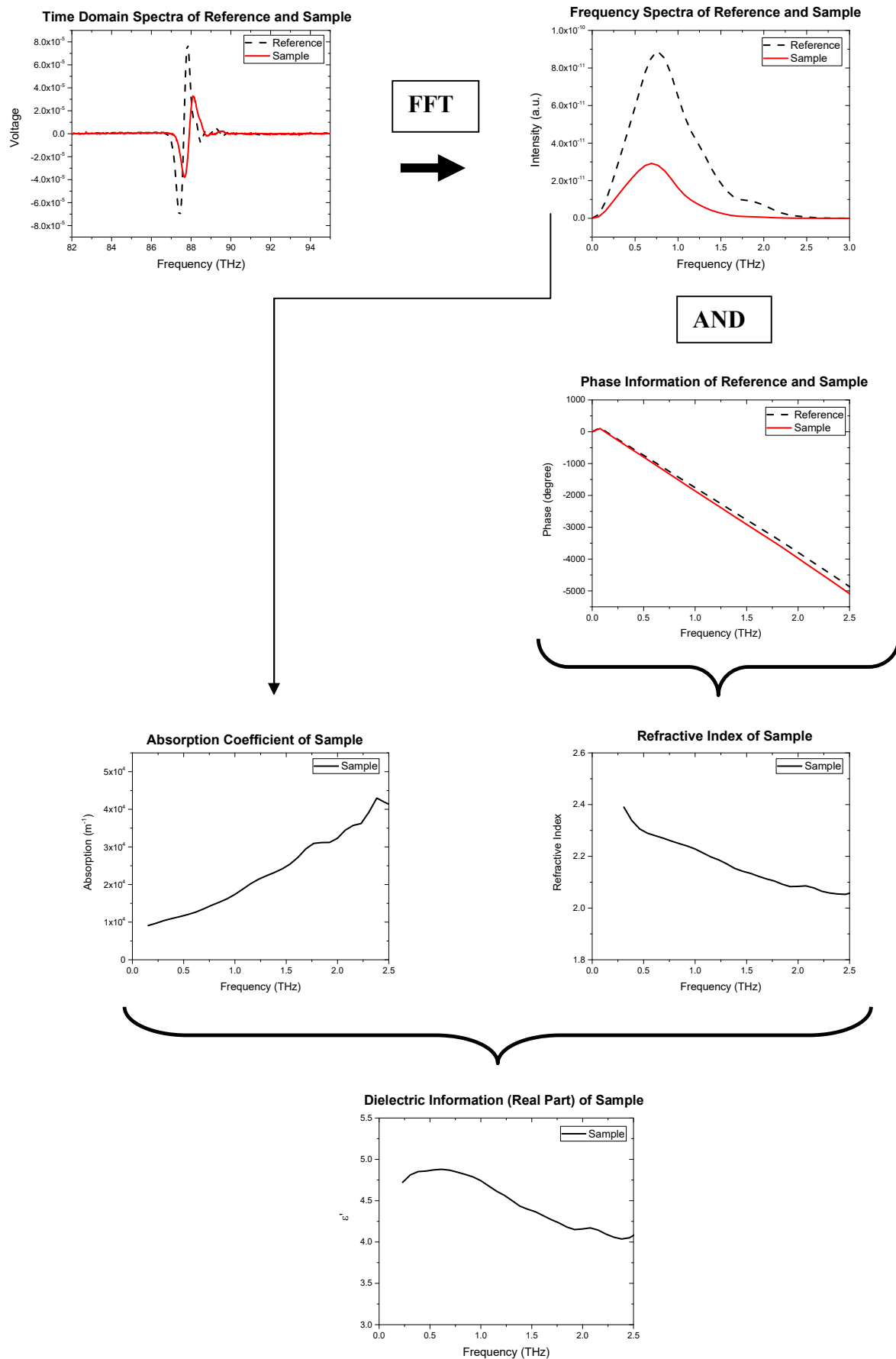


Figure 11. Representation of analysis of data

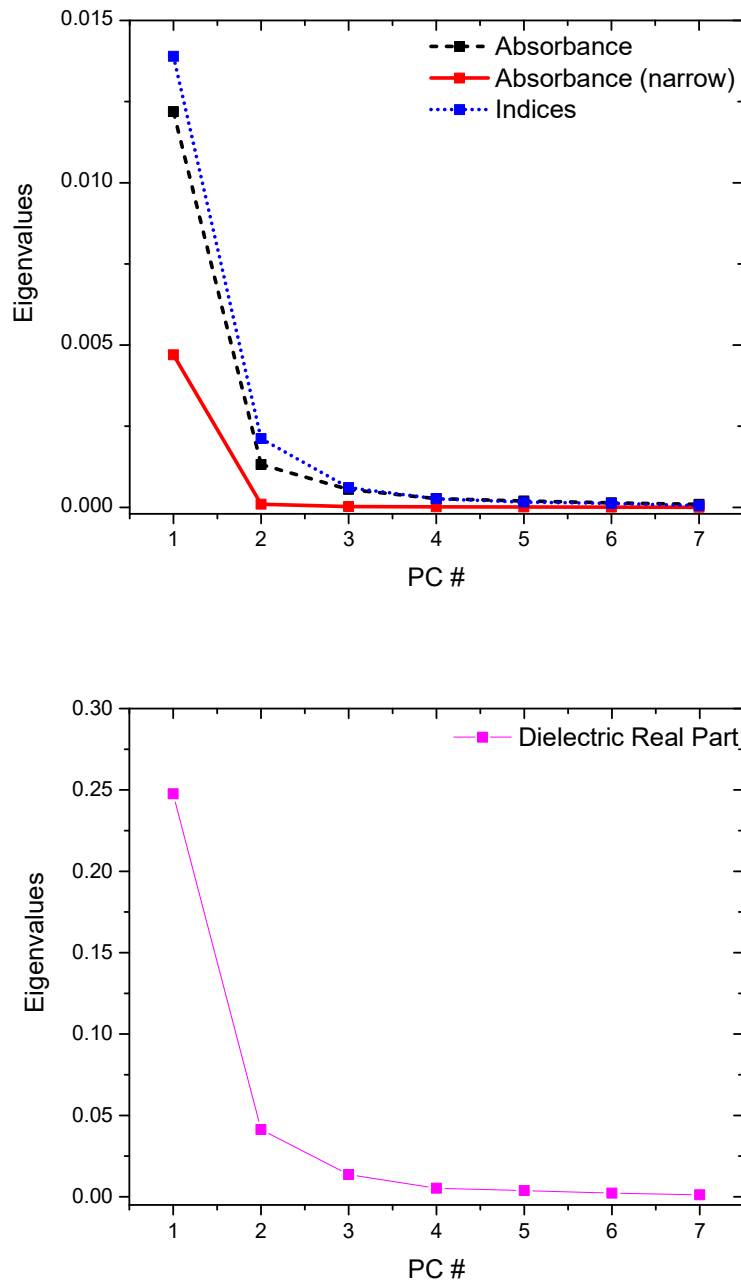


Figure 12. Eigenvalues of different principal components

2.3 Reproducibility of THz-TDS

Reproducibility of the system is measured by calculating the standard deviation of absorbance data within the analysis range that is used for wine samples. For all of

wine analyses, each of the samples had its own reference measurement taken before the sample measurement. Therefore, the same order is applied for reproducibility measurements but only 1 sample is used and measurement repeated for 3 times whose absorbance spectra is given in **Figure 13**. Also, 3 different wine sample measurements with absorbance data were selected for comparison (Sample 13-15).

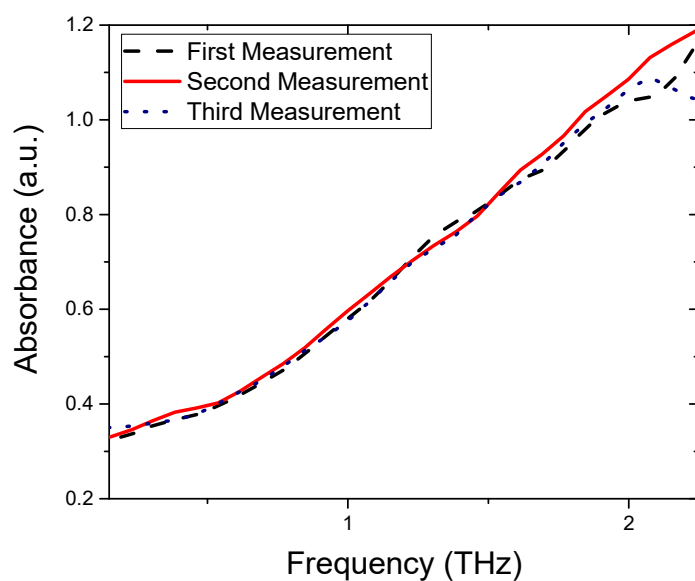


Figure 13. Reproducibility of the system

Standard deviation on each frequency of absorbance spectra which belong to 3 measurements of the same sample and 3 different samples between 0.23 – 1.92 THz were calculated. Then mean values are also calculated and results are shown in **Table 4** and **Table 5**. Finally, mean value of standard deviations of the same sample measurements are found 27% less than the mean value of standard deviations of 3 different samples that have the closest absorption profiles.

Table 4. Frequency dependent standard deviations for absorbance spectra of the same sample

Frequency (THz)	Absorbance (1st Meas.)	Absorbance (2nd Meas.)	Absorbance (3rd Meas.)	Standard Deviation
0.231	0.336	0.346	0.353	0.008
0.308	0.355	0.365	0.360	0.005
0.384	0.367	0.383	0.366	0.009
0.461	0.378	0.391	0.378	0.008
0.538	0.396	0.402	0.401	0.003
0.615	0.419	0.427	0.426	0.004
0.692	0.444	0.456	0.449	0.006
0.769	0.471	0.485	0.479	0.007
0.846	0.505	0.518	0.512	0.006
0.923	0.544	0.558	0.542	0.009
0.999	0.580	0.597	0.575	0.011
1.076	0.617	0.633	0.616	0.009
1.153	0.661	0.669	0.659	0.005
1.230	0.710	0.703	0.700	0.005
1.307	0.755	0.735	0.729	0.014
1.384	0.785	0.763	0.756	0.015
1.461	0.807	0.797	0.801	0.005
1.538	0.840	0.846	0.840	0.003
1.615	0.874	0.894	0.869	0.013
1.691	0.893	0.927	0.905	0.017
1.768	0.933	0.966	0.951	0.017
1.845	0.981	1.017	0.992	0.018
1.922	1.017	1.051	1.026	0.018
Mean				0.0095

Table 5. Frequency dependent standard deviations for absorbance spectra of three different samples

Frequency	Absorbance (Sample 13)	Absorbance (Sample 14)	Absorbance (Sample 15)	Standard Deviation
0.231	0.359	0.368	0.342	0.013
0.308	0.372	0.369	0.351	0.011
0.384	0.385	0.381	0.363	0.012
0.461	0.398	0.396	0.373	0.014
0.538	0.416	0.411	0.388	0.015
0.615	0.441	0.432	0.409	0.016
0.692	0.469	0.463	0.437	0.017
0.769	0.498	0.496	0.470	0.015
0.846	0.528	0.528	0.506	0.013
0.923	0.562	0.561	0.542	0.011
0.999	0.602	0.596	0.581	0.011
1.076	0.643	0.635	0.618	0.013
1.153	0.681	0.682	0.656	0.015
1.230	0.723	0.723	0.701	0.013
1.307	0.766	0.752	0.739	0.014
1.384	0.795	0.783	0.769	0.013
1.461	0.818	0.817	0.798	0.011
1.538	0.852	0.850	0.840	0.006
1.615	0.894	0.887	0.896	0.005
1.691	0.946	0.949	0.931	0.010
1.768	1.003	1.020	0.963	0.029
1.845	1.041	1.040	1.011	0.017
1.922	1.063	1.053	1.044	0.010
Mean				0.013

2.4 FT-THz Instrument

In this study, a modified FT-THz instrument (Thermo Scientific Nicolet 6700) was used for analysis at higher frequencies and as a check point for THz-TDS [Figure 14]. The instrument is modified with silicon (Si) coated Mylar® beamsplitter and a DTGS PE detector in order to reach frequencies below 400 cm^{-1} . With those modifications, the spectrometer is able to collect IR spectra of samples in Near-IR, Mid-IR and also Far-IR regions up to 50 cm^{-1} which partially overlap with the effective THz region available in our main setup.

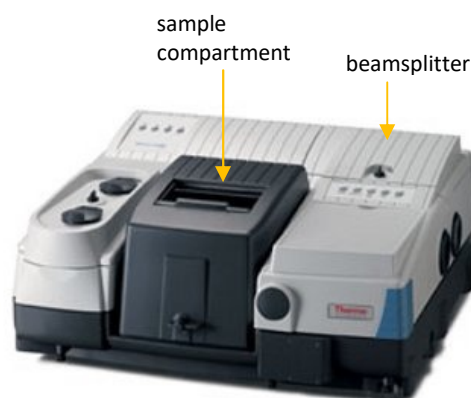
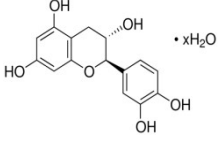
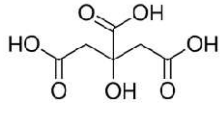
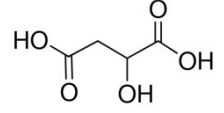
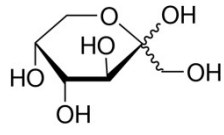
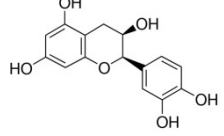
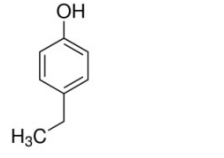
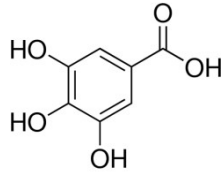
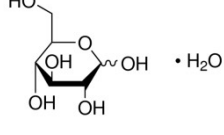
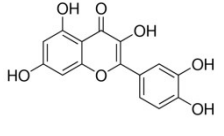


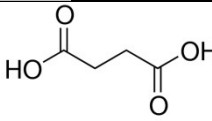
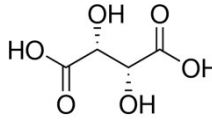
Figure 14. Thermo Nicolet 6700 FT-THz spectrometer

2.5 Sample Preparation

The samples were prepared and analyzed both in solid and solution forms in different solvent media. The analyte chemicals that are used in this thesis are listed in the Table 6 with IUPAC names, manufacturers, molecular formulas and structures.

Table 6. List of chemicals of interest, their manufacturer, formula and the structure

Name	IUPAC name	Manufacturer	Molecular formula	Molecular structure
(+) Catechin hydrate	(2R,3S)-2-(3,4-Dihydroxyphenyl) chromane-3,5,7-triol hydrate	Sigma-Aldrich	C ₁₅ H ₁₆ O ₇	
Citric acid	2-hydroxypropane-1,2,3-tricarboxylic acid	Merck	C ₆ H ₈ O ₇	
DL-Malic acid	DL-Hydroxybutane dioic acid	Sigma-Aldrich	C ₄ H ₆ O ₅	
D-(-)-Fructose	(3S,4R,5R)-1,3,4,5,6-pentahydroxy hexan-2-one	Sigma-Aldrich	C ₆ H ₁₂ O ₆	
(-)-Epicatechin	(2R,3R)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol	Sigma-Aldrich	C ₁₅ H ₁₄ O ₆	
4-Ethylphenol	4-ethylphenol	Sigma-Aldrich	C ₈ H ₁₀ O	
Gallic acid	3,4,5-trihydroxybenzoic acid	Sigma-Aldrich	C ₇ H ₆ O ₅	
D(+)-Glucose monohydrate	(2R,3S,4R,5R)-2,3,4,5,6-pentahydroxy hexanal;hydrate	Sigma-Aldrich	C ₆ H ₁₄ O ₇	
Quercetin	2-(3,4-dihydroxyphenyl)-3,5,7-trihydroxy chromen-4-one	Sigma-Aldrich	C ₁₅ H ₁₀ O ₇	

Succinic acid	butanedioic acid	Merck	C ₄ H ₆ O ₄	
Tannin		Merck		
Tartaric acid	2,3-dihydroxybutane dioic acid	Sigma-Aldrich	C ₄ H ₆ O ₆	

2.5.1 Solid Samples

Solid samples were prepared in pellet forms in order collect their spectra with both THz-TDS and FT-THz techniques. Chemicals were either taken from chemical warehouse inside METU-Chemistry Department or were purchased from Sigma-Aldrich. No further purification was needed nor applied. Both pure and diluted pellets were prepared for analyses. Dilution was achieved with polyethylene (PE) powder (Micro Powders Inc) that has an average particle size of 10 μm in diameter. For pure pellets, 100 mg from each analyte chemical was used; while for diluted pellets, ca. 95% dilution ratio was obtained with 95 mg PE and 5 mg analyte. Before the production of pellets, particle size of solid chemicals was tried to be reduced as much as possible in order to minimize the scattering of light. Hence, they were pulverized by using mortar and pestle. They were also used for obtaining homogeneous mixing for diluted pellets. Since the particle size of chemicals was quite reduced after that step, it was important to use a waxed paper to minimize the loss of analyte while transferring them to the die [Figure 15]. Before transferring them, it should be ensured that anvils and plunger were cleaned carefully with a volatile liquid like acetone or ethanol.



