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Chromatographic Methods Applied to the Characterization of Bio-Oil from the Pyrolysis of Agro-Industrial Biomasses

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Abstract

Biomass conversion into solid, liquid and gaseous products by pyrolytic technology is one of the most promising alternative to convert the biomass into useful products and energy. The total characterization of the products from the pyrolysis of biomass is one of the great challenges in this field, mainly due to their molecular complexity. Pyrolysis is a process that causes degradation of biomass in a non-oxidative atmosphere, at relatively high temperatures, producing a solid residue rich in carbon and mineral matter, gases and bio-oil. The yield and properties of the products depend on the nature of the biomass and the type of the pyrolysis process (type of reactor, temperature, gas flow, catalyst). Due to the high molecular complexity of bio-oil, many different technical had been developed to their complete characterization. This chapter describes the principles of the techniques and main application of chromatographic methods (GC, LC, GC × GC, LC × LC, Nano-LC) in the analysis of bio-oils derived from thermo-degradation of biomasses. Especial attention is carried out to two-dimensional techniques that represent the state of the art in terms of separation, sensibility, selectivity and velocity of data acquisition for characterization of complex organic mixtures. For proper use of bio-oil in the chemical industry, it is essential the identification and unambiguous determination of its major constituents. Only then, it is possible to propose a recovery route of some of these components for the development of an industry dedicated to a bio-refinery. For this, chromatographic methods, especially GC × GC/MS, are fundamental because they allow analysis with high sensitivity and accuracy in identifying each constituent of the bio-oil.

Keywords: Chromatography, bio-oil, biomass, pyrolysis

1. Introduction

The current world energy scenario shows a tendency to decrease the use of mineral resources source, considering its environmental impact, the socioeconomical and market problems if compared to renewable sources [1–3]. In addition, the diversification of energy sources is necessary in order to meet the growing energy demand. In this context, it arises the biomass coming from different sources of natural resources, which is a renewable alternative, abundant and suitable for competition with conventional fossil fuels [3, 4]. The biomass conversion into solid, liquid and gaseous products by pyrolytic technology is a promising alternative to convert the biomass into useful products and energy [5–7]. The total characterization of the products from the pyrolysis of biomass is one of the great challenges in this field, mainly due to their molecular complexity [8].

Pyrolysis is a process that causes degradation of biomass in a non-oxidative atmosphere, at relatively high temperatures, producing a solid residue rich in carbon and mineral matter, gases and bio-oil [6, 7, 9, 10]. The yield and properties of the products depend on the nature of the biomass and the type of the pyrolysis process (type of reactor, temperature, gas flow, catalyst) [7, 11–13].

One of the main products of biomass pyrolysis is the bio-oil and, due to its high molecular complexity, it has been subject of several characterization studies in order to identify its compounds and indicate their best uses [8, 10, 14, 15].

This chapter describes the principles of the techniques and main application of chromatographic methods (GC, LC, GC × GC, LC × LC and Nano-LC) in the analysis of bio-oils derived from thermo-degradation of biomasses. Especially, attention is done to two-dimensional techniques that represent the state of the art in terms of separation, sensibility, selectivity and velocity of data acquisition for characterization of complex organic mixtures.

2. Pyrolysis of biomass

Energy production in the twentieth century was dominated by fossil fuels (coal, oil and gas) that represented, still in the beginning of the century, about 80% of all energy produced in the world. Nuclear power, hydropower and renewable energy sources (solar, wind, geothermal and small hydropower plants), which are the most attractive from an environmental point of view, represented only 1.5% of world production in those years [1]. Currently, the depletion of natural resources of coal and oil, together with the greenhouse effect, has attracted great interest for the production of sustainable energy [2]. The search for alternative fuels has led some countries to opt for biofuels, which in turn increased interest in energy from biomass [3].

The conversion of biomass into fuels and chemicals can economize fossil reserves and thus boost research, social and economic activities, especially in countries where the biomass is an abundant raw material in the agro-industrial sector [4]. The use of renewable sources for obtaining chemicals such as bio-plastics, bio-fertilizers and bio-polyesters can help reducing the demand of insumes from petrochemical origin.

The biomass can be converted into energy using thermal, biological, mechanical or physical processes. The biological processing (biological catalysis) is very selective and produces a discrete number of products in high yield, but it requires a raw material containing sugar or carbohydrates and a water content exceeding 40% [5]. The thermochemical methods are more suited to dry biomass (moisture content <10%) and rich in lignin (which is less suitable for biochemical conversion, since it is hardly broken through enzymatic activity), such as wood and agricultural waste. Furthermore, the thermal conversion process provides often multiple products in a short reaction time, typically using inorganic catalyst to improve the quality of the product. The main processes used for the thermal conversion of biomass into a more useful form of energy are combustion, gasification and pyrolysis [5, 6].

Pyrolysis is defined as a thermal decomposition of the biomass that occurs in the absence of oxygen at temperatures between 350 and 700°C, producing gases, liquid and solid products [5, 7]. Processes using lower temperatures and long residence time favor the production of charcoal, while processes using higher temperatures and long residence time increase the conversion of biomass into gas. A great production of liquids is favored using processes at moderate temperature and short residence time [9, 10]. It should be noted that the three products are always generated, but the proportions may be varied by adjusting the process parameters such as heating rate, gas flow, pressure, particle size of biomass and residence time of the biomass in the reactor [6, 11]. Also in accordance with these parameters, the pyrolysis can be classified as slow, fast, flash, catalytic or vacuum [12, 13].

The slow pyrolysis, or carbonization, employs low temperatures and long residence times, favoring the production of charcoal. In the case of fast pyrolysis, the heating occurs at higher rates (above 20°C min^{-1}), while in the flash pyrolysis, heating rates are between 500 and 1000°C s^{-1} [14].

Fast pyrolysis produces 60–75% by weight of bio-oil (liquid) 15–25% by weight of biochar (solid) and 10–20% by weight of noncondensable gases, depending on conditions and the biomass used. The solid residue from this process can be charcoal or ash only, depending on the final temperature and on the content of mineral matter originally present in the biomass used. This residue can be used as fuel or as a soil additive or still for the production of ceramic material. The gas produced can be recycled back to the process and facilitating cleavage reactions of the original biomass [14]. The main objective of fast pyrolysis is to prevent the breakdown of primary products in small noncondensable gas molecules, or even prevent them from being recombined and polymerized. In any of these cases, it obtains a smaller yield of bio-oil [10].

The order of the reactions that occur during the pyrolysis process and the yield of obtained compounds will depend on parameters such as the heating rate, the pyrolysis temperature, pretreatment of biomass and catalyst effects. The study of these reactions and the effect of these parameters are important for obtaining high yields of the desired products [5]. To perform the pyrolysis, it can be used different types of reactors, according to the main purpose of the process.

2.1. Pyrolysis reactors

2.1.1. Fixed bed reactors (FxBR)

In this type of pyrolysis reactor, the fluid phase (gas) passes through the particulate solid phase (biomass). This FxBR aims to promote intimate contact between the phases involved in the process—the gaseous fluid phase with the stationary phase (particles of biomass) [15–22]. The FxBR is constituted by tubular structures of stainless steel or quartz, being used as an oven or grill support during the controlled heating of the system. The inert gas passes through the compartment with the biomass (in steady state) carrying the products out of the reaction bed during the pyrolysis [15]. Typically, units for feeding biomass, ash removal and outlet or collecting gases are added to these reactors. Such reactors operate with high residence time of the biomass in the oven and low inert gas velocities [16–22]. In this way, they are considered suitable for research in laboratory scale or bench. Some units operating in China can convert up to 600 kg/h of biomass to bio-oil [23].

Pyrolysis in FxBR is very used to the slow pyrolysis, with the main purpose of producing coal or ashes.

2.1.2. Fluidized bed reactors (FzBR)

In this type of reactor, a fluid (gas) is passed through a granular solid material at high speed, sufficient to suspend the solid material and cause it to behave like a fluid. This process known as fluidization provides extensive use in studies on the pyrolysis of biomass and is ideal for the technique of fast pyrolysis because it can achieve the necessary requirements for its realization and is virtually the only ones used in the world on a commercial scale for the production of biofuels and chemicals [24–27].