Figure 15. Pellet die

Pellets should have uniform thickness and the surfaces should be smooth as much as possible in order to reduce the reflection and scattering. Therefore, grounded solid should be manually dispersed on the lower anvil by using plunger. Then upper anvil should be gently placed on the analyte. In order to make fine pellets, press is used and pressure and wait time was optimized right after checking the pellets with naked eye. It was observed that for well-grounded chemicals, it is enough to apply lower pressure compared to grounded solids with large particle sizes. In this study, a fixed pressure and wait time were applied with 2 tons and 30 seconds respectively, except for tannin (pure), because the pressure was too high to get the pellet from the anvil without cracking it. Therefore, 0.5 tons of pressure was applied for tannin and pellet was easily released from the anvil.

2.5.2 Solutions and Wine Samples

The same chemicals were used in order to make solution measurements and they were prepared with various concentrations, especially during optimization studies of liquid sample cell, which is mentioned in detail in “Results and Discussion” part. Solvent was always either just the deionized water or ultra pure ethanol from Sigma-Aldrich or the mixtures of them with different Volume/Volume ratios.

Analyses of solutions were completed with the liquid sample cell [**Figure 19**]. Cell windows were TPX (polymethylpentene) plates in our study, because transmittance of TPX is higher than the other rigid polymeric materials like polycarbonate, polyethylene or teflon that can also be used as windows⁶⁴. The thickness of TPX plates was 2.2 mm.

In this study, 20, 30, 36, 40, 70, 100, 210, 250 and 500 μm spacer thicknesses were tried in order to find optimum thicknesses for FT-THz and THz-TDS measurements separately. It was predicted that thicker spacers would be useful for THz-TDS measurements, while thinner ones would be more suitable for FT-THz measurements because of difference between the powers of the light sources.

The sample thickness of wines was determined according to the previous experience obtained from wine chemical studies that are coming from solutions. **Figure 16** presents THz time domain spectra of empty cell and a solution with 15% ethanol in water. It is seen that approximately 50% (of intensity) transmitted through the solution with 70 μm thickness which indicates the thickness is appropriate for the analyses of wine samples, too.

Prior to measurement, no pre-treatment was applied on wine samples such as filtering or ethanol addition to increase its alcohol content etc. and the samples were kept with closed glass veils in refrigerator ($\sim 4^{\circ}\text{C}$) until analysis. During the measurements, each of the sample spectrum had been collected right after its own reference spectrum. The information about all wine samples measured are given in **Table 7**.

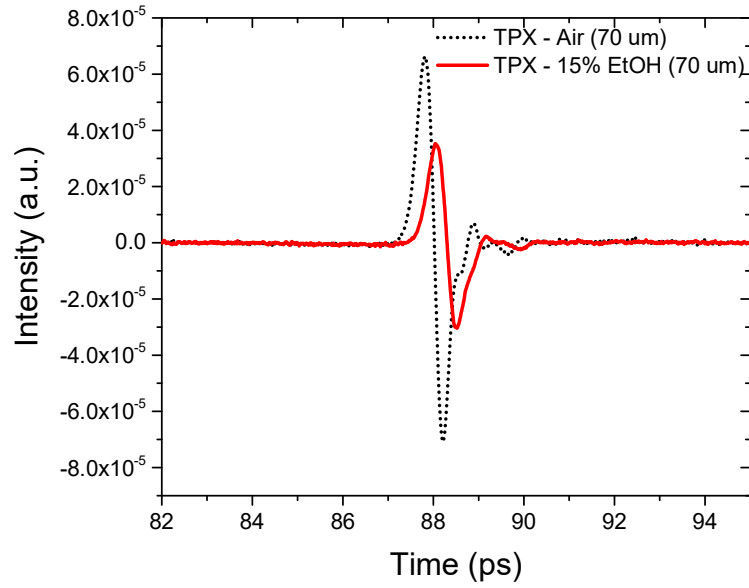


Figure 16. Signal loss due to 15% ethanol solution with 70 μm thickness

Table 7. Information of wine samples

Sample #	Brand	Sample is taken after the bottle is opened	Alcohol ratio (v/v) (as on label)	Production and bottling year	Cultivar
1	Buzbağ 2015 (red)	3 weeks	12.5%	2015 - *no info*	Öküzgözü / Boğazkere
2	Buzbağ 2015 (red)	4 days	12.5%	2015 - *no info*	Öküzgözü / Boğazkere
3	Angora 2016 (red)	A few minutes	13%	2016 – 2017	Cabernet Sauvignon / Alicante / Merlot / Öküzgözü
4	Buzbağ 2015 (red)	3-4 weeks	12.5%	2015 - *no info*	Öküzgözü / Boğazkere

5	Mistik (red)	1 hour	14%	2016 – 2018	Ege / Trakya region
6	Cadde (red)	A few minutes	12%	2015 – 2017	50% Öküzgözü / 30% Horozkarası / 10% Boğazkere / 10% Merlot
7	Turasan – Peribacası (red)	A few minutes	13.5%	2016 – 2017	Kalecik Karası / Boğazkere
8	Doluca (red)	A few minutes	14.5%	2016 – 2017	Kalecik Karası / Boğazkere / Shiraz
9	Angora 2017 (red)	30 minutes	13.8%	2016 – 2017	Cabernet Sauvignon / Alicante / Merlot / Öküzgözü
10	Cumartesi (red)	A few minutes	13.5%	2016 - *no info*	*no info*
11	*no info* (rose)	A few minutes	*no info*	*no info*	*no info*
12	Sandeman (white)	A few minutes	19.5%	*no info*	*no info*
13	Donini Collezione (red)	A few seconds	12%	2016 - *no info*	Merlot
14	Buzbağ – Diyarbakır (red)	A few seconds	12.5%	2015 - 2018	Boğazkere
15	Château Vartely (red)	A few seconds	13.5%	*no info*	Merlot
16	Terra – Varietal Series (red)	A few seconds	12.5%	2014 – 2017	Boğazkere
17	Akberg (red)	A few seconds	12.5%	*no info*	Merlot

18	Buzbağ – Elazığ (red)	A few seconds	13%	2015 - 2018	Öküzgözü
19	Turasan – Cappadocia (red)	A few seconds	14.5%	2017 – 2018	Öküzgözü
20	Château Vartely (red)	A few seconds	13.1%	*no info*	Cabernet Sauvignon
21	Akberg (red)	A few seconds	12.5%	*no info*	Cabernet Sauvignon
22	Akberg (red)	A few seconds	12.5%	*no info* - 2018	Boğazkere / Öküzgözü
23	Sade (red)	A few seconds	13.5%	2016 - 2017	Öküzgözü
24	DLC (red)	A few seconds	14%	2016 – 2017	Boğazkere

CHAPTER 3

RESULTS AND DISCUSSION

3.1 Minimization of External Effects

Strong absorption bands of water vapor at Far-IR region hinder collection of the spectra, thus detection of the samples. Therefore, it is important to eliminate or minimize the amount of water vapor from the sample compartment. Both in FT-THz and THz-TDS systems, purged air is used in order to achieve that goal. **Figure 17** presents the effect of water absorption on the raw spectra of air when the lid is kept open for 10 and 30 seconds. There are many strong and sharp bands even within this small range which can be determined by the increase in intensity of absorbed light by the water vapor. Similarly, **Figure 18** shows the purging effect on the intensity of the bands as the sampling region dries. There appears to be no significant difference after 15 minutes.

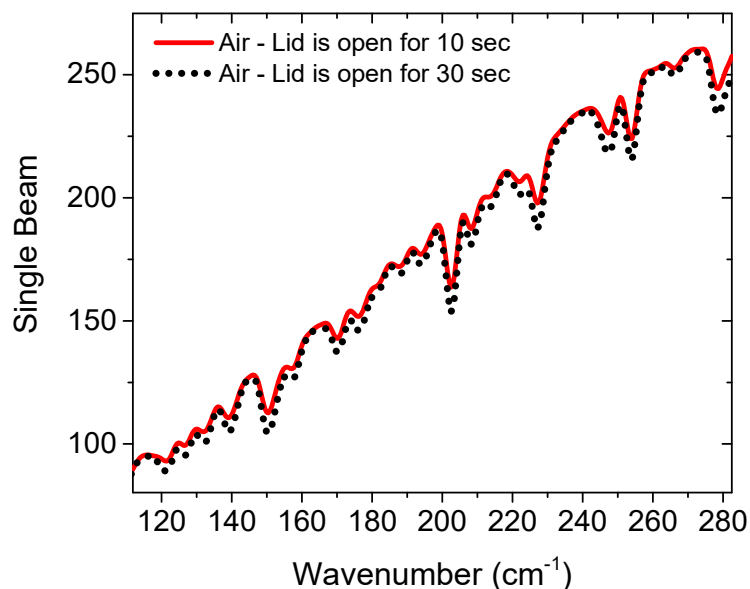


Figure 17. Effect of opening the lid on FT-THz spectra

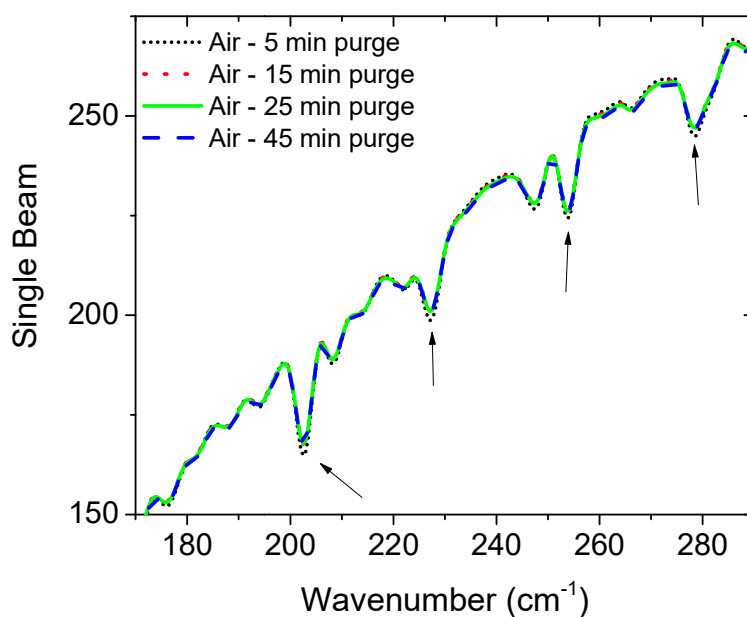


Figure 18. Effect of purge time on the FT-THz spectra

3.2 Optimization of Liquid Sample Cell

For both FT-THz and THz-TDS measurements, not only water vapor but also liquid water has significant interference effect on the spectra. Therefore, capability of the liquid sample cell was investigated by changing mainly 4 parameters: amount of water or ethanol, the path-length (thickness of spacer) and the concentration of the solutions.

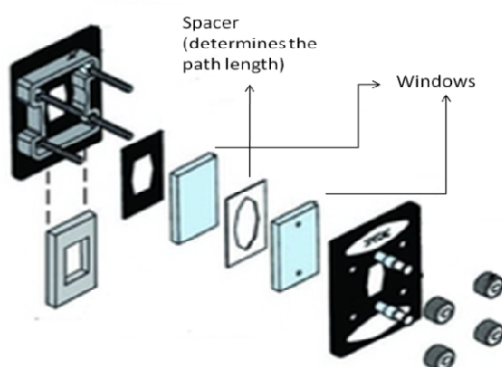


Figure 19. Diagram of liquid sample cell

Another important point is the material which sample cell windows are made of. It is known that some polymeric materials are more suitable for analyses in the THz region compared to others. One of them is Teflon [Figure 20]. Even though Teflon has more than 80% transmission in the THz region up to 5 THz, the spectrum is not flat and there is a structure in the spectrum which makes it less suitable for qualitative analyses. On the other hand, TPX and HDPE have almost no structure hence their transmission is fairly flat below 1.5 THz waves compared to Teflon [Figure 21]. Still, transmission depends on the thickness because of scattering effect. If thickness is too high, then degree of scattering increases and intensity of light at the detector decreases. So, transmission of a piece of CD box (polystyrene) and a TPX plate were compared by using FT-THz instrument [Figure 22]. The thickness of TPX was 2.2 mm and CD box was ~1 mm. It is seen that transmission of TPX plate is still higher than the polystyrene CD box.

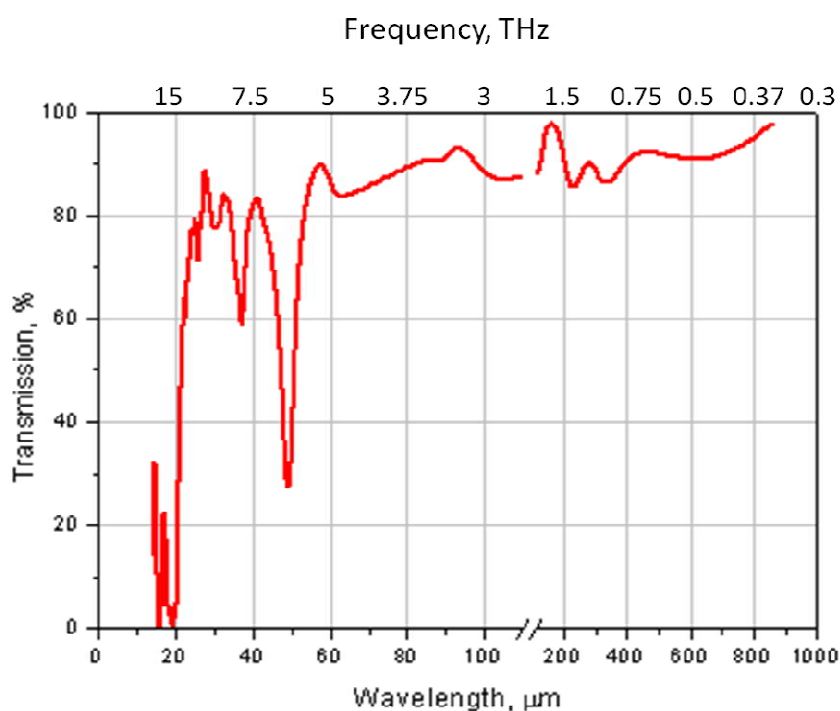


Figure 20. THz spectrum of Teflon⁶⁴

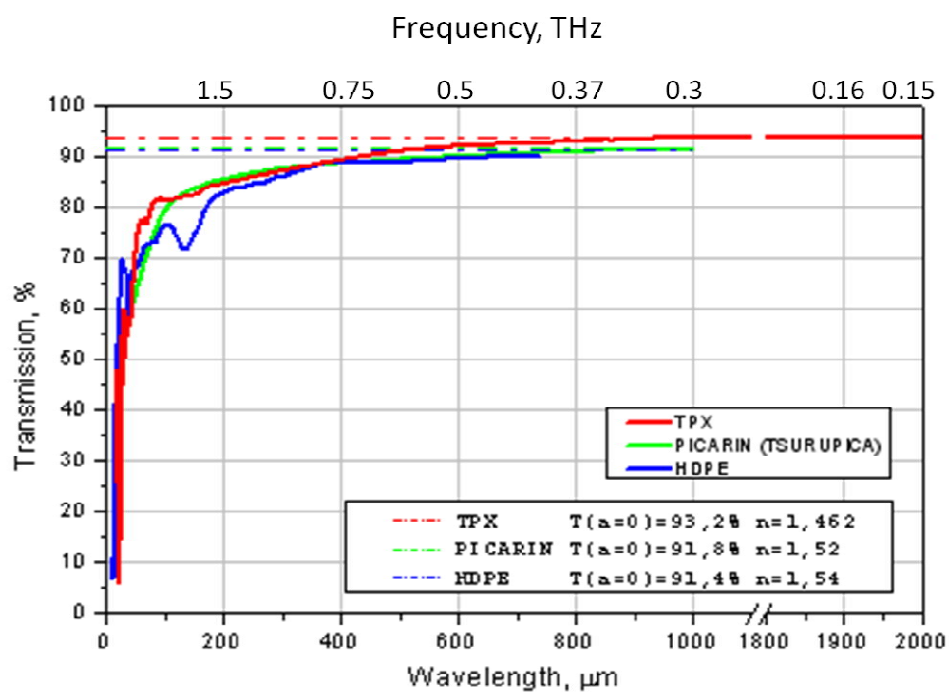


Figure 21. THz spectra of TPX, Picarin and HDPE⁶⁴

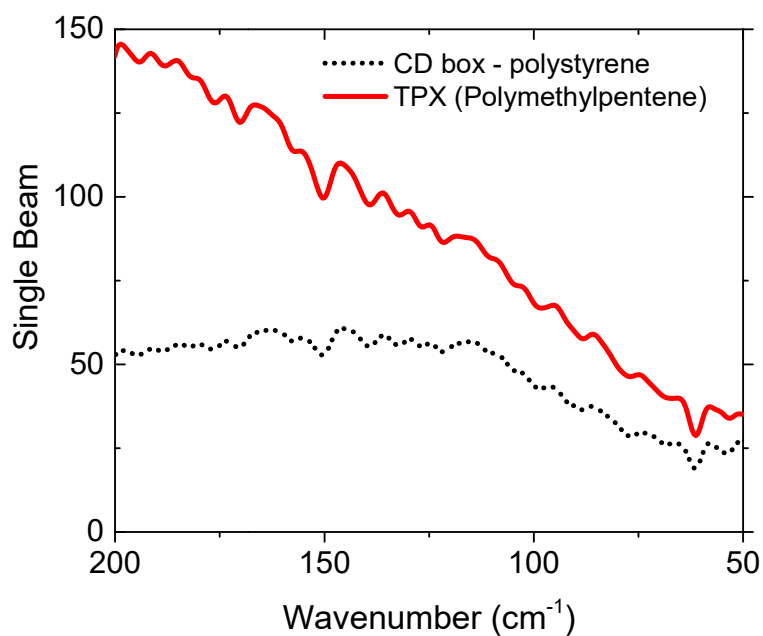


Figure 22. FT-THz spectra of two different types of sample cell windows

3.2.1 Studies for FT-THz Measurements

The measurement capabilities of FT-THz instrument and THz-TDS system are different from each other. Most important difference between two systems is the light source that is used. Intensity of light at the THz region created in FT-THz instrument is quite low compared to THz-TDS system. Therefore, it is important to determine the capability of sample cell and make optimization separately for each system. As mentioned earlier, there are four main parameters that affect the signal to noise level and the intensity of light arrives to the detector (sensitivity). Effects are summarized in **Table 8**.

Table 8. Effects of optimization parameters on measurements

	Absorption – Information about sample	Intensity of light - Sensitivity
If the amount of water <i>increases</i>	Decreases	Decreases
If the amount of ethanol <i>increases</i>	Increases	Increases
If the thickness <i>increases</i>	Increases	Decreases
If the concentration <i>increases</i>	Increases	Decreases

The effect of thickness on the transmission spectra is observed by using just water inside the sample cell [**Figure 23**]. Transmission decreases significantly above 36 μm and becomes almost zero when the thickness is 70 μm or above. This behavior is specific to water since it attenuates THz light significantly.

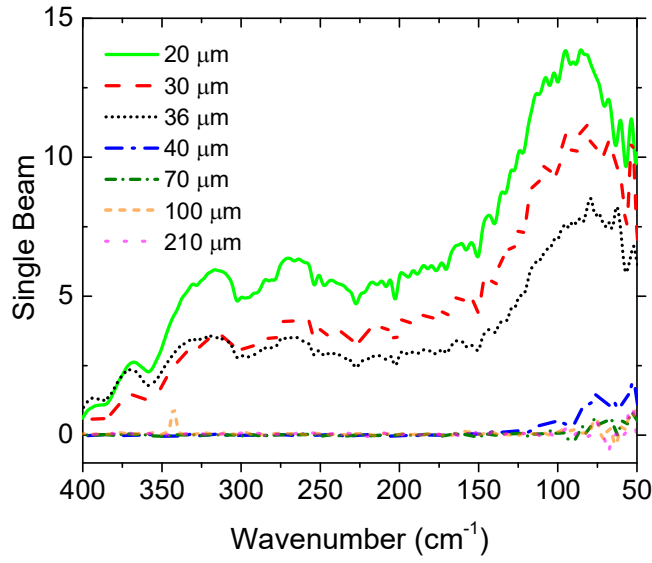


Figure 23. FT-THz spectra of water with different thicknesses

On the other hand, ethanol is more permeable than water in the THz region. Therefore, it is examined how the transmission changes with addition of ethanol to water since the wine samples do have alcohol content between 10-18 % (v/v) [Figure 24]. Even with the addition of 10% ethanol (v/v), transmission was significantly improved and working with 70 μm sample thickness became attainable.

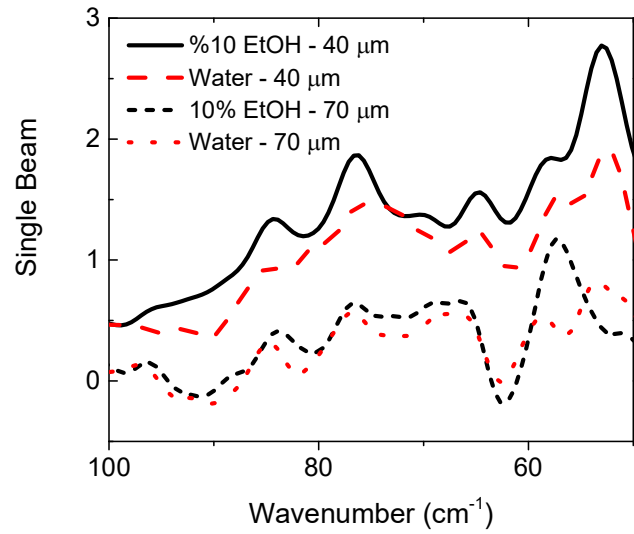


Figure 24. FT-THz spectra of water-ethanol mixture with different thicknesses

The capability of FT-THz system for determination the differences in ethanol concentration is examined by changing both the thickness and concentration [Figure 25 and Figure 26]. Figure 26 presents that it is possible to easily observe differences in the spectra for 1%, 3%, 5%, 10%, 20% and 30% ethanol in 30 μm thick sample cell. Not only 10% difference but even 2% difference between concentrations can easily be observed with FT-THz system. On another study, possibility of discrimination of alcohol samples with 1% differences was examined. It has been observed that 1% of ethanol change can be determined with 23 μm and 40 μm spacers [Figure 27 and Figure 28].

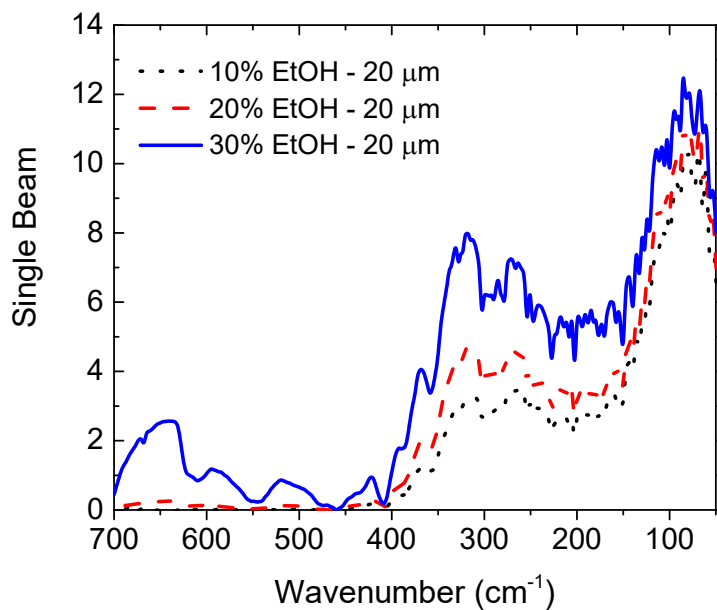


Figure 25. FT-THz spectra of water-ethanol solutions having 10% difference in ethanol concentrations (20 μm sample thickness)

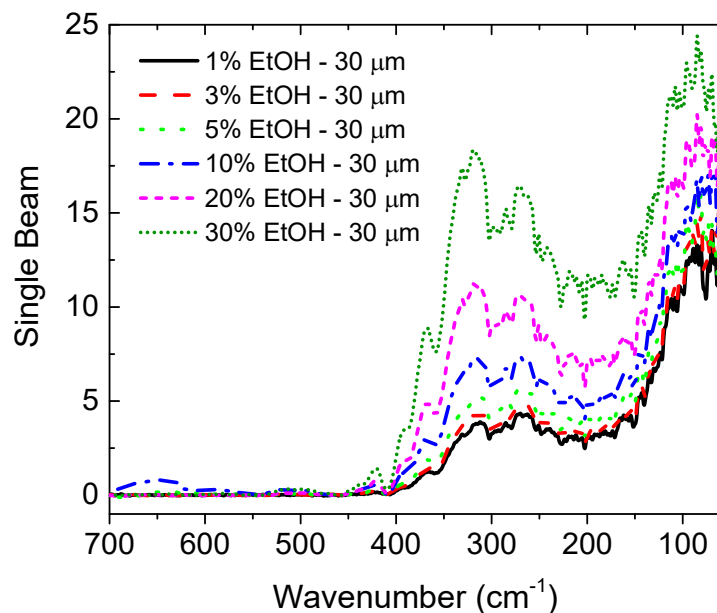


Figure 26. FT-THz spectra of water-ethanol solutions having varying EtOH concentrations (30 μm sample thickness)

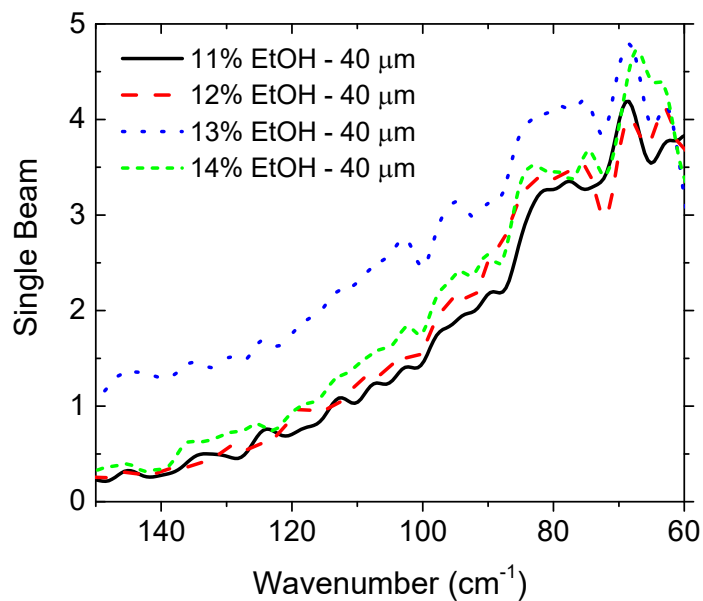


Figure 27. FT-THz spectra of water-ethanol solutions having 1% difference in ethanol concentrations (40 μm sample thickness)

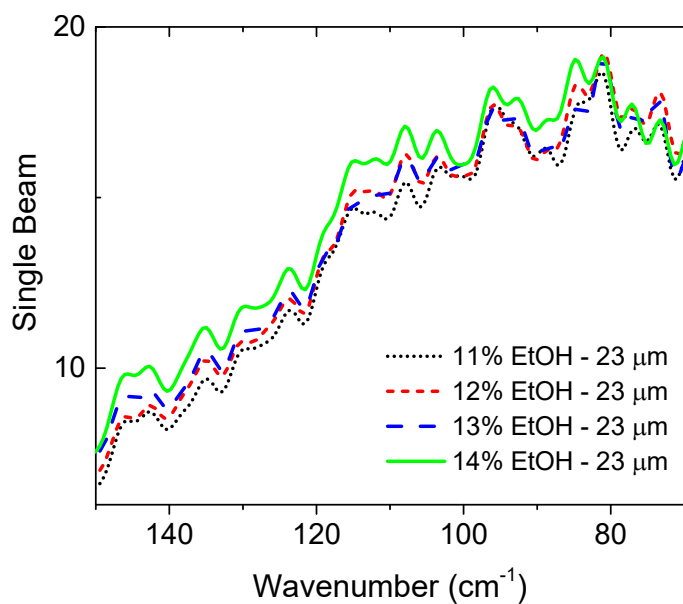


Figure 28. FT-THz spectra of water-ethanol solutions having 1% difference with ethanol concentrations (23 μm sample thickness)

Unfortunately, it is not easy to find such thin spacers. The 23 μm spacer was cut from a polymer film. It appears to be the best spacer to work with during FT-THz measurements. When the sample holder is prepared with 23 μm spacer, it is very difficult to fill the cell with a solution since the pressure inside this cell becomes too high. In addition, it is even more difficult to circulate different solutions. Even though 23 μm thickness gave the best differences, it was decided to keep this option as a secondary choice for FT-THz measurements. Increasing the spacer thickness and working with 40 μm was a better option in order to have higher sensitivity and more absorption information for the measurements of solutions with wine chemicals.

3.2.2 Studies for THz-TDS Measurements

Similar to the FT-THz studies, the sample thickness and the possible working concentrations were investigated for THz-TDS system, too. Initial studies had shown that it is easy to distinguish the ethanol/water mixtures with 5 % differences, especially up to 1.5 THz. **Figure 29** presents 10, 15, and 20% ethanol water solutions THz absorption spectra collected with THz-TDS system. The spectra were collected reference to water hence negative absorption means higher transmission relative to water, as liquid water has strong and very broad absorption in this region.

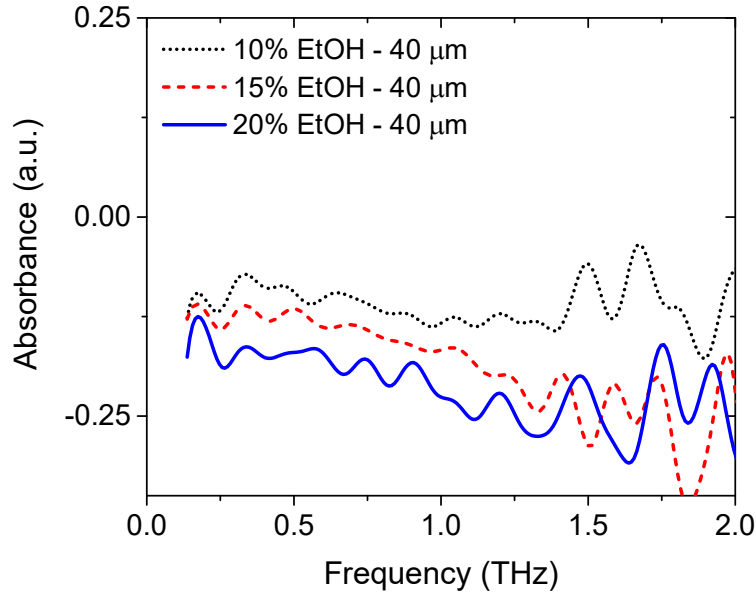


Figure 29. THz-TDS absorbance spectra of water-ethanol solutions having 5% difference with ethanol concentrations (40 μm sample thickness)

Time domain waveforms of solutions with 13%, 14% and 15% ethanol concentrations were prepared and collected [Figure 30]. However, after the application of FFT to the time-domain data, it was realized that the frequency domain data shows a very low frequency absorption band between 0-0.15 THz (shown with an arrow) that does not supposed to exist in these solution. After further investigation of the data it was realized that there is an artificial offset on the time-domain data (shown with an arrow on Figure 30), that is introduced by the lock-in amplifier. The off-set is very low but significant enough to introduce an artificial absorption band in the spectra. . To correct this behavior, a portion of data from a very early time (the first 30 data) where no signal is expected is averaged and was added to time-domain spectrum itself to level the base line to zero [Figure 31]. Therefore, offset was corrected and fake negative absorption feature between 0-0.15 THz was removed [Figure 32].

Figure 33 presents frequency domain of a single scan TDS data with no averaging. Even at this low signal it was possible to observe 1% ethanol concentration difference of solutions. With increasing the number of averaging and thus, SNR, system will be able to resolve the spectra better especially between 0.25 and 1.5 THz where we have enough bandwidth and good SNR.

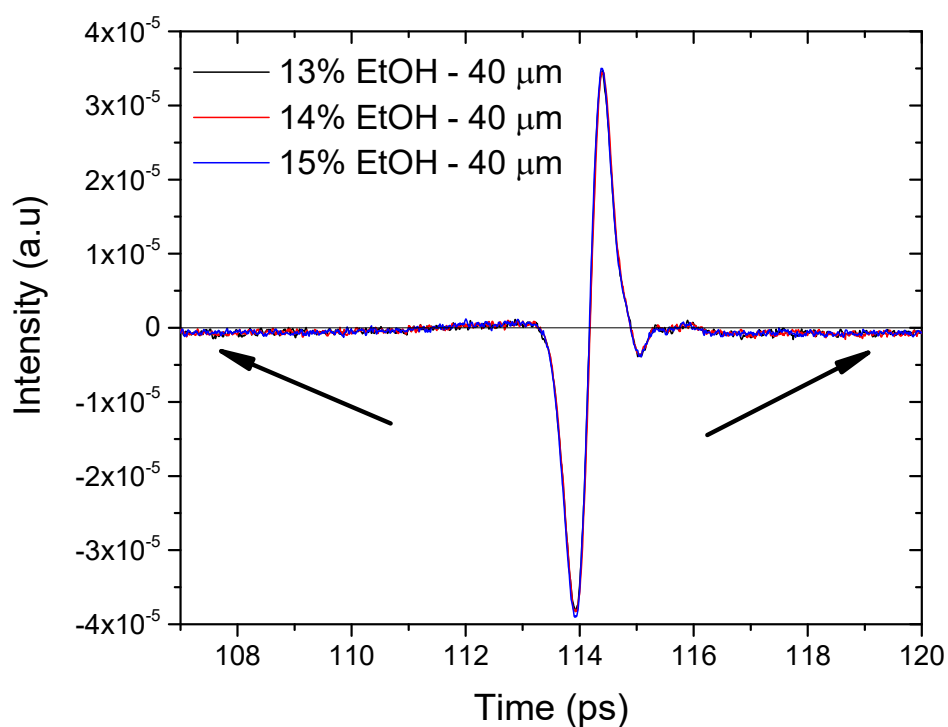


Figure 30. THz time domain spectrum of ethanol solutions with uncorrected offset line