The FzBR operates with suspended solids by the action of rising gases, which are introduced from the bottom of the reactor. To promote more effective heat transfer, there is used a bed of solid particles, generally consisting of sand finely dispersed in the biomass itself [23, 28].

The rapid exchange of heat between the heat source and the biomass is one of the most important points in the pyrolytic process. In this context, in FzBR, the dried and comminuted biomass is introduced into the reactor, wherein, in the heating zone, the material remains in a continuous movement, promoted by the carrier gas flow (inert gas), which maintaining the reactor with a low oxygen content as it is heated to high temperatures (500–900°C) using a heating rate between 100 and 500°C min^{-1} [23, 29, 30].

Among the many advantages of this model, we can mention its ease of construction and operation, good temperature control, the operation at atmospheric pressure, the easy scale-up and operation possibility with fine particle size biomass, which is common in agriculture, in forestry and industry. Beside this, the reactor has an excellent heat exchange between the heat source and the biomass, and an efficient gas-solid contact due to the movement of the particle bed. This effective contact simulates an isotherm condition, which implies an operation more secure and with optimum performance, generating yields of liquid products of approximately 70–75% by weight (dry basis), minimizing side reactions [24, 26].

But, the heterogeneity of the residence times in the reactor due to the agitation of the solids in the bed can compromise the uniformity of the products. Moreover, the wear caused by the moving particles and agglomeration of ash produced with the inert bed material may lead to loss of fluidization and therefore the pyrolytic process [23, 25, 28]. Therefore, the successful application of this technology depends on understanding and overcoming their disadvantages and thus the development of a reactor that meets the needs of the pyrolysis process in a whole.

Several research groups in the development of fast pyrolysis use the fluidized bed reactor. Among the many examples, one can cite: the Union Fenosa located in Meirama, Spain, which has a pilot plant with biomass feed capacity of approximately 250 kg h^{-1}. Still in Europe, the Wellman Company in the UK, which has a pilot plant with a capacity of 200 kg h^{-1}, in Canada, the DynaMotive, which employs a pilot plant with capacity of 8000 kg h^{-1} [23, 24].

2.1.3. Continuously feeding reactors (CFRs)

Continuously feeding reactors (CFRs) are those which operate at all times with an input a specific substrate(s) and output of product(s). These types of reactors are widely used in industrial scale [31].

In general, CFRs offer reduced fixed and operating costs and improve heat exchange capability [31, 32]. Furthermore, they provide an increase in quality of the final product, since the variations that exist between batches are eliminated [33]. Continuous processes are still able to reduce losses caused by operational problems during the process, and it is not necessary to interrupt the production line [33] (start-up and shutdown). The applications of these reactors are aimed to minimize the difficulties encountered in the process on an industrial level using reactors in pilot scale [31].

2.2. Catalytic pyrolysis

Studies of the composition of bio-oils obtained by pyrolysis of various biomasses (rice husk, coconut husk fiber, core tropical fruits, straw sugarcane, wood residues, etc.) found that the volatile fractions of bio-oils consist of a complex mixture of different classes of compounds such as, ketones, phenols, aldehydes, hydrocarbons [8, 15, 34–40]. These bio-oils have physicochemical characteristics that avoid their direct use as fuel, without any treatment. One of the reasons is its high oxygen content, thus leading to a high chemical instability during storage hindering their production on an industrial scale [41].

For industrial use of bio-oil, some enhancement process is needed (upgrading). The catalytic pyrolysis has emerged as an alternative for improving the quality of liquid products of pyrolysis acting as an upgrading process of bio-oil. The main variable of this process is the type of catalyst used, in particular those able to considerably reduce its oxygen content [41].

A method used worldwide is the hydrodeoxygenation (HDO), which converts and fragments heavy molecules in the biomass into hydrocarbons with lower oxygen content by the catalytic addition of hydrogen [42]. This process has been studied with the aim of producing a liquid mixture of hydrocarbons that do not have the undesirable properties of bio-oil and thus can be used as fuel. This method is considered the most efficient for the upgrading of bio-oils [42].

Several catalysts can be applied for HDO, being necessary the use of high temperatures and pressures, under an atmosphere of H_2. The most used are those consisting of cobalt and nickel and may be supported on alumina (Al_2O_3), silica (SiO_2), carbon, zeolites (ZSM-5), among others [43, 44].

The H-ZSM-5 catalysts, for example, have strongly acidic active sites, which can supply hydrogen for the pyrolytic reactions, favoring the deoxygenation of bio-oil. Thus, the great advantage of the use of zeolites for production of bio-oils is the possibility of working on hydrogen-free atmosphere and the use of ambient pressure during the catalytic pyrolysis process [45].

Zeolites have been shown to be highly effective for converting lignocellulosic biomass into aromatics through catalytic pyrolysis. Whereas this type of biomass has low amounts of hydrogen in their composition (low H/C ratio), the maximum yield of hydrocarbons that can be obtained in the absence of H_2 as external reagent is limited. Thus, an upgrading using zeolites generally results in yields of aromatic hydrocarbons and olefinic near 30% [46, 47]. In addition, in the catalytic pyrolysis occurs greater coke formation when compared with conventional pyrolysis [48].

Catalysts conventional hydrotreating as Co-Mo and Ni-Mo and supported noble metal catalysts have been studied to produce stable products with high calorific value from pyrolysis [49], however, require high pressures of H_2 that lead to hydrogenation of the aromatic ring, resulting in reduction in calorific value and increase in H_2 consumption [50].

2.2.1. Catalytical pyrolysis in-situ versus ex-situ

Depending on the method of contact of catalyst and vapors of the pyrolysis, catalytic pyrolysis can be classified as in situ or ex situ.

In the in situ catalytic pyrolysis, the catalyst is mixed with the biomass to be pyrolyzed. Thus, the most suitable type of reactor for this is the fluidized bed reactor since this biomass is directly mixed with the catalyst. As the catalyst is exposed to a concentrated stream of vapors generated by depolymerization of the biomass components, the catalysis reactions are facilitated [51–54].

For the ex situ catalytic pyrolysis, biomass is pyrolyzed in a separate compartment of the catalyst. The pyrolysis vapors generated in the first compartment are diluted in an inert gas which is inserted between the two compartments being transported into a second compartment which is filled with the catalyst. As the second gas flow between the compartments, the contact time with the catalyst decreases [51]. An advantage of this technique is the use of different temperatures for both the pyrolysis reactor and for the catalysis reactor, thus allowing the use of catalysts which are sensitive to high temperatures [51–55].

2.3. Microwave assisted pyrolysis (MWAP)

The different pyrolysis processes have various conventional and unconventional heating methods. The development of an efficient heating method, with a precise control of heating

parameters and with a reduction of adverse effects on the quality of the product is one of the challenges to be overcome in the development of efficient pyrolytic processes [56]. The use of microwave can be an efficient way of heating the biomass in a thermochemical conversion processes. Tech-En Ltd (UK) did the first study of the use of microwave pyrolysis in early 1990 [57, 58]. This is a technology which can be a very effective alternative for pyrolysis of biomass, presenting several advantages such as reduction in waste volume, fast heating, better chemical reactivity, ease of control, energy saving, overall cost-effectiveness, portability of equipment and processes, a cleaner source of energy compared to conventional systems [59].

Microwave heating allows a more careful control of the parameters of the pyrolysis process, enabling the maximization of the production of liquids or gases, once these parameters may induce or modify specific chemical reactions resulting in different product profiles. The process can be modulated, aiming the product optimization in accordance with conditions of temperature, power and residence time used [60].

The biggest advantage in the use of microwave heating as compared with the conventional process is the significant reduction in temperature and consequent energy gain in the pyrolysis process [61].

Another important aspect of the heating by microwave is capability to obtain basically the volatile organic compounds at lower temperatures when compared to conventional heating. Moreover, obtaining bio-oil and gases is nearly synchronized. This heat and mass transfer characteristics of the process are related to the selective heating of the components to absorb the microwave with more intensity [60].

Obtaining a greater heating uniformity in the process may be possible if the temperature is homogenized at some point during the process, so that, without this, different product compositions are obtained due to the temperature profile formed [60].