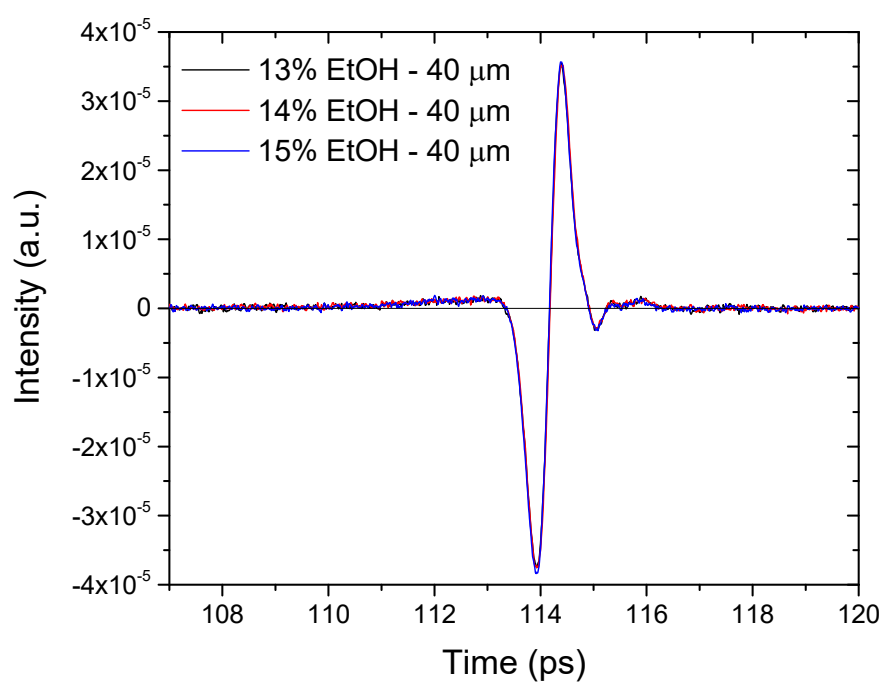


Figure 31. THz time domain spectrum of ethanol solutions with corrected offset line

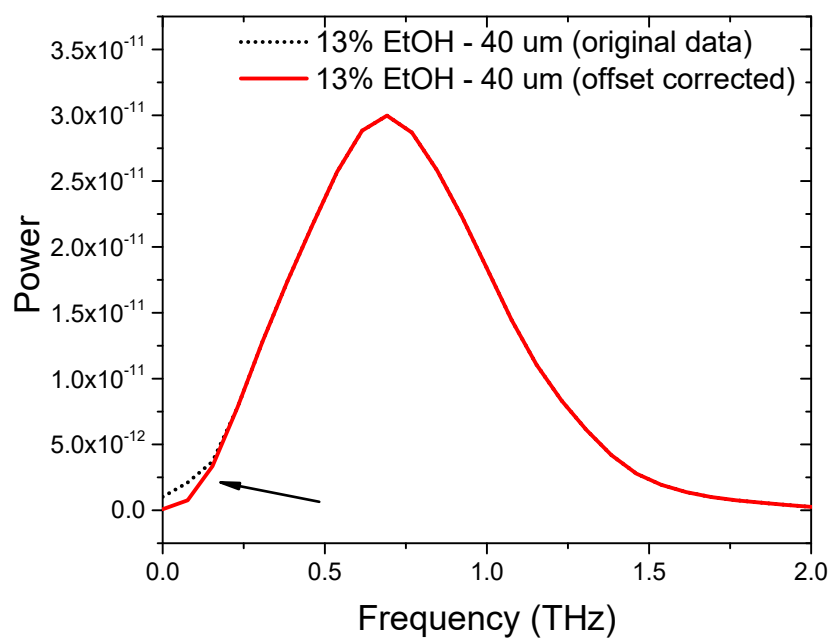


Figure 32. Effect of offset correction on frequency domain spectrum

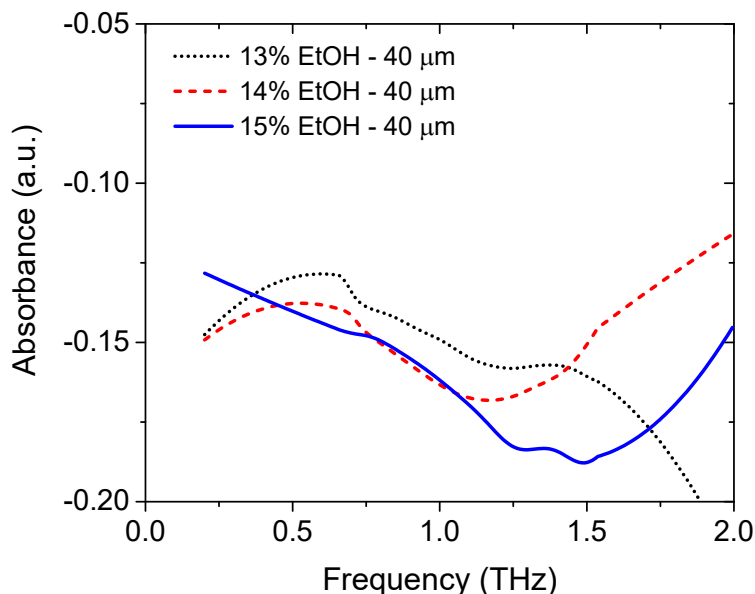


Figure 33. THz-TDS absorbance spectra of water-ethanol solutions having 1% difference with ethanol concentrations (40 μm thickness)

3.3 Terahertz Measurements

Analyses of both solutions and solid samples were completed with THz-TDS system. Firstly, spectra of solutions were collected and then solid samples spectra were made in order to observe the vibrational or rotational features of wine chemicals if exist in order to be used as a chemical marker during wine analysis.

3.3.1 Solutions

At the beginning, concentration of solutions was chosen 0.4 g/10 mL in ethanol-water solution. The medium of solutions was chosen as 15% ethanol in order to simulate the wine medium. This concentration is higher than the ones as they appear in a typical wine⁶⁵. However, no strong marker absorption bands were observed for almost all chemicals measured [Figure 34]. Although the spectra appear as flat and featureless, a closer look suggests some broad features that may be considered as a

band, especially in the spectra of acids. However, due to its low signal-to-noise ratio it was really hard to label those as vibrational features of the wine chemicals and use them as markers during wine analysis. There was a need for the confirmation either from solid sample measurements or with theoretical calculations.

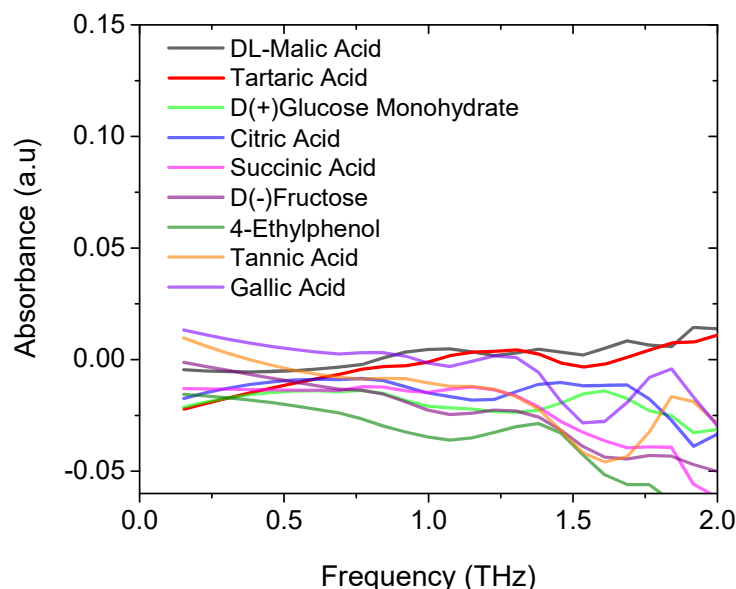


Figure 34. THz-TDS absorbance spectra of wine chemicals in solution form

3.3.2 Solid Samples

Due to difficulty in characterization of the wine chemicals in solutions, the solid samples of the pure and polyethylene mixtures as pellets were prepared for further analyses. **Figure 35** and **Figure 36** present the selected wine chemicals that have significant absorption features. These are DL Malic Acid, Gallic Acid, D(-)Fructose, D(+)-Glucose Monohydrate, Tartaric Acid and (-) Epicatechin. On the other hand, the others (Citric Acid, (+) Catechin Hydrate, 4-Ethylphenol, Quercetin, Succinic Acid and Tannic Acid) have featureless absorption spectra. Unless confirmed the bands beyond 1.5 THz is generally considered as artificial due to very low signal-to-noise level beyond that frequency.

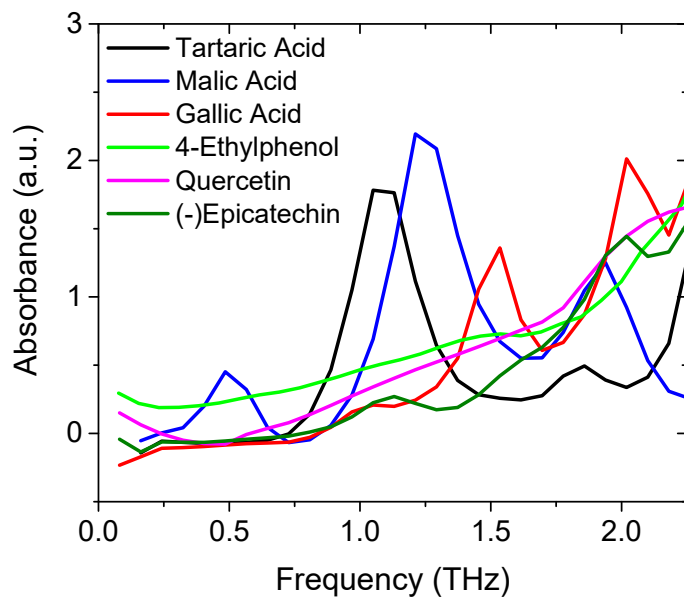


Figure 35. THz-TDS spectra of some of acids and phenolics in pellet form

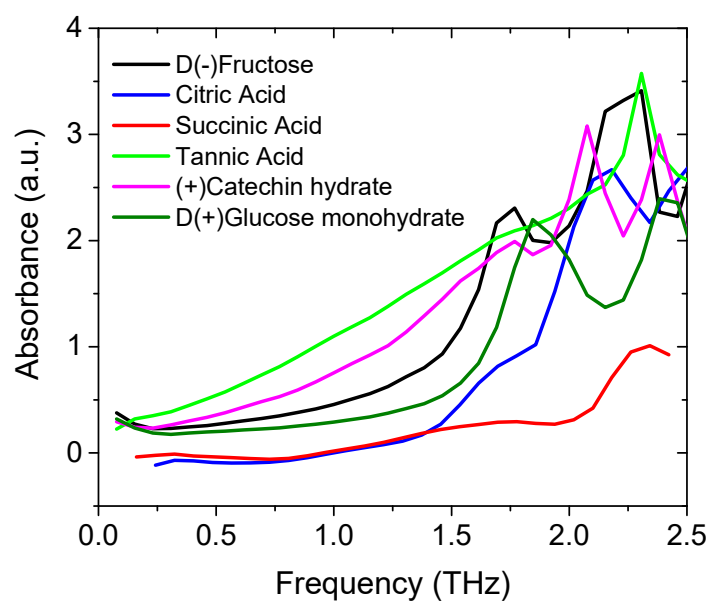


Figure 36. THz-TDS spectra of some of acids and sugars in pellet form

3.4 Malic Acid Measurements

After collection of the spectra belong to the solid form of the wine chemicals, a set of measurement was designed with malic acid, because it has intense absorption features at different frequencies.

Figure 37 presents FT-THz spectra of 5% solid solution of malic acid in PE and liquids samples with three different concentrations, 1.0 g, 12.5 g and 17.5 g of malic acid in 25 ml of 80% ethanol solution. In these measurements a 70 μm spacer and higher ethanol concentration were chosen in order to increase the THz transmission through sample significantly since the THz transmission through the sample is very low and hinders the measurements.

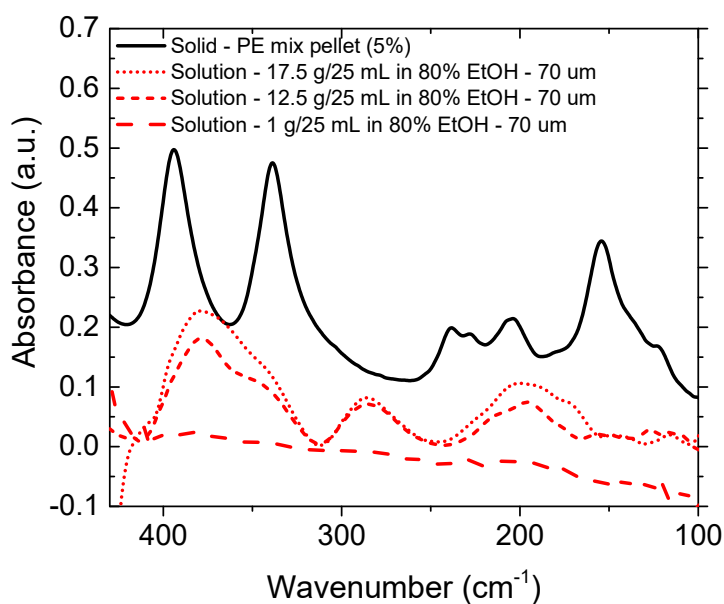


Figure 37. FT-THz spectra of DL-Malic Acid in both pellet and solution forms

The vibrational features observed in solid sample are quite strong, thus showing a fingerprint for malic acid in this region. The main strong bands associated to various vibrational motions of malic acid are located at 394, 338, and 151 cm^{-1} wavenumbers in addition to the weaker ones at 200, 225, and 235 cm^{-1} . However, when it is dissolved in 80% ethanol solution all these features broaden out and red shifted in

frequencies. In addition, the band observed at 151 cm^{-1} disappears. When a band exists in solid phase but disappears in liquid phase it is usually a sign for being a phonon mode rather than an intra-molecular vibrational mode associated with the motions of the molecules. In order to prove that this is a phonon mode, one needs to carry out solid-state calculations. Unfortunately, we do not have such a capability in our group.

Figure 37 presents spectra of solutions with three different malic acid concentrations. Unfortunately, FT-THz is not sensitive enough at 70 μm thickness and no bands have been observed and the spectrum is fairly flat. On the other hand, 12.5g and 17.5g ones have broad and well separated bands at 378, 285 and 200 cm^{-1} .

Figure 38 and **Figure 39** present spectra of malic acid solutions with different concentrations and different spacer thicknesses. No absorption feature is observed including the ones that are present in solid spectra collected or the ones reported in the literature⁶⁶. Spectra of saturated solutions with 250 and 500 μm spacers show a strong rise at high frequencies. This feature might be the tail of a strong absorption band located at frequencies greater than 3THz, such as the one observed in 151 cm^{-1} (ca. 5 THz) of solid sample with FT-THz. The reason is because in a saturated solution there still could be associated solid-like samples due to strong hydrogen bonding which results a similar feature.

The solution spectra at low to moderate concentrations, however, have a very broad and weak band between 1 and 1.5 THz. The observed feature is too broad and too weak to be used as a marker band for identification and quantification of malic acid in wines. The observed negative absorption profile of the high concentration malic acid samples (5g and saturated solutions) is mostly because of solvent replacement. The water and ethanol–OH bands in the THz region have high absorption thus signal attenuation. When these materials are replaced with a material which has lower number of OH groups in high concentration, the sample becomes more transmissive compared to the reference solution. This then results in negative baseline in the absorption spectra.

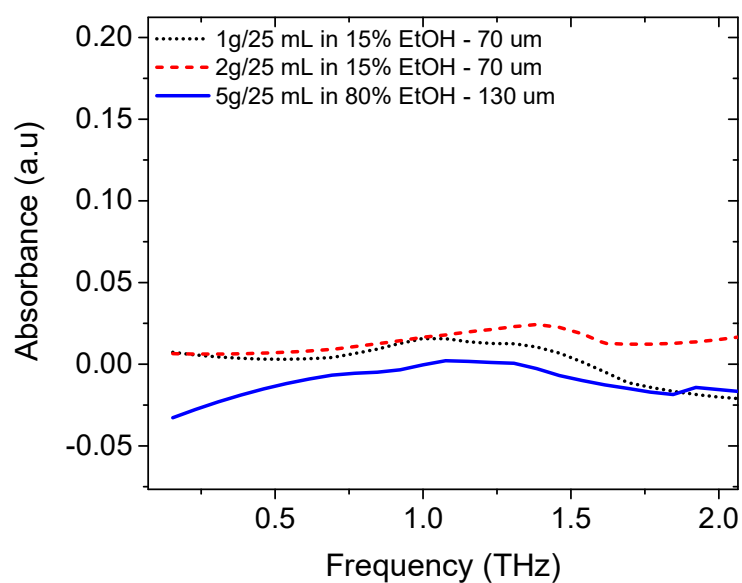


Figure 38.THz-TDS absorbance spectra of DL-Malic Acid solutions at different concentrations

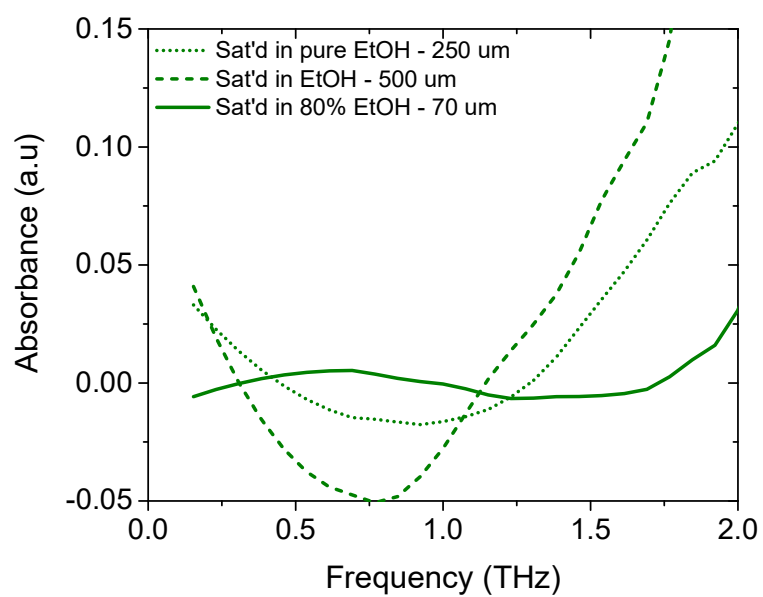


Figure 39.THz-TDS absorbance spectra of saturated DL-Malic Acid solution

Similar to the high frequency region of the malic acid spectrum collected with FT-THz, it appears that the bands observed in solid sample broadens and shifts to lower frequencies. Also, when dissolved in solution the weaker band observed in solid malic acid at ca. 0.5THz disappears. This is most likely due to being an intra-molecular mode such as phonon mode, in nature. Another explanation could be that if an absorption band broadens too much, then it might not be possible to observe the feature.