Furthermore, the temperature selection depends on the desired product. Processes at low temperatures provide greater bio-oil yields and lower energy cost. The determination and use of the appropriate power to microwave are also important. Lower powers, with lower heating rates favor formation of biochar, whereas higher power with higher heating rates favors the gasification reaction. In both cases, there is a reduction in the yield of bio-oil [60, 61].

The use of microwaves also has some challenges for the applying in the pyrolysis such as the need for different systems for measuring the temperature [62], the limitation of usable materials for propagation of microwave [63] and obtaining heating equipment compatible with the process and with an efficient scale [64].
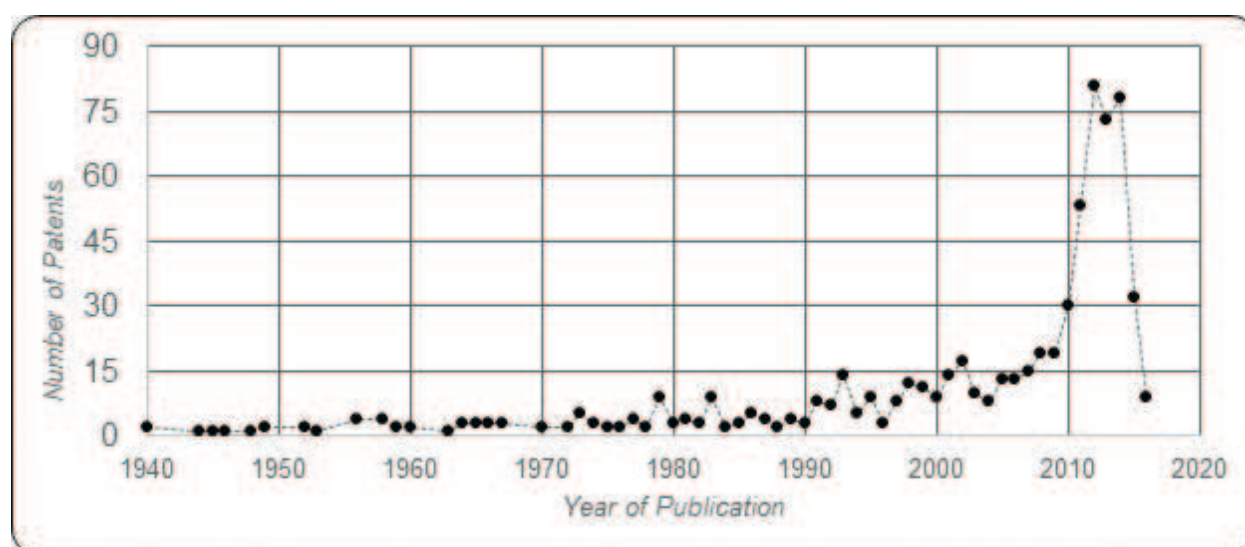
2.4. Pilot plants of pyrolysis

The pilot plant consists of many components (steps/processes) that together form a unique process, which enables testing technologies to evaluate the quantity and quality of products desired [65, 66]. According to RESEM®, a Sino-American Company specialized in equipment for pyrolysis [61], there are different sizes of pilot plants, reactors differentiated according to the type and number of samples to be processed or products to be produced [67, 68].

The development of new technologies for the production from clean energy sources [69, 70], are associated with the emergence of pilot plants for the production of bio-oil through pyrolysis of biomass in both, laboratory and industrial scale, mainly because it is a simple, reproductive and fast process [71].

Pilot plants are equipments that consist of a closed or open system with physical, chemical and thermodynamic operation, in order to perform a technological process on a small scale. So, a prototype, designed for industrial processes, can be installed either in research laboratories or in industries [66]. In these prototypes, new and different technologies, shapes, sizes and physical structures for generating and processing information for use in pyrolytic systems can be tested before the scale-up [67].

According to **Figure 1** that shows one graphic with the research scenario related to global patent on the theme in pyrolysis plant, from 1940 to this year, they were deposited 673 patents worldwide [72, 73].

Figure 1. Distribution of patents related to pyrolysis pilot plants in the last 80 years (according https://patentes.google.com and https://patentscope.wipo.int [72, 73]).

Few of these patents actually generated commercial pyrolysis plant, indicating the need a lot of investment in this regard, especially in countries with great potential for use of biomass and lower oil reserves. As already described, in recent decades, there has been a growing concern about the processing of biomass through pyrolysis due to the interest in their products, both biochar [74], as bio-oil. In this sense, grow-related searches to biomass conversion technologies studies from laboratory scale to the development of bio-refinery [75] involving processes for the production of fuels and chemicals [76, 77].

The experimental setup on a pilot scale is based on the adjustment process through the system variables, and especially in the reactor used technology.

The reactors in pilot plants can be classified in two systems [78]:

- Batch system: The biomass is loaded at the beginning and from there it collects the products and the flow is not continuous.

- Continuous system: The biomass remains in continuous flow and the products generated are also continuously collected.

With the technological advancement of pyrolysis technique, some models of reactors have been exploited to optimize the process, cost and quality of the products generated, and the main ones are the fluidized bed (bubbling and circulating), in addition to these, some others can be found like fixed bed, ablative, vacuum, rotary cone, plasma, microwave and solar, among others [79–99].

2.4.1. Fixed bed pilot reactor

The pilot fixed bed system, similar to the bench reactors (Section 2.1.1), consists in fixed reactor with an inert carrier gas, similar to the bench scale. The technology of the fixed bed reactor is considered simple, reliable [100], and efficient for the production of bio-oil in the pilot plant. In these reactors, the solids move down and collide in counter current with a heated inert gas. They are used in small-scale productions [101]. The cooling and cleaning gas system consist in a filtration and in the use of separators cyclones. The biggest problem is associated with the removal of bio-oil and losses involved in this process.

The fixed-bed pilot plants developed from the late 1980s, with a final capacity of 5 tons per day are considered small, from the point of view of the transfer for industry, considering that these technologies are still in development stages for commercial applications.

2.4.2. Fluidized bed pilot reactor

As mentioned in Section 2.1.2, in the pilot fluidized bed reactor, a mixture of fluid and solid is obtained by introducing a pressurized fluid through the solid particles with smaller diameter [102, 103]. The general scheme of this type of reactor is very similar to that used in fluidized bed in bench scale.

Two main types of fluidized bed reactors are used in pilot plant:

Bubbling fluidized bed reactor: According to the literature [104], the bubbling bed reactors are simple to construct and operate. They provide a better ability to control the temperature, fluid-solid contact, heat transfer and capacity of storage of solids. Heated sand is used as a bed solid phase, rapidly heating the biomass in an oxygen-absent atmosphere. The biomass is decomposed into coal, steam, gas and aerosols. The fluidized gas stream entrains the compounds produced out of the reactor [105].

After the pyrolytic reaction, biochar is removed by a cyclone separator and stored. An important factor for the full operation of these reactors is that the biomass needs to be in small particles (less than 2–3 mm) to achieve high heating rates in the oven.

Circulating fluidized bed reactor: It has similar characteristics to the reactor in bubbling fluidized bed except for the shorter residence time. This design results in higher speed and higher yield of bio-oil compared to fluidized bed reactors [106]. The reactor moves around its main axis. One advantage is its high performance even with a more complex hydrodynamics.

2.5. Bio-oil from pyrolysis of biomass

The bio-oil, or pyrolysis oil, is a dark brown color liquid with a characteristic smell and comprising a complex mixture of hydrocarbons and oxygenated compounds with an appreciable amount of water, originated from the natural moisture of the biomass as well as a product of reactions that occurred during the pyrolysis process [26, 41, 107].

The literature registers that the bio-oil can contain more than 400 different chemical compounds from different chemical classes, varying among organic acids, aldehydes, ketones, alcohols, esters, furans, sugar derivatives, phenols, among others [108, 109]. In addition to the oxygenated compounds, many aliphatic and aromatic hydrocarbons can be found [8].

The anhydrous-sugar levoglucosan (1, 6-anhydro-β-ᴅ-glucopyranose) is the main component of bio-oil, being derived from the thermal depolymerization of the cellulose and from it, many other sugar derivatives can be formed. The yield of these anhydrous-sugars is affected by the source of biomass and by the experimental pyrolysis conditions. The increase in pyrolysis temperature reduces the concentration of levoglucosan, in contrast to other products because it stimulates breakdown of this molecule [110].

Mixtures of compounds as phenols, cresols and catechols (monomers and oligomers) are formed from the lignin [111, 112]. Phenolic compounds found in bio-oils are composed mainly of simple phenols with a hydroxyl and which may contain other substituent group on the benzene ring, forming mixed functions (carbonyl, carboxyl, alkyl or aryl radical) [108].

Due to the presence of acids, especially acetic and formic, bio-oil may show pH values in the range 2–4 [41, 113] that constitutes a problem, since it will affect storage conditions (equipment, transport) and its processing [113, 114].

The oxygen content in bio-oil is approximately 35–40% by weight. The specific composition depends mainly on the type of biomass used, the pyrolysis conditions (temperature, residence time and heating rate) and the storage conditions of bio-oil [10, 41, 113]. The high oxygen concentration results in a low energy density that is less than 50% of the value for conventional oils. It also affects the bio-oil miscibility with other petroleum fuels and the stability of bio-oil [41, 114].

The water constitutes 10–30% bio-oil, and their quantity depends on the original biomass, and the pyrolysis conditions, since the moisture is coming from biomass and also from dehydration reactions taking place during pyrolysis [41, 108, 113]. Depending on the feedstock and process conditions, the ratio of aqueous and oil phase can vary from 50:50 to 30:70, and the presence of two phases can hinder the application of bio-oil. This high water content may cause problems in the ignition engines by reducing the rate of vaporization of the oil, which prevents its application directly as fuel [115]. In many instances, drying the biomass prior to the pyrolysis is sufficient to reduce this problem.

The instability of bio-oil is mainly due to the presence of highly reactive organic compounds (ketones, aldehydes, organic acids), which can undergo reactions to form ethers, acetals or hemiacetals [41, 116]. This kind of reactions may increase the average molecular weight oil, water content and its viscosity, resulting in a low quality oil and that, when stored, results in phase separation. However, the addition of polar solvents such as methanol or acetone can significantly reduce the viscosity of the bio-oil [41].

The ash content of the bio-oil can also cause problems in some applications. The composition of the ash is dominated by alkali metals (potassium and sodium) that are responsible for severe corrosion and deposition turbines on heating surfaces during combustion [115, 117].

However, the crude bio-oil, before use as fuel, must be chemically modified through complex processes such as hydrodeoxygenation, hydrocracking, decarbonylation or decarboxylation to reduce oxygen content, which is the main unwanted constituent in the bio-oil for energy purposes [113]. Another alternative improving (upgrading) of the bio-oil is catalytic pyrolysis using zeolites alumina or metals as catalysts [114].

For the use and recovery of chemicals from bio-oil can be used conventional separation techniques, such as solvent extraction, column chromatography and distillation (single, fractionated, or under vacuum). Solvents commonly used for extraction of compounds of interest in bio-oil include water, alcohols, ethyl acetate, hydrocarbons such as toluene and mixtures thereof [108].

Fractionation in open or pressurized column has also been used as a pretreatment for the separation of compounds from bio-oil [118–120].

Among its uses, bio-oils are potential fuels to diesel engines, gas turbines and boilers. They can be used also as raw materials for obtaining hydrocarbons by catalytic conversion or hydrotreating [121]. Considering its phenolic fraction, bio-oil appears as a substituent for petrochemical phenol in the production of phenolic resins (phenol-formaldehyde) or can be used in pharmaceutical, food or fine chemicals industries [108, 122]. Furthermore, bio-oil may be fractionated to obtain many other products of commercial interest, such as abrasives, filter elements, battery separators, electric components, refractory materials, adhesives for wood, paints, varnishes, enamels, etc. [8, 123].

The reaction of bio-oil with ammonia, urea or other amino compounds produces amides and amines stable, nontoxic to plants and can be used as organic fertilizer. The bio-oils derived from wood residues can be commercially applied in smoking food [10]. In case of bio-oils with high concentrations of hydrolyzable sugars, it may be favorable to the production of ethanol by fermentation, whereas bio-oils with high phenol content are mentioned as attractive starting material for the production of adhesive [123].

As application examples of some of the most important bio-oil compounds can be mentioned: the levoglucosan (food additive, pharmaceutical industry); the levoglucosenone (synthesis of antibiotics and rare sugars); furfural (pharmaceuticals, pesticides); acetic acid (chemical industry); formic acid (wood preservatives, antibacterial agents); and hydroxyacetaldehyde (pharmaceuticals, fragrances) [124].

2.5.1. Aqueous phase of bio-oil

As mentioned above, the amount of aqueous phase in the bio-oil will depend on the original biomass composition, its original moisture and the pyrolysis conditions [108, 113]. However, it cannot be removed by conventional methods such as distillation [10]. The phase separation will occur when the amount of water exceeds the maximum level in bio-oil (usually above 30–45%) or by extraction methods [125].

The addition of water allows to readily separating the bio-oil into organic and aqueous phase. The aqueous phase contains mainly components of higher polarity, such as levoglucosan and other anhydrous-sugars, furan, furfural, organic acids of low molecular weight, hydroxyacetone, hydroxyacetaldehyde and guaiacol [126–128]. The separation of the aqueous phase of the bio-oil is also commonly performed using dichloromethane and sodium bicarbonate solution, obtaining an acid extract [129, 130]. Although solvent extraction is widely used in phase separation of bio-oil, it can affect the qualitative and quantitative composition of the extracted sample due to the different affinity of the solvent for each class of chemical compounds present in the sample [131].

The aqueous phase of the bio-oil cannot be directly discarded as wastewater, since some compounds may be above discharge limits. Different processes may be applied for the treatment of wastewater together with the pyrolysis process or subsequently in a wastewater treatment plant [132, 133].

Currently, several studies have been conducted for the treatment of aqueous phase and the application of its compounds as industrial raw material. Upgrading processes such as the hydrodeoxygenation and moderate catalytic cracking allow the production of hydrogen, hydrocarbons, alcohols and olefins from the aqueous phase [127, 130, 134].

Due to high amount of oxygenated compounds from C2 to C6 such as aldehydes, ketones, acids, and carbohydrates, several gasoline additives, alcohols and diols can be produced from the aqueous phase by hydrogenation processes [135, 136]. Light alkanes C1 to C6 and liquid alkanes C7 to C15 may also be produced from the aqueous phase carbohydrates through upgrading processes as dehydration and hydrogenation [137].

The sugars present in the aqueous phase (levoglucosan, pentoses and hexoses) are recognized as key compounds for the production of furan derivatives, such as furfural and 5-hydroxymethylfurfural [126]. The conversion of sugars into furan derivatives can improve the economic perspective for using the aqueous phase as source of raw material for a wide variety of chemicals [124, 126].

The acidity of the aqueous phase, as already mentioned [127], can cause corrosion in equipments (based on carbon steel); however, these organic acids may also be valuable byproducts that can be used in industry as solvents and wood preservatives [124, 127].

2.6. Analytical techniques applied to bio-oil and aqueous phase

The full chemical characterization of pyrolysis oils is very complex, mainly because they are formed by the degradation of carbohydrates and lignin, that have an abundant content of water and a great variety of organic functions associated a small amount of inorganic material [138].

The composition of bio-oils can be divided into four distinct fractions: moderately polar monomers, detectables by GC (40%); polar monomers directly detectables by HPLC or by GC after derivatization (12%); water (28%); lignin and pyrolytic materials (20%) [139] not detectables by GC.

Gas chromatography is the most widely used technique in chemical analysis of bio-oils and other complex mixtures. Despite its wide application, the GC [7, 107, 110, 111] has some limitations when applied to mixtures with a great variety of compounds and different concentration ranges. Co-elution is a major impediment for the separation and unambiguous identification of a compound. In this sense, there was developed the comprehensive two-dimensional gas chromatography (GC × GC) which is being applied to this kind of sample [8, 15, 119, 140], which greatly reduces the number of co-elutions in a chromatogram, through two dimensions separations.

Known since the 90s, the GC × GC is an analytical tool that differs from other techniques due to the sequential use of two chromatographic columns, which allows a significant increase in selectivity, favoring the structuring of the peaks in the chromatographic space. Regarding the dimensional gas chromatography, GC × GC shows most significant increase in the sensitivity and resolution, allowing a higher peak capacity, that is, a higher number of peaks separated and identified [141, 142].