3.5 Analyses of All Wine Chemicals

After malic acid measurements, the sample cell was adjusted with 250 μm thickness spacer and the rest of the solution spectra were collected from saturated ones in order to see the absorption signals clearly. The other concentrations did result fairly flat and featureless spectra.

3.5.1 (+)Catechin Hydrate

(+) Catechin is one of the compounds found in wine belong to Flavan-3-ols group⁶⁷. It is found in fruits like grapes, berries, apples and also in various kind of tea. Amount of (+) Catechin in wine varies between 14.24 mg/L and 78.67 mg/L for 6 different commercial Sicillian red wines from Merlot cultivar⁶⁸. Hydrate form of the compound was used in order to collect the spectra in both FT-THz and THz-TDS systems.

Figure 40 presents spectra of the pure and PE mixture (solid solution) pellets and saturated solution in ethanol collected with FT-THz and THz-TDs spectrometers in region of 0.2-2 THz. The pure and PE mixture spectra have a broad absorption feature with a peak point at ca. 1.77 THz. FT-THz spectra of both sample forms also have similar absorption signals but they do not seem as broad as the features in THz spectra and they located at 1.72 THz frequency. This shows that there is shoulder band located approximately at 1.5 THz, where FT-THz does not have enough signal.

On the other hand, the band is not observed in spectrum of saturated solution. This might be due to the band broadening similar to the one we have observed in malic acid spectra. In addition, the band may also disappear if it is associated with a phonon mode. A closer look in the saturated spectra suggests very weak and broad absorption band between 1 and 1.75 THz. This might be fairing the band broadening and shift rather than being a phonon mode. In addition there is a very weak but also recognizable band located at 0.54 THz, though, the signal to noise level is too low to be sure. There is also a sign for a high frequency band beyond 2 THz.

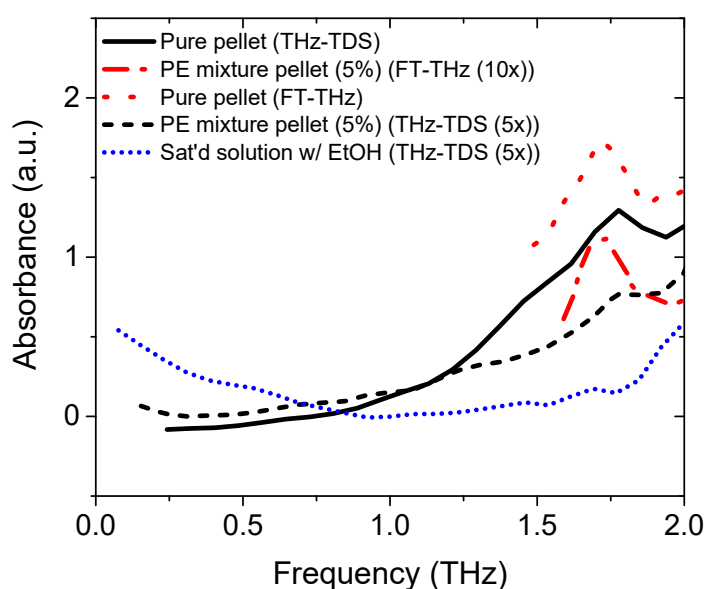


Figure 40. THz-TDS absorbance spectra of (+) Catechin Hydrate

3.5.2 Citric Acid

Citric Acid is one of the acidic compounds that present in wine. Its amount changes between 0.22 and 0.88 g/L for 16 different Croatian wine samples⁶⁹. Citric acid may be added to wine after process is finished in order to increase its acidity and adjust the taste⁷⁰. THz-TDS and FT-THz spectra of Citric Acid solid sample and solution were collected and presented in **Figure 41**.

FT-THz and THz-TDS spectra belong to both pure and PE mixture pellets have absorption features at the same frequencies, 1.72 and 2.14 THz. The reason for a better resolved absorption bands on FT-THz spectra is because the FT-THz spectrometer has a better frequency resolution for data collection. In this study, the THz-TDS spectrometer is limited with the reflection of the THz field in time domain due to sample thickness, that results in a lower resolution in the frequency domain and becomes a limitation for the instrument.

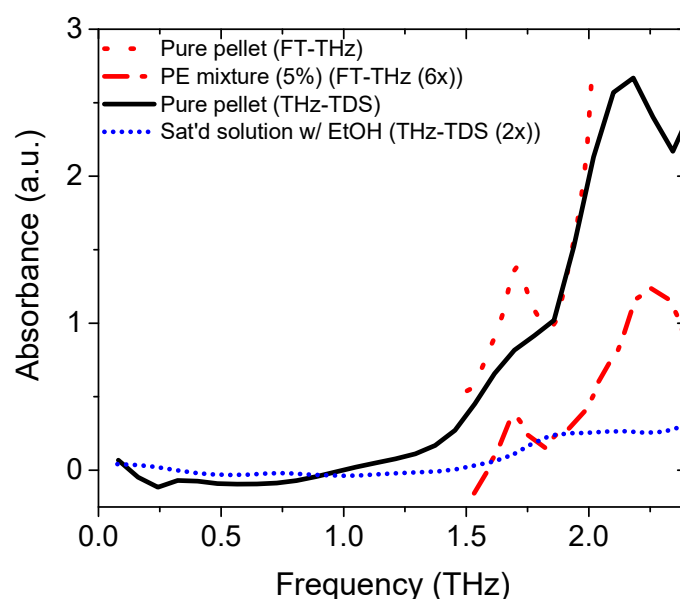


Figure 41. THz-TDS absorbance spectra of Citric Acid

3.5.3 DL-Malic Acid

Malic acid is one of the components found in grapes that the amount is highly affected by the climate. It increases with the cold summers and decreases as the climate gets warmer⁷¹. Its amount changes between 121 – 400 mg/L for 45 different red wine samples which belong to 2006 and 2007 vintages⁷². DL-Malic Acid, the racemic mixture, is used during the measurements in this study.

Figure 42 presents the THz-TDS and FT-THz absorption spectra of solid malic acid, and 5% malic acid in HDPE pellets along with its saturated solution in ethanol in a

frequency range of 0.1 and 2 THz. Especially in the TDS spectra of the solid form, clear and well resolved features are observed. The features are confirmed with the FT-THz within the common range of 1.2-2 THz. The strongest absorption feature within this range is observed at ca. 1.25 THz. Additionally, a fairly strong feature is observed at ca. 1.9 THz. However this band appears to be shifted in THz-TDS spectra of 5% sample. Considering the band is also observed clearly in FT-THz spectra of the same sample, this is most likely due to the very low signal-to-noise ratio at this region when the HDPE is used as host matrix. In addition to these, a weak but observable absorption band at 0.48 THz is also recorded in the THz-TDS spectrum of pure pellet. The feature is also noticeable in the 5% sample. Even though the solid spectra has a strong and a medium bands in its pure and mixture sample, THz-TDS spectrum of saturated solution does not show any of these absorption features.

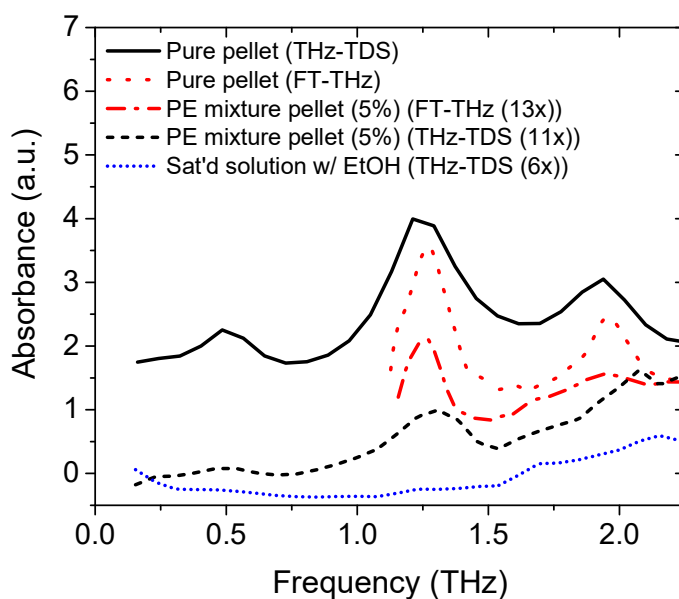


Figure 42.THz-TDS absorbance spectra of DL-Malic Acid

3.5.4 D (-) Fructose

Sugar is one of the most important factors for fermentation process and it is mostly found either in glucose or fructose form. It is consumed by the yeast during fermentation and remaining sugar is called residual sugar³⁸. The amount of residual fructose varies between 0.3 and 17.7 g/L in 26 different wine samples including red, white, ice and sparkling wines⁷³. D (-) Fructose is used for the measurements.

Figure 43 presents THz-TDS and FT-THz absorption spectra of pure and solution samples of fructose between 0.15 and 2.5 THz. There is no feature observable below 1.5 THz in any of the spectra. However, strong and well resolved absorption peaks are observed between 1.5 and 2.5 THz in the solid spectra. These features are confirmed by the FT-THz spectra of 5% sample and are located at ca. 1.75 and 2.2 THz frequencies. The reason why these nice features are not clearly observed in the THz-TDS absorption spectra of 5% sample is most likely because of the very low S/N in this region when HDPE is used. No absorption feature can be observed from saturated fructose solution in ethanol.

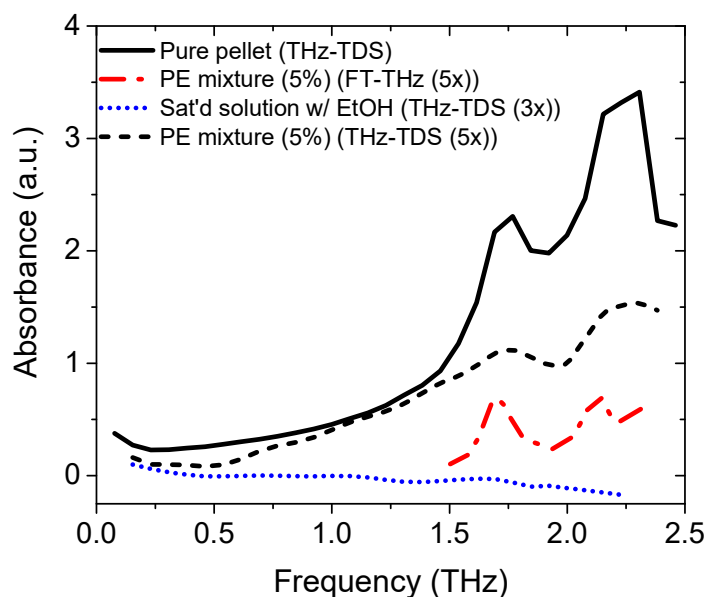


Figure 43. THz-TDS absorbance spectra of D(-)Fructose

3.5.5 (-) Epicatechin

(-) Epicatechin is one of the compounds in wine belongs to flavan-3-ols subclass under flavonoid group⁶⁷. Its mean amount is 40.58 mg/L for 76 different wine samples belong to Shiraz cultivar and it is one the significant contributors for cultivar discrimination⁴⁰.

Figure 44 presents THz-TDS and FT-THz absorption spectra of pure and solution samples of Epicatechin. There are distinct features of Epicatechin in the range that can be used for discrimination. A weak but significant absorption feature is observed at ca. 1.1 THz frequency both in the THz-TDS spectrum of pure pellet and also in the FT-THz spectrum of PE mixture pellet. In addition, a shoulder appears at ca. 1.6 THz, which broadens the main feature significantly. All the features are observed in the saturated solution spectra of Epicatechin in ethanol. The features, though, are all broaden compared to the solid forms. The strongest feature appears to lose its intensity and red-shift toward lower frequencies. On the other hand, surprisingly there appears to be very small shift (if any) for the shoulder or the band at 1.1 THz. It can be concluded that these features result from an intra-molecular motion of the compound, thus does not appear to be significantly affected by the environmental changes.

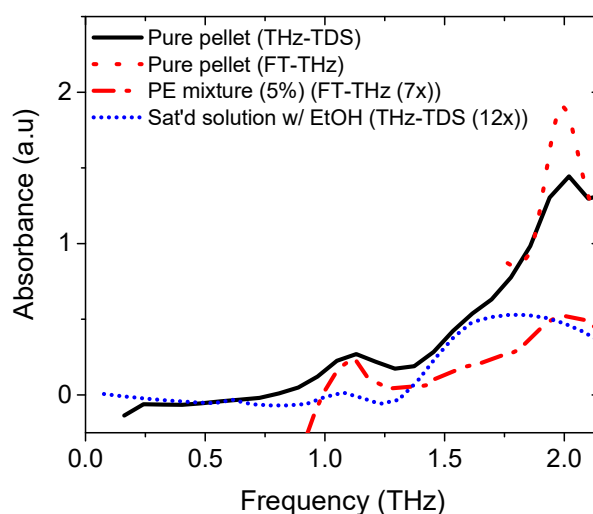


Figure 44. THz-TDS absorbance spectra of (-) Epicatechin

3.5.6 4-Ethylphenol

The amount of 4-Ethylphenol varies greatly in different kind of wines and it highly affects the odor of wine especially if it is in high concentration. Its concentration varies between 2 $\mu\text{g/L}$ – 2660 $\mu\text{g/L}$ for 61 different bottle of commercial Australian wines⁷⁴.

THz-TDS and FT-THz absorption spectra of pure and solid solution samples of 4-Ethylphenol are given in **Figure 45**. It has a broad absorption profile between 1.23 and 1.61 THz frequencies. The features observed in solid sample are not strong enough to really determine the nature and position of these bands, possibly due to the low light transmission from the pure sample. On the other hand, the features of 5% 4-Ethylphenol in HDPE is clearer in the THz-TDS spectrum. The broad nature of the bands is clearly observed. A broad band at 1.6 THz is also recorded in FT-THz spectrum of pure pellet. Unfortunately, the solution spectra of 4-Ethylphenol could not be collected.

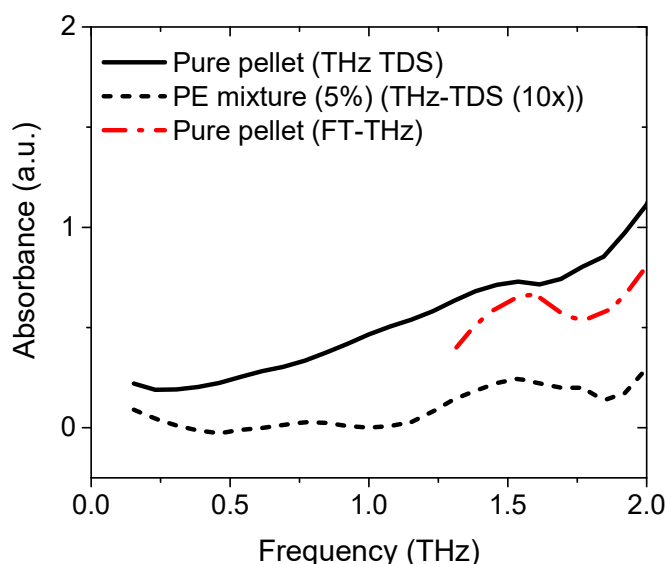


Figure 45. THz-TDS absorbance spectra of 4-Ethylphenol

3.5.7 Gallic Acid

Gallic acid is one of the compounds that affects the color of red wine through contribution to copigmentation⁷⁵. Concentration of gallic acid varies between 28.34 and 100.73 mg/L in the 11 commercial Sicilian red wines from Nero d'Avola cultivar⁶⁸.

Figure 46 shows THz-TDS and FT-THz absorption spectra of pure and solution samples of gallic acid. Similar to the other acids, the spectra are rich with the absorptions bands, which are nicely observable in all the spectra including the saturated solution sample. THz-TDS spectrum of pure pellet gallic acid has three main features. Intense absorption bands are located at ca. 1.5 THz 2.1 THz. The weak and relatively broader feature is at 1.05 THz. The sharp absorption features at 1.5 and 2.1 THz frequencies are also clearly observed in FT-THz spectra of both pure and PE mixture pellets. THz-TDS spectrum of saturated solution has similar absorption features with the pure pellet spectra at 1.1 and 1.53 THz frequencies. In addition, the rise toward the 2 THz in the spectra also suggests that the band would have been observed if the bandwidth would have been reached with the saturated solution. The existence of all the features in solution strongly suggests that the observed features are all intra-molecular modes.