Studies employing liquid chromatography for characterization of bio-oils have also been recently undertaken [143–149]. In view of the complexity of the samples, the comprehensive two-dimensional liquid chromatography (LC × LC) [150] in the same way as it happens for GC × GC becomes an important tool for characterizing bio-oils, because there is a large increase in the resolving power when compared to one-dimensional methods. Another great alternative is the use of micro HPLC columns that allow very small mobile phase flows, facilitating the coupling to more efficient detectors than conventional ones [151–153].

3. Gas chromatographic methods applied to the analysis of bio-oils

3.1. One-dimensional gas chromatography (1D-GC)

One-dimensional gas chromatography (1D-GC) coupled to mass spectrometry (GC/MS) or flame ionization detector (GC-FID) is an important analytical tool that provides chemical profile information of bio-oil, aiming its correct destination as fuel or in the chemical industry. There are several studies in the literature about the use of 1D-GC to chemical characterization of bio-oils from pyrolysis of various biomasses [12, 154, 155]. The large Brazilian biodiversity contributes to many options of biomasses, significantly increasing the total number of identified chemical compounds and the potential use of these materials. There are several studies using 1D-GC to analysis of bio-oil from Brazilian biomasses such as straw and sugarcane bagasse (*Saccharum officinarum*) [38, 70, 156], eucalyptus sawdust (*Eucalyptus globulus*) [70, 156], Amazon tucumã (*Astrocaryum aculeatum*) [157], mangaba seed (*Hancornia speciosa*) [39], coconut fiber (*Cocos nucifera*) [34], fruit of palm (*Arecaceae*) and pine wood (*Pinus*) [158], among others. A large part of this work has employed the GC/MS to identify families and major compounds, usually containing oxygen in its constitution, as phenols, furans, alcohols, ketones, aldehydes, esters and carboxylic acids, regardless of the biomass used. As an illustration of this type of analysis, **Figure 2**

shows the chromatogram of a sample of sugarcane straw bio-oil using the GC/MS system with DB-5 column (60 m × 0.25 mm × 0.25 μm), developed in our laboratory. In this work, 208 and 336 compounds were detected in the analyzed bio-oil sample directly and by SPME with PDMS fiber, respectively. Among these, 33 and 35 compounds were identified in two cases.
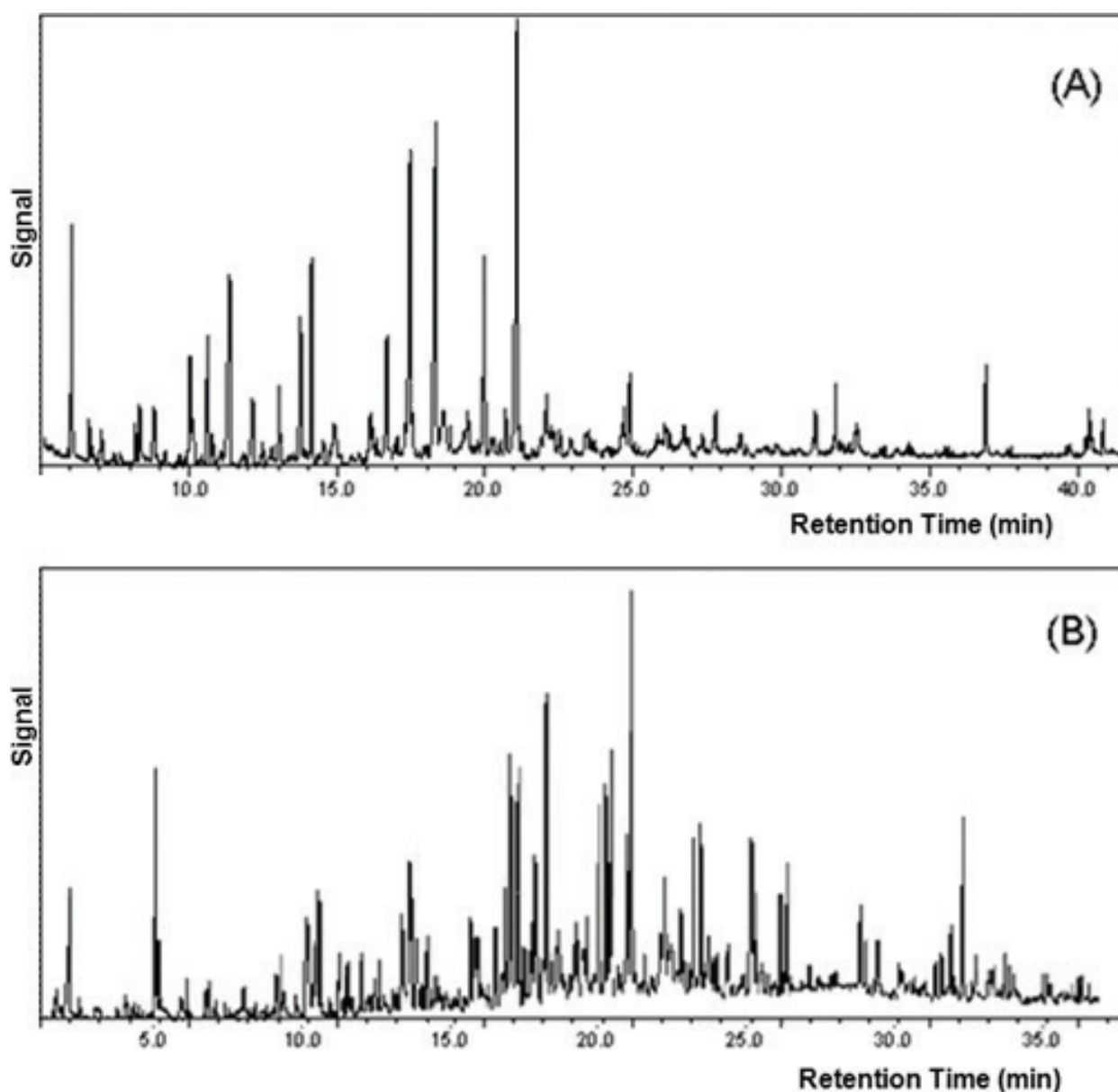
Figure 2. Total ion chromatogram (GC/MS) for bio-oil (A) and SPME (B) of the pyrolysis of straw sugarcane. Chromatographic Conditions: DB-5 column (60 m × 0.25 mm × 0.25 μm); oven temperature: initial oven temperature was 40°C, hold for 2 min, heating to 280°C at a 5°C/min, where it stayed for 2 min.

In general, phenolics compounds are the most family in the majority of bio-oils. Phenols are widely used in fine chemical industry, food processing, pharmaceutical and production of phenolics resins [34, 108, 158].

Hydrocarbons (saturated, unsaturated and aromatic) are also identified by GC/MS in most bio-oils; however, their percentage is very small compared to other components, with few

exceptions. In the study of Santos et al. [39] were identified various hydrocarbons (saturated, unsaturated and aromatic) through GC/MS in the bio-oil from pyrolysis of mangaba seed (*Hancornia speciosa*). The total relative area of peaks which is one of the ways used to estimate the quantitative composition of bio-oil ranged from 8.1 to 13.8% of total compounds tentatively identified in this bio-oil. In this same work, carboxylic acids were the family of major compounds found (from 72.5 to 84.4%). Carboxylic acids, although lower quality becomes the bio-oil, are possible hydrocarbon precursors, enabling further study of the bio-oil upgrading.

Patel et al. [54] characterized by GC/MS the crude bio-oil from sugarcane bagasse produced by fast pyrolysis in a fluidized bed and evaluated the efficiency of Mo_2C/Al_2O_3 catalyst in the deoxygenation and quality of bio-oil. The catalyst resulted in an increase in the content of phenolics and furans, which arouse great industrial interest.

Spilã et al. [159] developed a method for separation and analysis of the aqueous fraction of bio-oil by adding water to raw bio-oil followed by extraction with ion exchange resin and ethyl ether. The ether soluble compounds were analyzed by GC/MS, since the insoluble fraction was evaporated and solubilized in methanol for analysis (GC/MS, CHN and pyrolysis-GC/MS). The characterization method was applied to bio-oils derived from wood, scots pine and wheat straw. Further studies have applied the same separation method to different biomass such as forest waste [160] and wood [161] with subsequent chromatographic analysis (GC/MS and GC-FID).