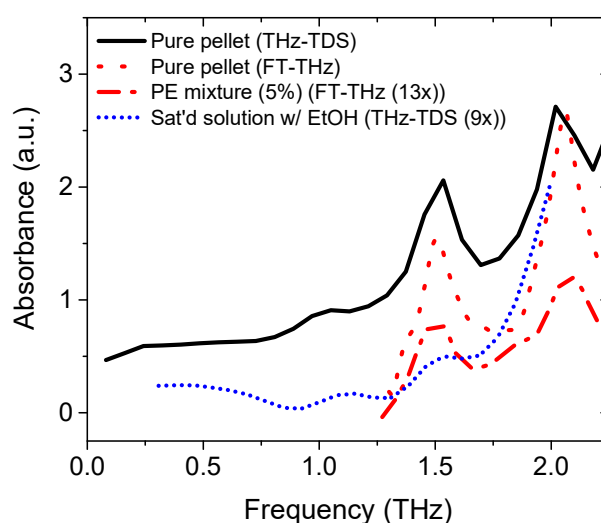


Figure 46.THz-TDS absorbance spectra of Gallic Acid

3.5.8 D (+) Glucose Monohydrate

Glucose is one of the most abundant sugar content in wine with fructose. It is consumed during fermentation and the amount of residual glucose in 16 different wine samples varies between 0.2 – 15.4 g/L⁷³. D(+) Glucose Monohydrate is used for all of the measurements.

Figure 47 displays THz-TDS and FT-THz absorption spectra of pure and solution samples of glucose. It is exciting to observe reasonably strong, well resolved features of the glucose in all the solid samples. On the other hand, there is no observable feature below 1.5 THz in all the spectra. For THz-TDS spectrum of pure pellet, those are located at 1.84 and 2.40 THz frequencies. Even though the feature at 1.84 THz is observed as a single band in the THz-TDS spectra, the feature is resolved into two bands in the FT-THz spectra of both pure and PE mixture pellets. This is most likely due to the lower resolution in the THz-TDS spectra due to instrument and sample limitations. Spectrum of saturated solution does not have observable absorption features as the solid samples do.

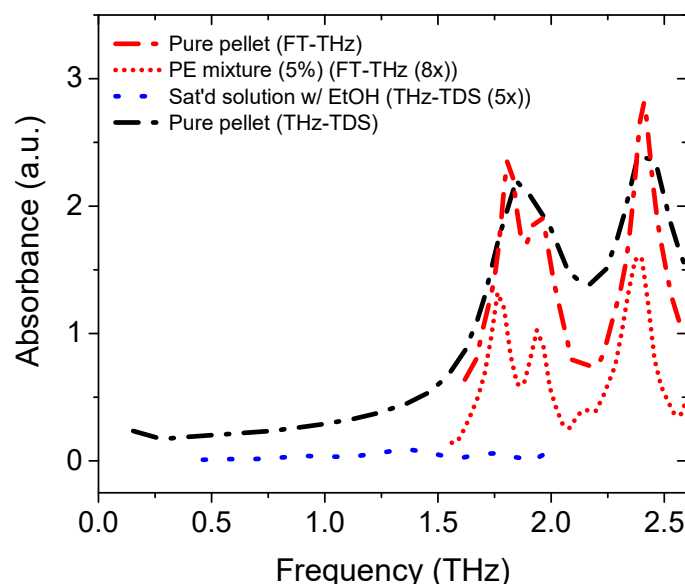


Figure 47. THz-TDS absorbance spectra of D(+)-Glucose Monohydrate

3.5.9 Quercetin

Quercetin, is found in wines, is known for its antioxidant effect⁷⁶. It is present in wine in both conjugated and free forms. Concentration of free quercetin in 65 different types of wine samples varies between 0.3 – 15.8 mg/L⁷⁷.

THz-TDS absorption spectra of pure and solution samples of Quercetin are given in **Figure 48**. It does not have a sharp absorption feature, though has a very broad but reasonably strong absorption band around 2.25 THz. In addition, it has an increasing absorption profile with frequency in range of 0.15 and 2.5 THz. Possibly this is due to scattering effect of the samples that could not be corrected with reference. The solution spectrum shows a band located at ca. 2 THz, which is most likely the same band but red shifted as it has been seen in other solution samples.

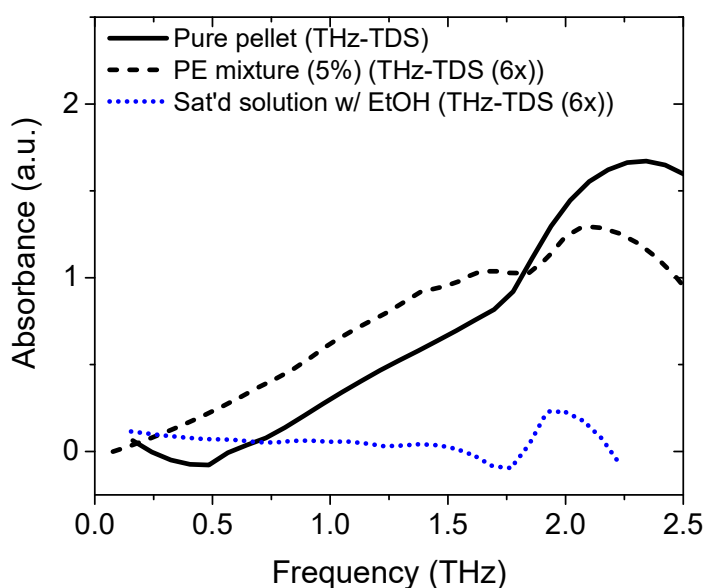


Figure 48.THz-TDS absorbance spectra of Quercetin

3.5.10 Succinic Acid

Succinic acid one of the components that is responsible for salty and bitter taste in wine and formed after fermentation. It is stable in wine because it is resistant to

microbial attack³⁸. Typical concentration of succinic acid in wines is between 0.5 – 1.5 g/L⁷⁸.

Figure 49 presents THz-TDS absorption spectra of pure and solution samples of succinic acid. THz spectra of the both samples are rich in features but unfortunately they are broad and overlapping, especially the ones of solid sample. The strongest feature is around 2.25 THz while there are two similar intensity bands around 1.25 and 1.75 THz. They are better resolved in the solution spectra. The fact that they exist in solution suggest that these are intra-molecular modes of the succinic acid.

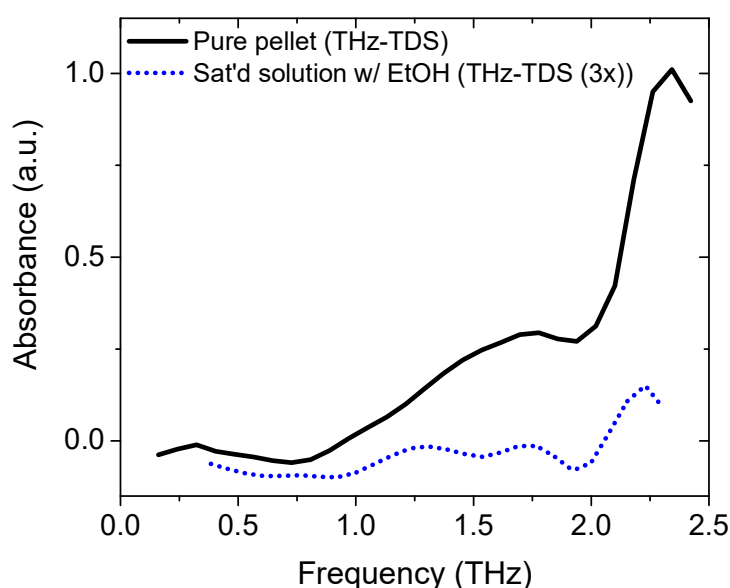


Figure 49.THz-TDS absorbance spectra of Succinic Acid

3.5.11 Tannin

Tannins are the polymeric compounds that give wines their astringency and cause dry feeling in the mouth. Kallithraka and Bakker showed that peak areas on chromatogram belong to saliva proteins decreases after consumption of grape juice and wine⁷⁹. Results suggest that when those proteins combine with phenols, they precipitate and result in astringent feeling in the mouth.

There are two main types of tannins in wine which are named as hydrolysable and condensed tannins. Condensed tannins are flavonoid derived and consist of large number of flavan-3-ol molecules⁸⁰. Both types of compounds mostly come from the grapes³⁸.

In **Figure 50** it is seen that even though there is no sharp absorption band in the pure pellet THz-TDS spectrum, there is a shoulder type absorption band at 1.7 THz which can easily be spotted. Increasing absorption profile between through our bandwidth might be the scattering effect. The features seen at 1.7 THz on the solution spectrum might verify the possibility that this signal is actually the intra-molecular band.

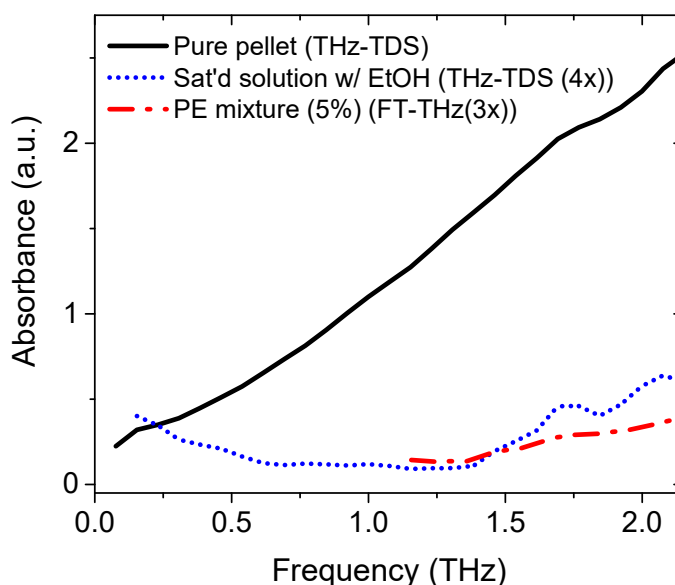


Figure 50. THz-TDS absorbance spectra of Tannin

3.5.12 Tartaric Acid

Tartaric acid is the other major grape acid as malic acid; however, amount of tartaric acid does not change during ripening, therefore it is used in order to increase the wine's acidity because of the fresh taste that the acidity gives³⁸. Amount of tartaric acid changes between 0.8 – 2.98 g/L for 16 different Croatian wines⁶⁹.

Figure 51 shows THz-TDS and FT-THz absorption spectra of pure and solution samples of tartaric acid. There is a sharp and strong feature at ca. 1.1 THz and a broad and weak absorption feature at 1.85 THz in the THz-TDS spectrum of solid sample. Those features match with the absorption bands in THz-TDS spectrum of PE mixture sample and also FT-THz spectrum of pure solid sample. The strong feature observed at ca. 1.1 THz of the solid sample disappears in solution spectra while the weak one red shifts. The fact that the weak one exists while the strong one disappears clearly shows that the band at 1.1 THz is a phonon mode while the weak one at 1.85 is an intra-molecular band.

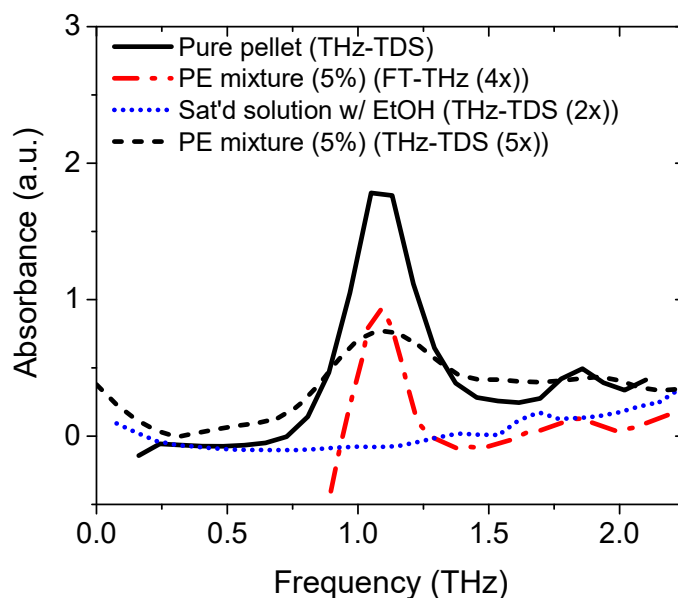


Figure 51. THz-TDS absorbance spectra of Tartaric Acid

3.6 Wine Measurements

Before starting the wine measurements, reviews about the wine brands and ratings are taken from the Vivino⁸¹ software and listed in **Table 9**. This application is used at least once by 30 million people around the world and has approximately 100 million ratings about 10 million different wines. Basic arithmetic was applied on the ratings of our wine samples in this thesis and it was decided that the samples with

3.3 or higher rating (out of 5) are “*higher quality samples*” compared to the others. It is important to note that the taste, odor and color of wines can change by the harvesting year because of climate effects. Therefore, ratings could change; however, it was not possible to gather all of the rating information considering the harvesting year of the wine samples one by one, because there was not enough information about the years neither on the bottles, nor in the software. The given are the overall ratings for the samples with the *same brands* and *matching cultivars*, therefore only difference between the sample listed in the software and in this thesis might be the samples production year.

Table 9. Ratings of wine samples

Sample #	Brand	Score (out of 5)	# of ratings	Multiplication (Score * number of ratings)
1, 2, 4	<i>Buzbağ</i> (red)	3.3	738	2435.4
3, 9	Angora (red)	3.1	1351	4188.1
5	Mistik (red)	2.9	36	104.4
6	Cadde (red)	3.2	17	54.4
7	Turasan – Peribacası (red)	2.9	79	229.1
8	Doluca (red)	2.9	244	707.6
10	<i>Cumartesi</i> (red)	3.3	44	145.2

11	*no info* (rose)	-	-	-
12	<i>Sandeman</i> (white)	3.3	1035	3415.5
13	Donini Collezione (red)	3.2	877	2806.4
14	Buzbağ – Diyarbakır (red)	3.2	138	441.6
15	<i>Château</i> <i>Vartely</i> (red)	3.5	185	647.5
16	Terra – Varietal Series (red)	3.2	153	489.6
17	<i>Akberg</i> (red)	3.5	15	52.5
18	<i>Buzbağ –</i> <i>Elazığ (red)</i>	3.5	333	1165.5
19	Turasan – Cappadocia (red)	3.0	98	294
20	<i>Château</i> <i>Vartely</i> (red)	3.5	544	1904
21	Akberg (red)	2.5	5	12.5
22	<i>Akberg</i> (red)	3.5	5	17.5
23	Sade (red)	3.1	121	375.1

24	<i>DLC (red)</i>	3.4	83	282.2
SUM				19768.1
MEAN				3.24

Absorbance, refractive index and dielectric properties of samples were calculated after measurements using purged air with sample cell windows as reference and the results are given in **Figure 52-57** respectively. Even though there is no distinct feature in the absorbance spectra, there appears to be a linear increase in absorption as the frequency increases. In addition, it is clearly seen that absorbance of Sample 12 is lower than the other wine samples in all frequencies. This can be explained with higher alcohol content (ca. 19.5%) compared to the others (13.5% on average). Even though there is no feature that may help on identification of the wines, there are small but significant differences in absorption values of wines that could be used for classification. The further details will be discussed in statistical treatment of the data with PCA.

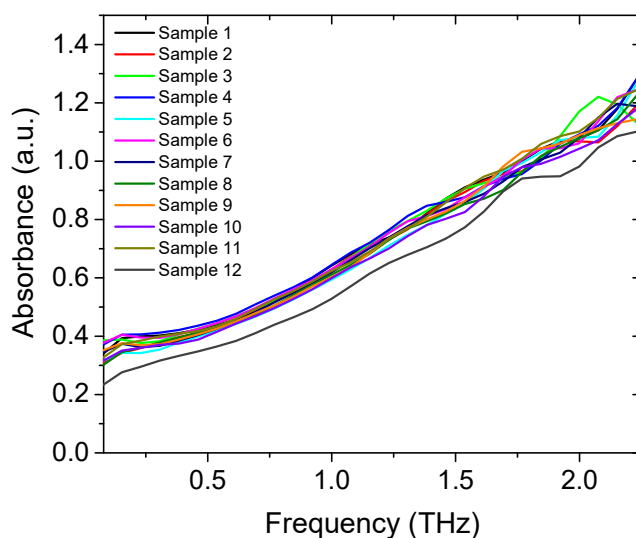


Figure 52.THz-TDS absorbance spectra of wine samples (Sample 1-12)

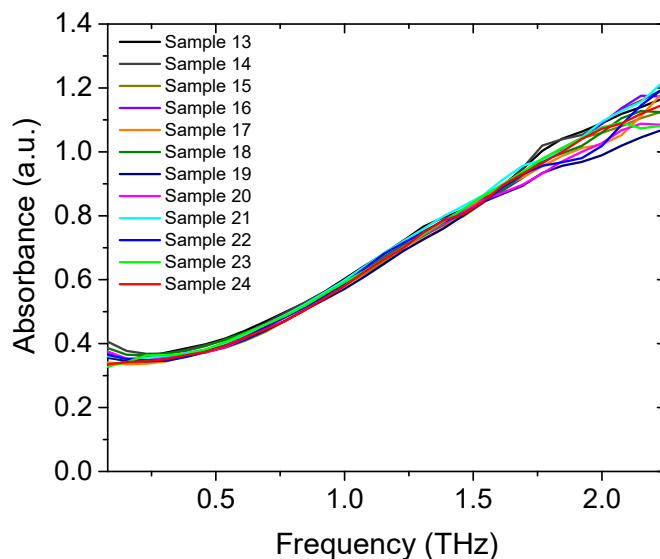


Figure 53. THz-TDS absorbance spectra of wine samples (Sample 13-24)

The refractive index change with frequency of all wines samples are given in **Figure 54** and **Figure 55**. As opposed to the absorption profile, the index decreases with frequency. Even though there is no feature similar to the absorption profile, there are significant enough differences in their index values. The indexes are around 2.3 and 2.4 at the very low frequencies (< 0.5 THz) while they are around 1.9 and 2.1 at 2.25 THz. On average the index is ca. 2.2. The refractive index of the one with the higher alcohol content (19.5%, Sample 12) is less than the ones of other samples through 0.3 and 1.5 THz while the refractive index of the Sample 6 is significantly higher than the rest.

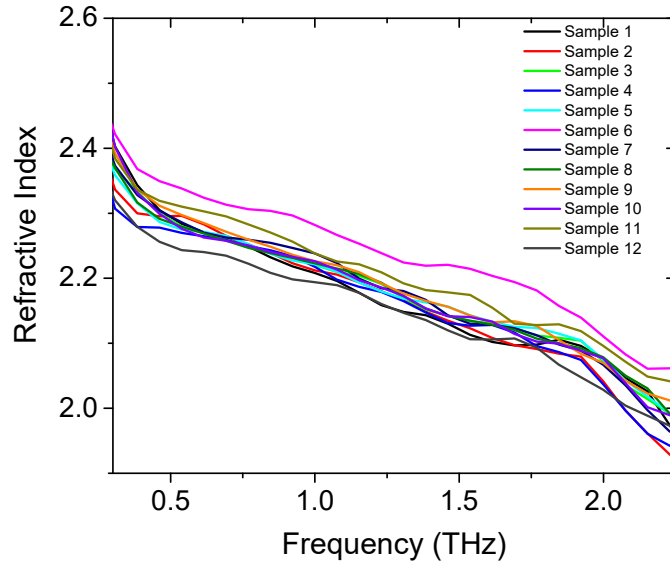


Figure 54. Refractive indices of wine samples (Sample 1-12)

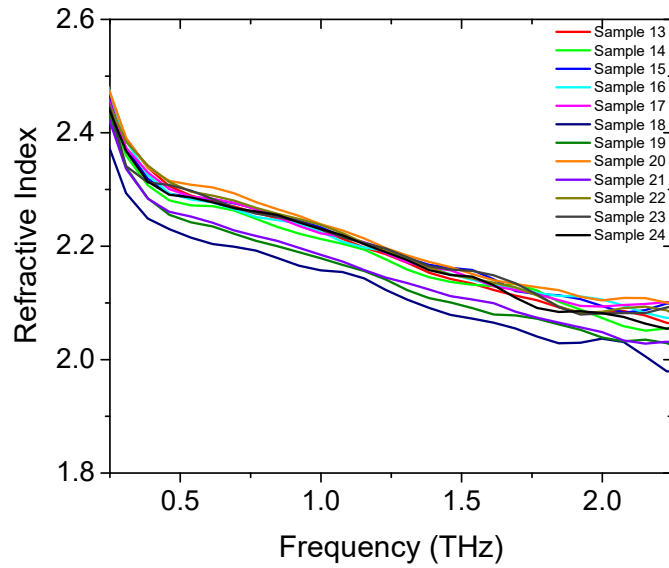


Figure 55. Refractive indices of wine samples (Sample 13-24)

The dielectric constants are unique properties of materials thus can also be used for material characterization. **Figure 56** and **Figure 57** present frequency dependent real parts of the dielectric behavior of the wine samples. While most of the wines samples' behavior is similar, the samples 6, 17, 19, and 24 separate themselves from

the general behavior of the rest. And these differences do appear in the PCA analysis of the sample, which is discussed in the next part.

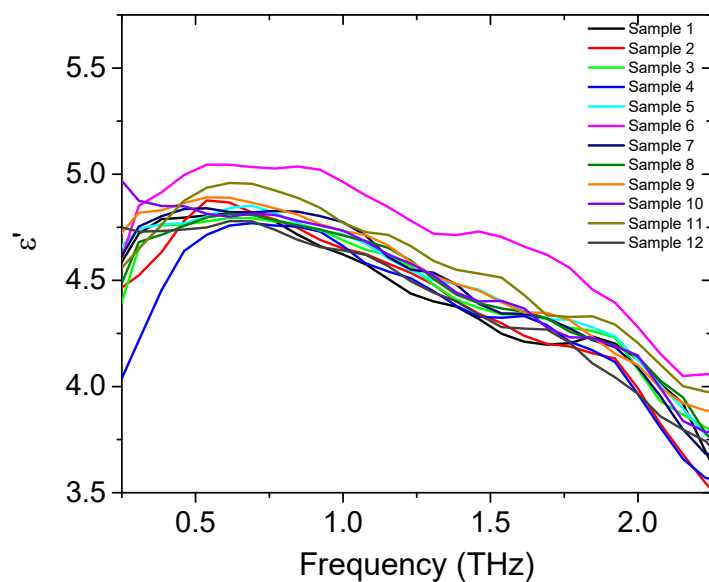


Figure 56. Calculated dielectric real parts of wine samples (Sample 1-12)

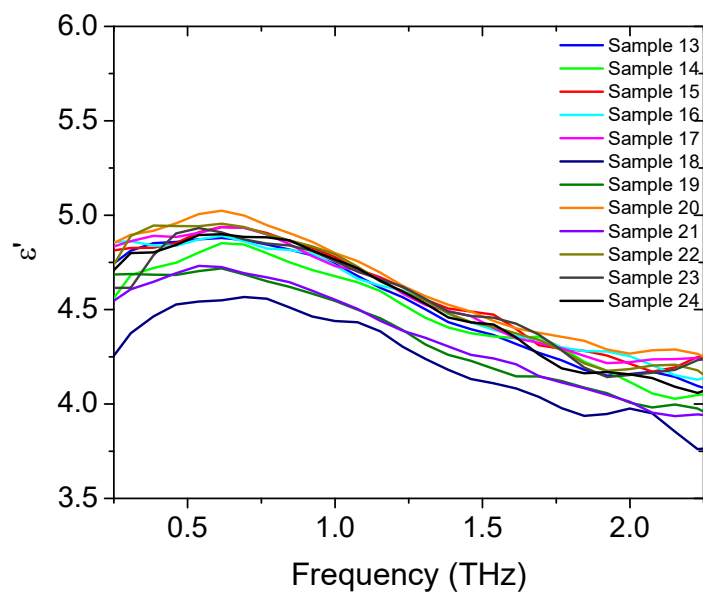


Figure 57. Calculated dielectric real parts of wine samples (Sample 13-24)

3.7 PCA Results

Information of absorbance, refractive indices and dielectric properties are used for Principal Component Analysis (PCA). Figures are plotted by using first three principal components and analyses on the figures are made.

Figure 58 presents the PCA analysis of the absorbance spectra of the wine samples in the range 0.23 – 1.92 THz frequencies. As it can be seen in the figure, wine samples with rating 3.3 and higher are grouped on the left side (with negative PC1 values) in the PCA graph which are marked with red squares. However, it is observed that three other samples are at the right side of the graph, which are marked with dashed blue lined squares, even though their rating is 3.3 and seem to be closer to each other, when compared to other group that is squared. The common feature of those wine samples is their brand. They all are Buzbağ samples, so there might be more common features among them that results in wide discrimination from the other main group. In addition, the reviewers of this brand may not be classified among the other since those are usually low income group, such as students. Thus, the analysis may have rightfully grouped those with respect to their quality. Also, distribution in PC1 to PC3 plot shows also the similar tendency with PC1 to PC2 plot [Figure 59]. Therefore, it can be concluded that both PC1 to PC2 and PC1 to PC3 plots done with absorbance spectra might be able to group the wine samples with higher quality when compared to the others.

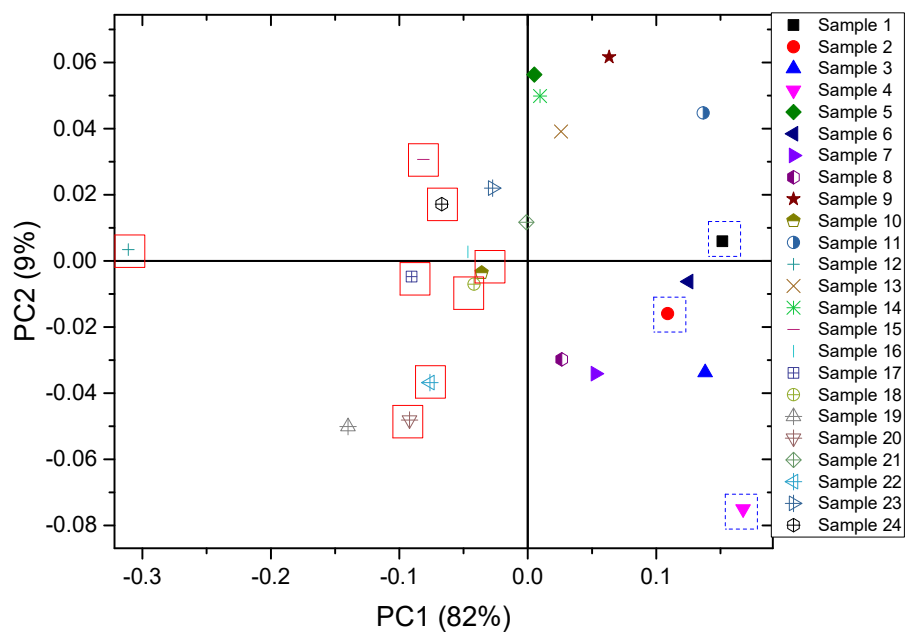


Figure 58. PCA graph done with absorbance spectra of wine samples indicating higher rating samples with red solid and blue dashed squares (blue dashed ones are Buzbağ samples) (PC1 to PC2)

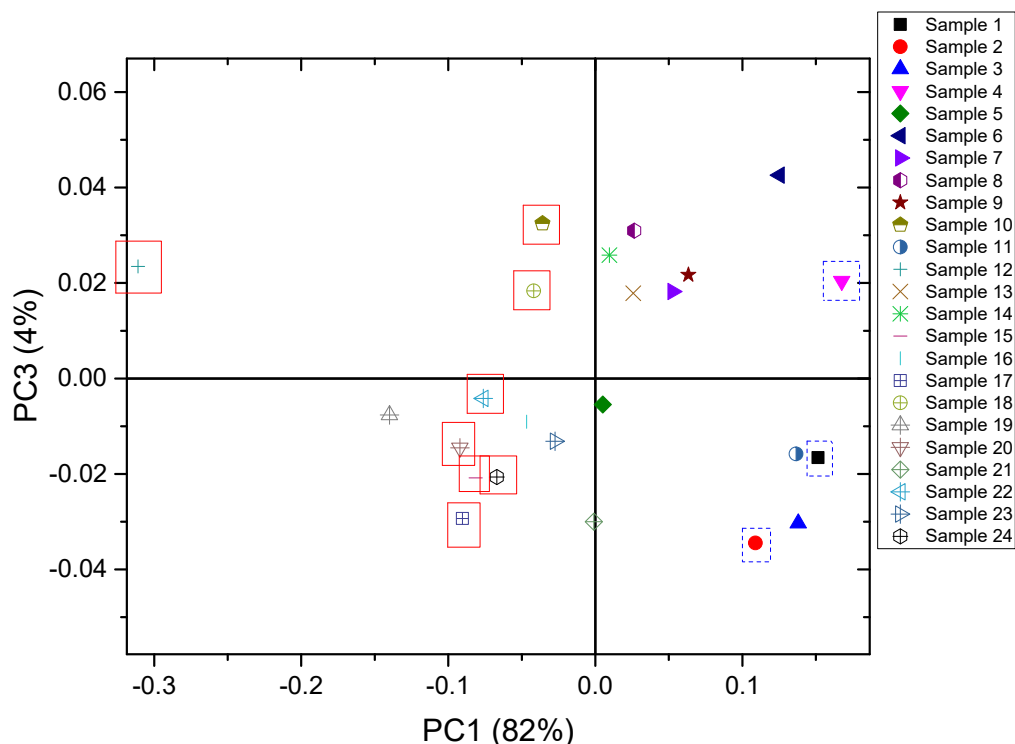


Figure 59. PCA graph done with absorbance spectra of wine samples indicating higher rating samples with red solid and blue dashed squares (blue dashed ones are only Buzbağ samples) (PC1 to PC3)

In **Figure 60**, PC1 to PC3 plot according to absorbance spectra actually shows another tendency in the distribution. It is seen that, the wine samples made from non-native grapes (Cabernet Sauvignon, Merlot etc.) have a tendency to make a group with each other with the negative PC3 values on the graph, which are marked with circles. There are only 2 wine samples that do not join that grouping. Therefore, it might be concluded that PC1 to PC3 plot may give clues about discrimination of wine samples according to their type of cultivars as native and non-native ones in addition to wine ratings, hence wine quality.

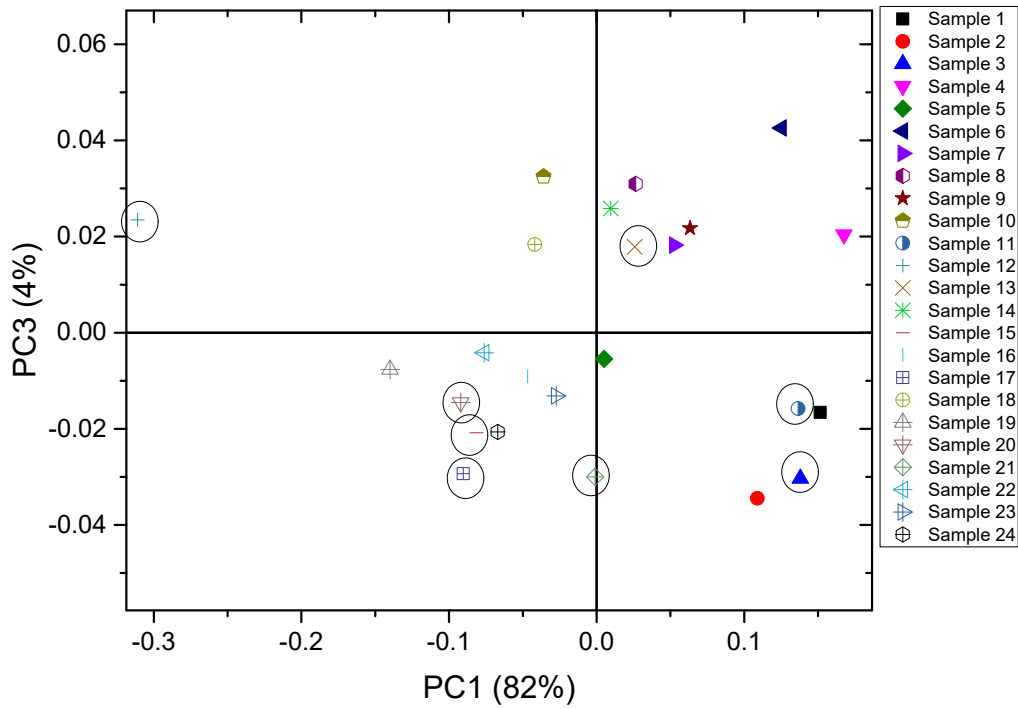


Figure 60. PCA graph done with absorbance spectra of wine samples indicating non-native type of cultivars with circles (PC1 to PC3)

In **Figure 61**, PC1 to PC2 plot was plotted by considering the absorbance spectra in the narrower range. In that graph, Sample 1 was not considered because it was observed as outlier and samples from number 2 to 24 are included.

All of wine samples are marked according to their type of cultivars (as native and non-native) with circles and also to their ratings. Squares with dashed lines belong to Buzbağ samples, while other squares mark other samples with rating 3.3 or higher. It was realized that wine samples made from non-native grapes with higher qualities have a tendency to make a group among each other on the left top quadrant, in general. In addition, PC1 to PC3 graph for the same range absorbance spectra show the similar grouping, however, they are getting more widespread among two quadrant [**Figure 62**]. On the other hand, Sample 12 is getting closer to the

aforementioned group. Therefore, the results suggest that both PC1 to PC2 and PC1 to PC3 plot of PCA using a narrower range on absorbance spectra of samples may be able to discriminate wines made from non-native grapes with higher qualities.