Wiggers et al. [162] performed a study on pyrolysis of soybean oil at pilot scale, in continuous system aiming higher yield of bio-oil. The authors conducted a prior distillation to separate light bio-oil fractions (LBO) and heavy bio-oil fractions (HBO). Various hydrocarbons were found in bio-oil and benzene, toluene, ethylbenzene, p-xylene, o-xylene and linear hydrocarbons (C_7 to C_{12}) being the majority for LBO; while for HBO the majority were fatty acids, toluene, ethylbenzene and linear hydrocarbons (C_8 to C_{17}).

Owing to the industrial importance of phenols and furfural, the author highlights the refining of the aqueous phase can extract these aromatic compounds of high value to the industry.

GC/MS can be also coupled directly to one pyrolyzer system since it is required to just check the potential of the chosen biomass to generate bio-oil, since the amount of sample used in this case is very short not allowing further analysis with the bio-oil produced. This technique is known as analytic pyrolysis and represented by Py-GC/MS.

3.2. Pyrolysis-gas chromatography/mass spectrometry (Py-GC/MS)

Analytical pyrolysis (Py-GC/MS) is an important characterization tool of the products generated by the pyrolysis of lignocellulosic material, in addition to allowing a better understanding of the function and effects of catalysts for the production of hydrocarbons or other desirable compounds prior to the pyrolysis in analytical laboratory scale. Direct analysis of condensable gases is held in a pyrolyzer coupled to a gas chromatograph with mass spectrometry detection (Py-GC/MS). In the pyrolyzer, a small amount of biomass is subjected to a heat treatment and condensable gases are simultaneously separated and identified by their mass spectra and retention times [163–165].

The Py-GC/MS have advantages, such as the use of small quantities of biomass, which allows the realization of several pyrolysis under various operating conditions (ratio catalyst/biomass, temperature, heating rate, etc.), in a simple and fast way [166, 167]. Through Py-GC/MS can also predict what kinds of industrial interest compounds are produced during the process, which the effect of catalysts, and if inhibition occurs, etc.

Liaw et al. [165] studied the optimal pyrolysis temperature for two standards cellulose and two different types of wood by Py-GC/MS, and yields variations were evaluated by PCA (principal component analysis). The PCA showed that pyrolysis products can be divided into groups and are strongly influenced by the nature of the raw material studied. However, the levoglucosan represents those few compounds that are influenced by temperature because as the temperature increases, its concentration decreases.

Barbosa et al. [168] developed method of determining the ratio between syringyl and guaiacyl content (S/G) in lignins from eucalyptus wood by Py-GC/MS using different markers of syringyl and guaiacyl units. Traditional methods, which involve chemical degradation, generally are laborious, time consuming and require large sample amounts. However, the method developed by the authors is fast, uses very small amount of sample and highly sensitive.

The analysis of macauba fruit (*Acrocomia sclerocarpa M.*) [169] by Py-GC/MS revealed the formation of alkanes, alkenes, dienes and cycloalkanes, being the main products the 2-propenal and acrolein (triglycerides derived). In the bio-oil from the oil pulp, most of the compounds found (71–79%) were aldehydes, cycloalkanes, alkenes and dienes. Through this method, it was possible to observe the differences in composition of biomass and its influence on the quality of bio-oil formed.

Py-GC/MS enables understanding of formation of fatty acids obtained by pyrolysis of seed and babassu oil, studying their composition and fragmentation mechanisms of the compounds produced by thermal degradation of the samples [170].

The evaluation of the use of different catalysts (ZnO, CaO, $ZnCl_2$ and $MgCl_2$) to obtain bio-oil from several biomasses [171–173] was also assessed by Py-GC/MS.

Despite numerous studies reported in the literature, 1D-GC has limitations when applied to the characterization of complex sample, such as low resolution and co-elution, which lead to incorrect identification of some analytes [112, 174, 175]. A wide range of chemical classes present in the matrix of bio-oil also complicates the choice of chromatographic columns proper polarity at all [165]. Therefore, currently, most jobs that show a characterization of bio-oil by one-dimensional chromatography also make use of the characterization by chromatography comprehensive two-dimensional gas (GC × GC).

3.3. Comprehensive two-dimensional gas chromatography (GC × GC)

The comprehensive two-dimensional gas chromatography (GC × GC) is a multi-dimensional chromatographic technique that emerged in the early 90s [176]. Due to its high separation power [177], GC × GC has been widely used use in the analyzes of pyrolytic bio-oils, once these are complex mixtures and their composition can present more than 400 organic compounds

belonging to a great number of distinct chemical classes. The bio-oil composition may vary according to the pyrolysis conditions and the kind of biomass chosen [178, 179].

This technique provides many advantages in relation to 1D-GC in the elucidation of the composition of complex samples [177]. Among these advantages one can be highlighted the increase in peak capacity, which leads to a better separation, not only between analytes, but also between them and the matrix. An increase in detectability, due to the narrow chromatographic bands resulting from the modulation, may also be considered another advantage.

Furthermore, the technique GC × GC compared to conventional gas chromatography also provides an increase in sensitivity and a generation of structured diagrams that facilitate the identification of unknown compounds [180–183].

The two-dimensional system consists of two chromatographic columns connected in series, one with a standard size (normally 30 or 60 m length and 0.25 mm internal diameter) and other shorter and with smaller diameter (~2 m length and 0.15 mm i.d.). A column set commonly referred as conventional consists of a nonpolar column in the first dimension (^{1}D) and a polar or with intermediate polarity in the second dimension (^{2}D). The two columns have different separation mechanisms (orthogonal separation), that is, the first column performs separation of compounds according to molecular weight or boiling point and second column by polarity, providing a major breakthrough in the separation of complex samples [141, 177, 184–188].

Figure 3 shows a GC × GC system where is possible to observe the modulator between both columns and some details of the peak processing. The modulators have the main function of to continuously collect fractions from the first column, re-concentered them and to inject in the second one [190–192]. This procedure is responsible for the increase in the signal-to-noise and the decreasing in detection limits, if compared to 1D-GC [188].

In the graphical representation of GC × GC, the register of the detector signal versus retention time is a continuous sequence of short chromatograms of each eluted fraction from the second column. After this, tridimensional graphics can be constructed considering the detector signal and the retention times in the first and second dimensions (^{1}t$_R$ and ^{2}t$_R$) [191, 192].

Data acquisition from 2D-GC can be better explained viewing **Figure 3b**. In this figure, one co-eluted peak corresponding to three analytes) eluted from the first dimension pass through the modulator being fractionated and eluted from the second column. The crude chromatogram originated corresponds to the sum of all the chromatograms obtained on ^{2}D. The second step (**Figure 2c** is the transformation (by and adequate software) of these data in a two-dimensional diagram (^{1}t$_R$ × ^{2}t$_R$). The last step is the tri-dimensional visualization of the results (**Figure 2c** that was also performed by adequate software [182].