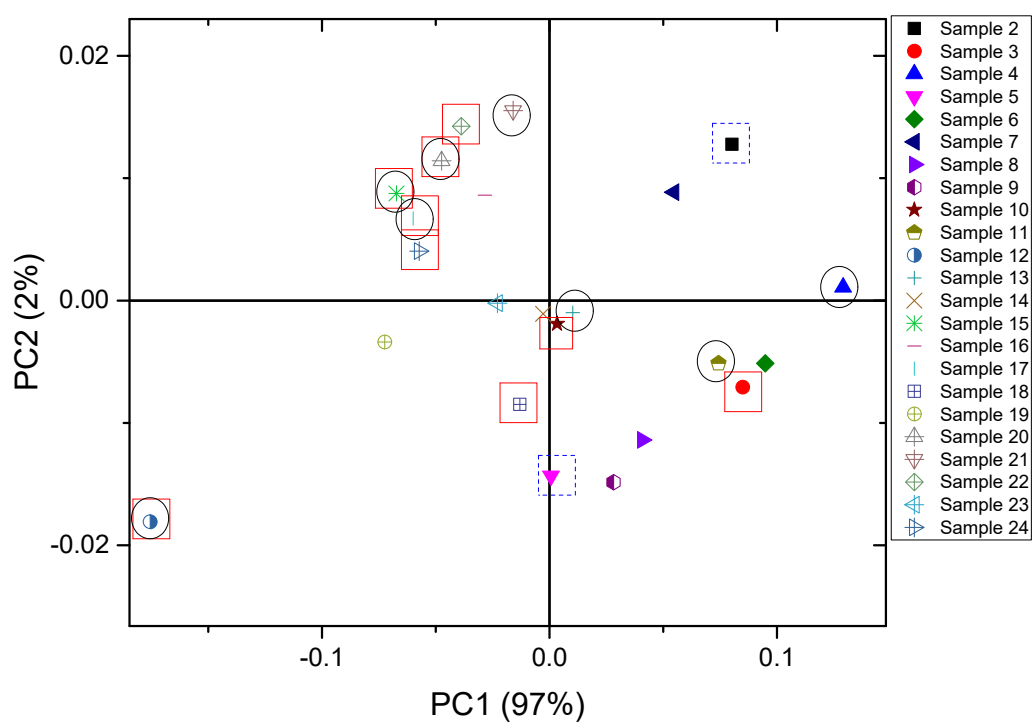


Figure 61. PCA graph done with absorbance spectra (narrow range) of wine samples indicating non-native type of cultivars with circles and higher rating samples with red solid and blue dashed squares (blue dashed ones are only Buzbağ samples) (PC1 to PC2)

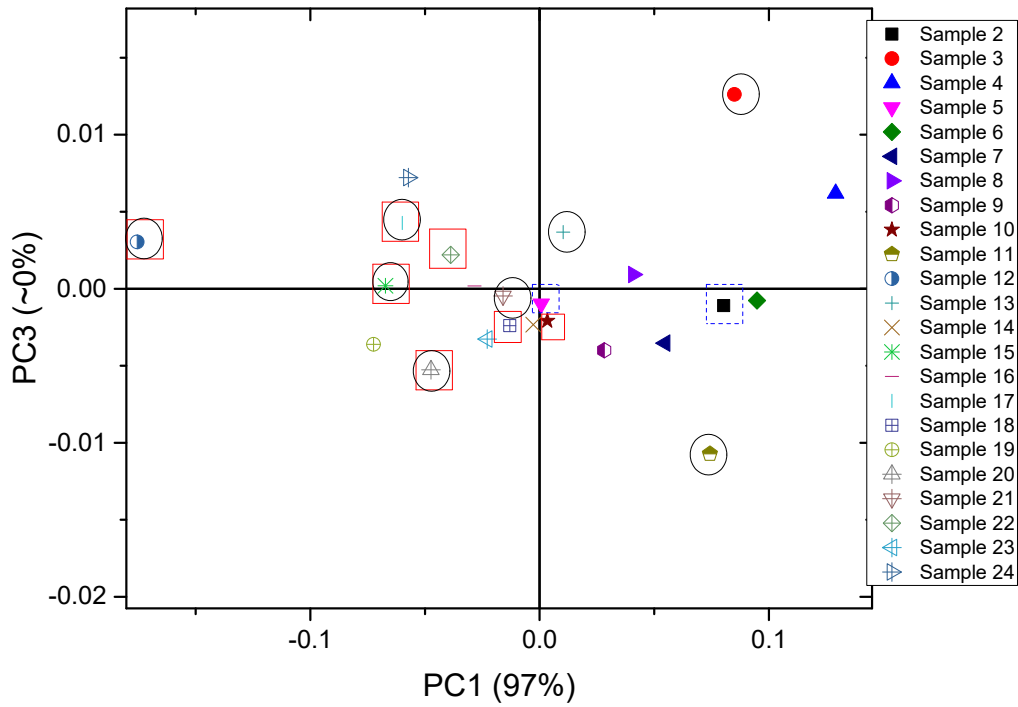


Figure 62. PCA graph done with absorbance spectra (narrow range) of wine samples (PC1 to PC3)

Figure 63 presents PC1 to PC2 plot by using the refractive indices of samples. Here, it was observed that some of the samples made from same type of grapes can be grouped. Wine samples made from mixture of Öküzgözü and Boğazkere grapes; and samples made from only Öküzgözü grapes are marked on the figure. On the other hand, PC1 to PC3 graph also shows the similar tendency [**Figure 64**]. Samples from Öküzgözü/Boğazkere mixture, Cabernet Sauvignon/Alicante/Merlot/Öküzgözü mixture, Boğazkere/Kalecik Karası mixture and from only Merlot grapes are marked as grouped samples. Additionally, it is observed that Öküzgözü/Boğazkere samples are placed on the plot almost between Boğazkere and Öküzgözü samples individually for both figures. Thus, it can be concluded that PCA by using refractive indices might be able to give clues about the cultivar information about the wine samples.

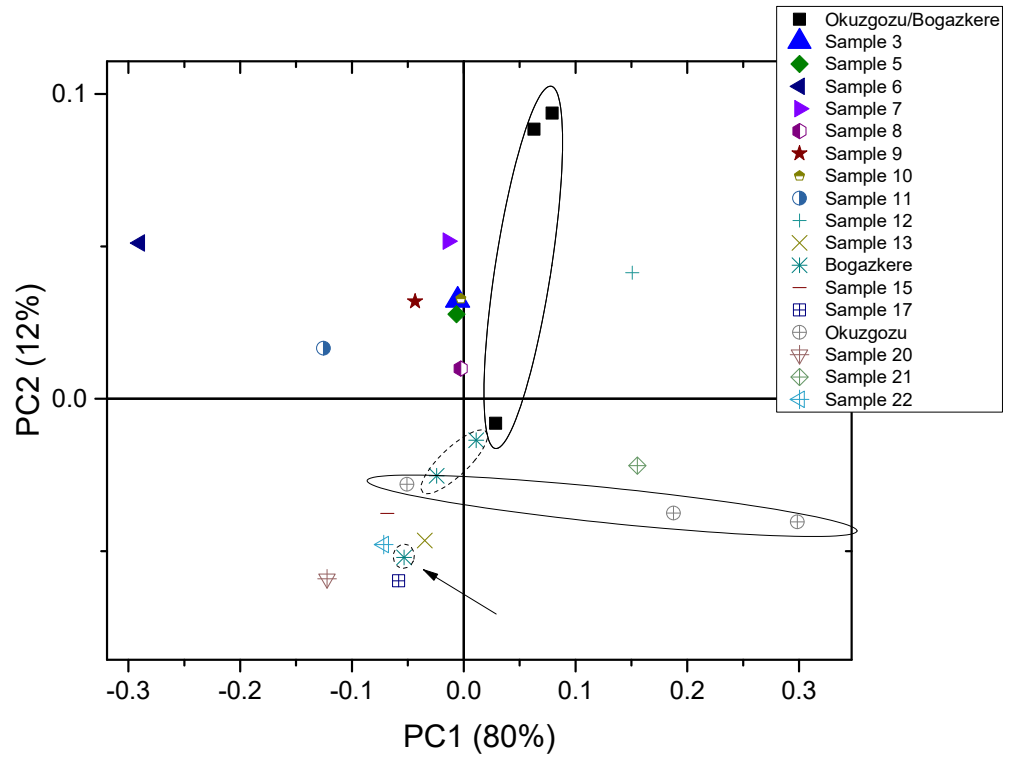


Figure 63. PCA graph done with refractive indices of wine samples (PC1 to PC2)

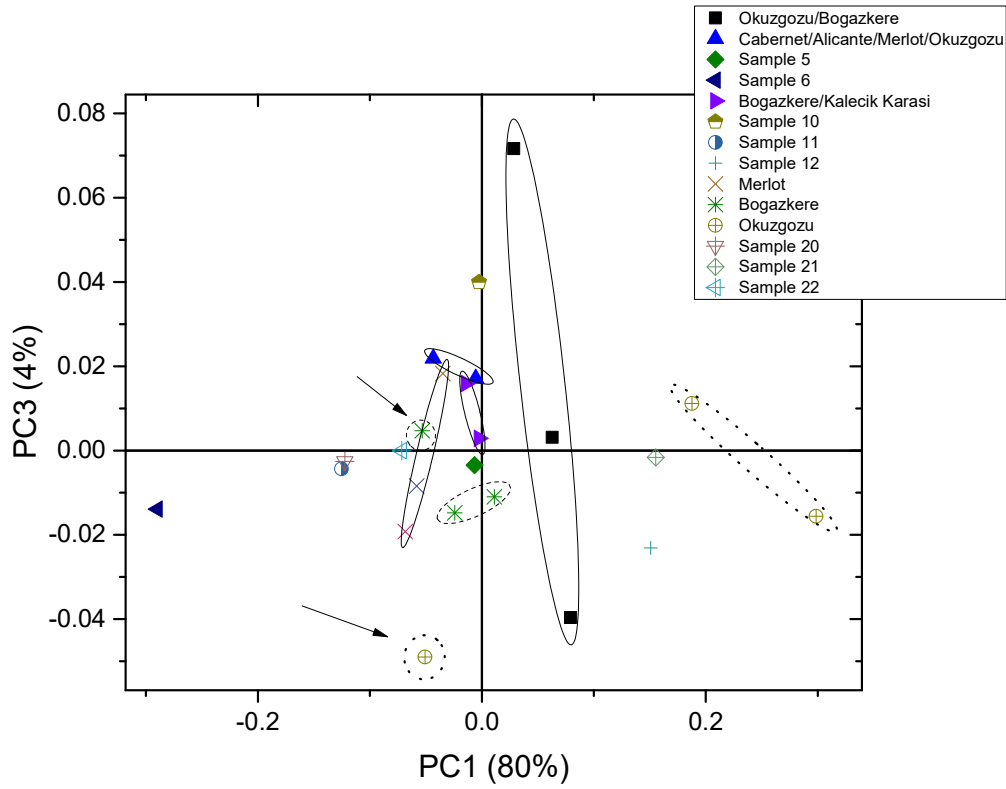


Figure 64. PCA graph done with refractive indices of wine samples (PC1 to PC3)

In **Figure 65**, information of dielectric properties of wine samples were used in order to plot the PC1 to PC2 graph. In that case, analysis can highlight the Boğazkere samples, while it was Öküzgözü samples for the analysis through refractive index information. In this graph, samples made from only Boğazkere grapes and also Boğazkere/Kalecik Karası mixture can be grouped among each other. In addition, PC1 to PC3 graph not only could be able to group Boğazkere/Kalecik Karası and Boğazkere samples, but also Öküzgözü/Boğazkere and Cabernet Sauvignon/Alicante/Merlot/Öküzgözü samples can be marked on the **Figure 66**. In addition, samples from Öküzgözü/Boğazkere mixtures are observed just between the Öküzgözü and Boğazkere samples on the plot.

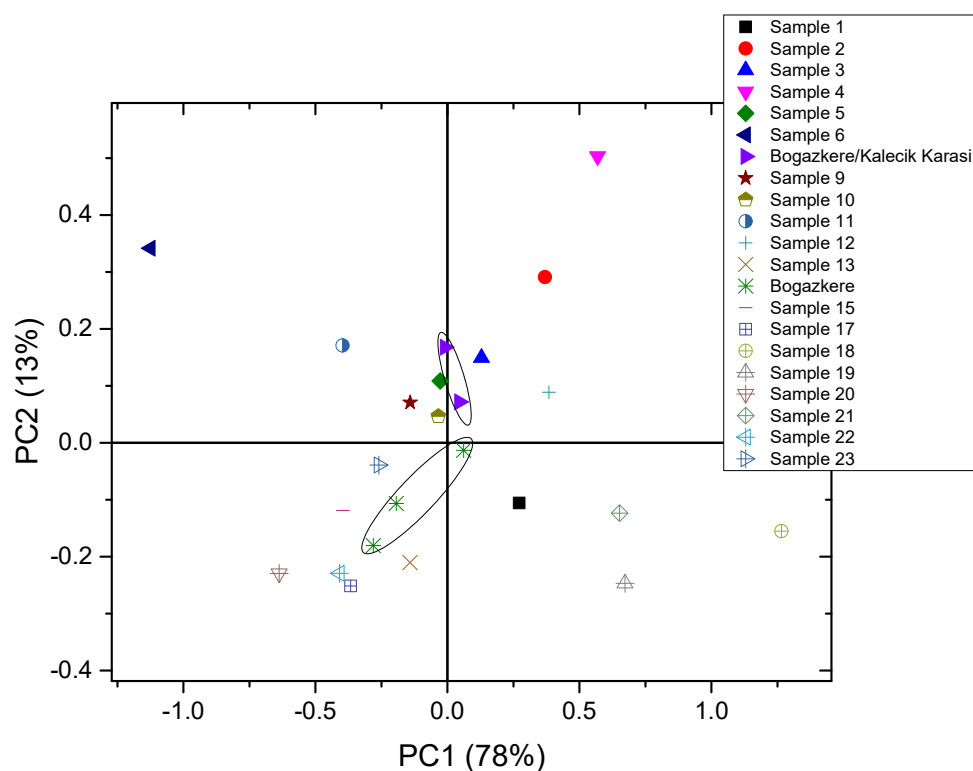


Figure 65. PCA graph done with dielectric real part of wine samples (PC1 to PC2)

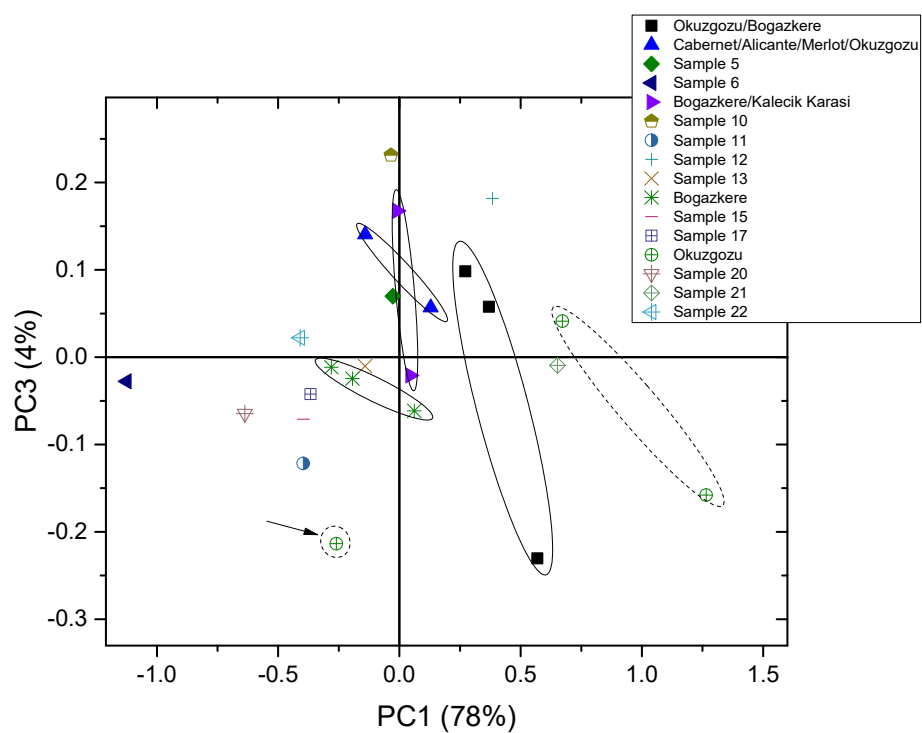


Figure 66. PCA graph done with dielectric real part of wine samples (PC1 to PC3)

CHAPTER 4

CONCLUSION

In this study, the use of THz-TDS and FT-THz spectroscopy techniques were investigated for wine chemicals and commercial wine analysis. The main aim was to determine chemical markers to identify the chemical content and its effect on the wine analysis and also to be able to classify wines according to their qualities and/or cultivars, grape types etc. Briefly, in this study liquid sample cell parameters were optimized for each of the spectroscopy system and spectra of the various wine chemicals were collected with both THz-TDS and FT-THz instruments. Finally, wine samples were analyzed by using THz-TDS. The distinct spectral features of the wine chemicals have been observed when solid samples were measured. However, no strong spectral features of many of the wine chemicals were observed in solution samples, that hindered their use in wine analysis. Therefore, overall amplitude differences on frequency dependent profiles of absorbance, refractive indices and dielectric properties of different wine samples were used for wine classification. The analysis was done with PCA. It was shown that combination of Terahertz spectroscopy and PCA could be able to make a correlation between absorbance spectra of the wine samples and their types of cultivars, as well as their relative quality ratings from users. Additionally, refractive indices and dielectric properties of samples can make a correlation with the types of grapes that the wine samples made from.

The study composed of three different parts, sample cell optimization, spectral analyses of wine chemicals, and wine samples. Firstly, liquid sample cell was tried to be optimized as much as possible because of water interference in the spectra. Since types of light sources and their power in the instruments were different and their capabilities to penetrate through the sample were highly different, various thicknesses for the sample cell were investigated. As it was predicted, thickness that works well for THz-TDS was too much for measurements done with FTIR instrument. While even 70 μm thickness is suitable for THz-TDS measurements, up

to only 40 μm thickness sample cell can be used with FT-THz instrument, which made the measurements quite difficult. Both techniques are used complimentary to each other in order to determine the nature of the observed vibrational bands. The comparison of solution measurements with the solid samples showed that many of the features observed from wine chemicals were actually result of intra-molecular motion, while few of those are phonon modes. A measurement of solid samples only could not reveal this information. However, it was realized that high amount of chemicals was need in order to observe in solution. This hinders their use in chemical identification in wine analysis and its use toward wine quality analysis. As a conclusion, quality analysis would not be investigated qualitatively through wine chemicals. Thus, a statistical approach is considered.

PCA analysis of the frequency dependent absorbances, indices and real part of the dielectric constants were done in order to observe possible classification of the wines according to their similarities and difference. It was observed that while absorbance spectra of samples might discriminate and group wines according to their overall rating scores given by the consumers, it can also give clues about differentiation of wines that are made from non-native grapes. On the other hand, refractive indices and information about dielectric properties of wines may help on grouping wines according to their cultivars. Those analyses seem to work especially for the native cultivars like Öküzgözü and Boğazkere.

As a result, combination of THz-TDS and PCA can give valuable information about quality and origin cultivar of wines. Since THz-TDS also has a future to be used in situ, the technique could be promising for both wine producers and consumers.

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