There are two main kinds of commercial modulators: thermal and with valves [193–196]. The thermal modulators (as by heating or by freezing) present more utilization in GC × GC [193, 195]. The cryogenic modulators can use liquid nitrogen or liquid CO_2 and act as a cold trap for the analytes [196].
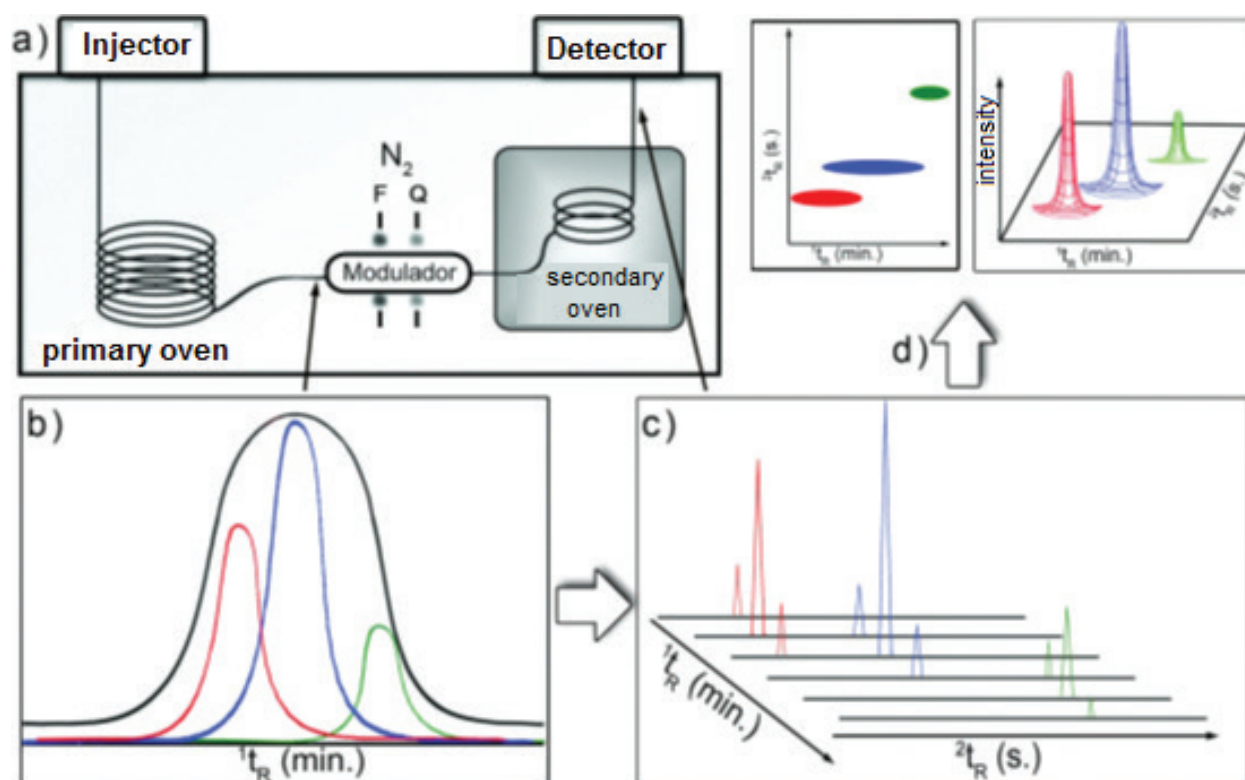
Figure 3. (a) GC × GC system; (b) three peaks co-eluted in the first dimension (^{1}D); (c) transformation of raw data into a two-dimensional chromatogram ($^1t_R × ^2t_R$); (d) reconstruction process of chromatographic peaks forming the color diagrams (two-dimensional and three-dimensional) [189].

GC × GC allows the utilization of several detectors, with few modifications to adapt them to the low volume and high velocity of data processing. One can be highlight the flame ionization detector (FID), time-of-flight mass spectrometry (TOF-MS) and quadrupole mass spectrometry (qMS) as the more used [197–203].

FID has been associated to GC × GC since the first works found in the literature, including the initial researches in the bio-oil characterization [204–207].

GC × GC promotes high speed separations, providing very narrow peaks, requiring detectors with equally rapid acquisition rates to get a sufficient number of points per peak [208, 209] to permit quantitation. The detector scan should be short and its internal volume has to be small [194]. The high acquisition rate of the FID (up to 200 Hz) [210] allied to a good response for almost all organic compounds and its good performance in quantitative analysis justifies the wide use of this detector in two-dimensional chromatographic analysis. The main difficulty in the FID employment is that it does not provide structural information about the separate peaks. Thus, detectors with mass analyzers gain space for identification and confirmation of the separate compounds.

The TOFMS is particularly effective for GC × GC, since it presents acquisition rates between 50 and 500 Hz [180, 182, 197–203], making it the preferred choice among researchers for these studies. The coupling of separation GC × GC with efficient detection TOFMS has an

additional advantage, which is its higher sensitivity than the full scan mode over conventional mass spectrometry detectors with quadrupole analyzer (qMS). Consequently, TOFMS outperforms the other detectors in qualitative and quantitative analysis. The disadvantages of systems of this type analyzer are its relatively high cost, the need for proper training and specific operating conditions for daily operation, especially due to its high sensitivity [191].

The GC × GC-qMS system is also showing its potential to analyze complex samples [211–213]. Initially, quadrupole mass spectrometers showed very low data acquisition rates (up to 20 Hz) which made them too slow to use in GC × GC systems [214]. However, decreasing the mass range investigated or monitoring only a few ions during the run, it is possible to obtain a higher scan rate [191]. This has been the subject of research of several research groups [180, 208, 214, 215], that is, to develop quadrupole mass analyzers faster and comparable to TOFMS. From this, it was introduced on the market a fast quadrupole system, which allows achieving data acquisition rates above 50 Hz allowing their use coupled to GC × GC system [209, 216].

In recent years, it is possible to find a lot of research in the literature applying GC × GC in the analysis of the chemical composition of bio-oils.

Among the initial studies of bio-oils through GC × GC, it can be highlighted the Works of Marsman [205–207] and Sfetsas [174]. Marsman et al. [206, 207] evaluated the compounds presented in the bio-oil from beech using GC × GC with FID and TOFMS detectors. Authors used GC × GC-FID and GC × GC/TOFMS for the identification of approximately 248 and 368 compounds, respectively, with concentration higher than 0.3% in beech (*Fagussylvatica*) hydrodesoxygenated (HDO). In these studies, it was also made a classification for these compounds, according to their chemical class into nine groups (acids, aldehydes, ketones, furans, guaiacols, syringols, sugars, alkyl phenols, alkyl-benzenes). The major compounds found in beech bio-oil were levoglucosan, hydroxymethyl furfural, furanone, furfural, mequinol and butanediol. Similarly, Sfetsas et al. [174] used the GC × GC technique for analyzing three oils pyrolysis, in which were tentatively identified, approximately 96 compounds with concentration higher than 0.3%, classified in acids, esters, aldehydes and ketones, hydrocarbons, aromatic hydrocarbons, phenols, sugars and other compounds not classified. Acetic acid, levoglucosan and hydroxy-propanone were the majority compounds.

Bio-oils derived from the fast pyrolysis of several Brazilian residual biomasses as orange bagasse [217], peach core [15, 189], rice husk [15] and sugarcane straw [8] were recently characterized by GC × GC. The analysis of bio-oil from orange bagasse [15], without any pretreatment, by GC × GC/FID and GC × GC/TOFMS was compared. The last one showed better results and 167 compounds were identified, belonging to acids, aldehydes, ketones, phenols, esters, ethers and some nitrogen-compounds. From these, 26 compounds appeared in concentration above 1%.

Bio-oils from peach core and rice husk analyzed by GC × GC/TOFMS in a conventional set of columns showed the presence of ketones, phenols, alcohols, ethers, acids, aldehydes sugar derivatives and hydrocarbons, with around 500 peaks in each sample [15].

Studying the bio-oil from peach core, by comparison with 1D-GC/qMS and GC × GC/TOFMS, Migliorini et al. [189] observed the superiority of the multidimensional technique. Another observation was the spatial structuration of the GC × GC color diagram, which allowed the identification of all the isomers of C1 to C4 alkyl phenols.

This group of researchers also applies another tool for the identification: the dispersion graphics (DG). These graphics, constructed using Excel™ software clarifying the distribution of compounds and allow to preview the presence of others homologues in a series of compounds, like alkyl substitutes in phenols or in aromatic hydrocarbons. **Figure 4** shows examples of DG for the separation of alkyl phenols. The results from GC/MS and GC × GC/TOFMS showed, for the same sample, 51 and 220 components, respectively. The chemical classes found were alcohols, aldehydes, anhydrides, ketones, esters, ethers and phenols and were observed by both techniques employed. However, using GC × GC were also found carboxylic acids, hydrocarbons and sugar derivatives such as levoglucosan [189].

The analysis of the aqueous extract of peach core pyrolysis is illustrated in **Figure 5**. This figure has been an example of the spatial distribution of the constituents in a GC × GC/TOFMS (**Figure 5A**) and an illustration of separation capacity offered by the second dimension (**Figure 5B**). In this last one, it is observed a separation of four peaks that co-eluted in the first dimension and is adequately separated in the second one, giving mass spectra of high purity.
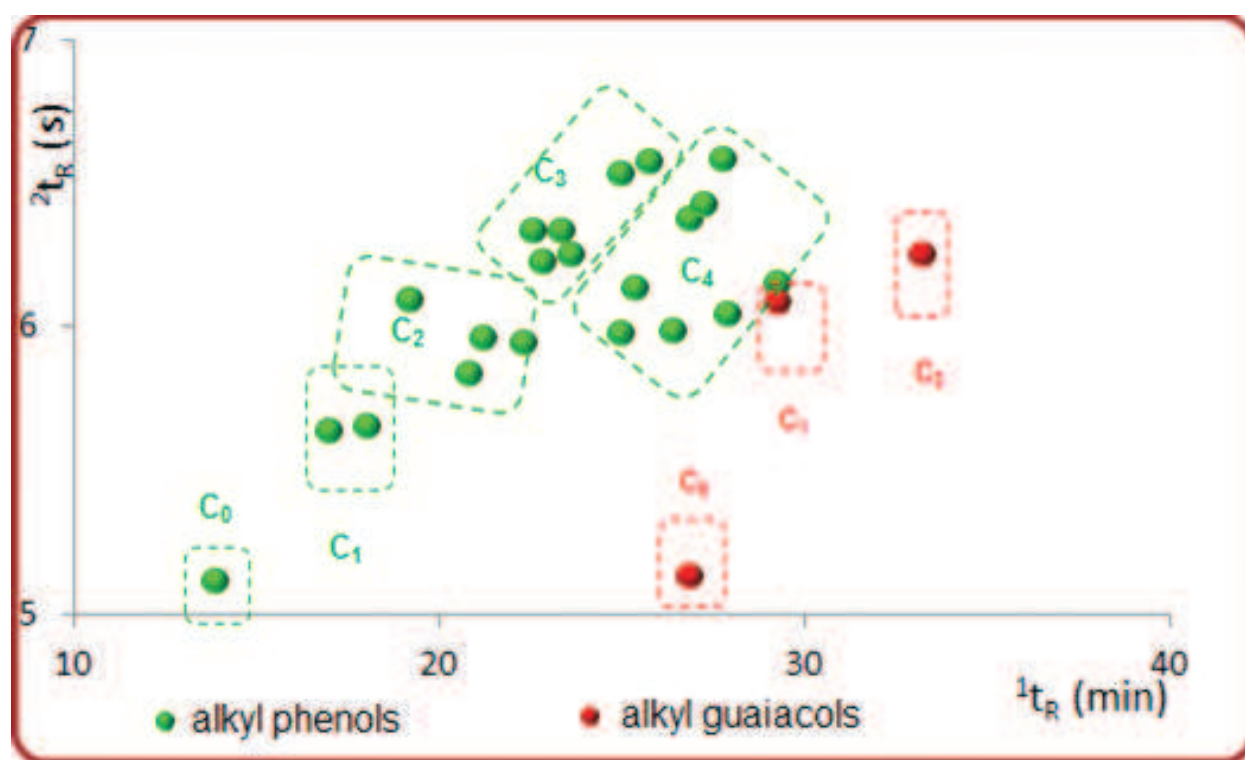
Figure 4. Dispersion graphic of the separation of phenols in the bio-oil from pyrolysis of peach core. Legend: C_x represent the side alkyl chain on the aromatic main chain of the phenols were x is equal to the number of carbon in the side chain. Based on Ref. [91].
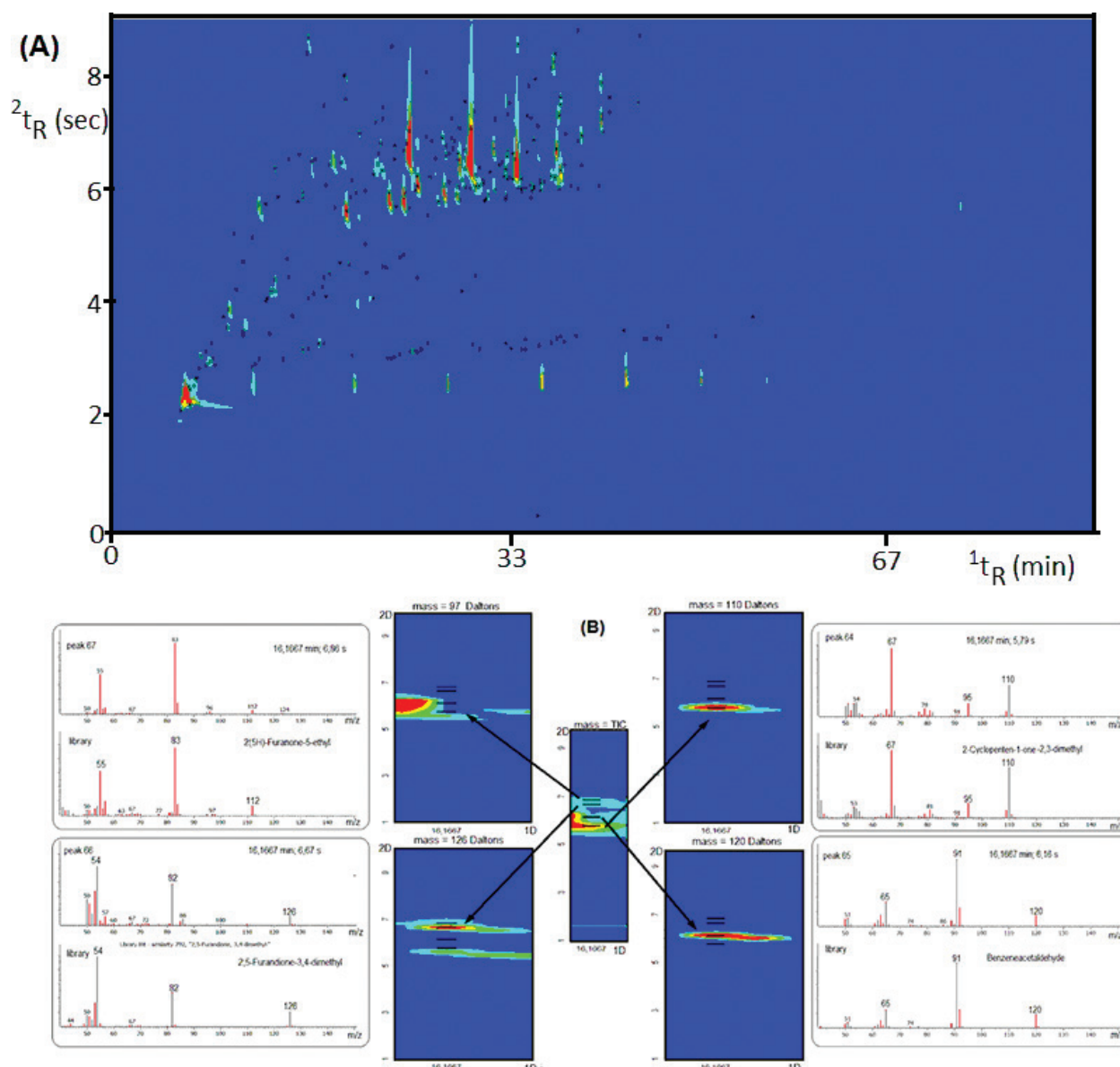
Figure 5. (A) Color two-dimensional diagram for analysis by GC × GC/TOFMS of the organic extract of aqueous fraction of the pyrolysis of peach core. (B) Example of chromatographic separation by GC × GC using the total ion current chromatogram (TIC) and the extracted ion chromatogram (EIC) for the following compounds (a) C2-cyclopentenone, (b) benzene acetaldehyde, (c) ethyl furandione and (d) ethyl furanone and their respective mass spectra. Chromatographic conditions: column set: DB5 (60 m × 0.25 mm × 0.25 μm) and DB17 (1.94 m × 0.18 mm × 0.18 μm); heating ramp: 40.0°C (5.0 min)–3°C/min–270°C.

In the study of fast pyrolysis of sugarcane straw, Moraes and coworkers [8] used GC × GC/ TOFMS, infrared spectroscopy with Fourier transform (FTIR) and scanning electron microscopy (SEM) to fully characterizing the products from the pyrolysis of sugarcane straw. The results of the analysis of bio-oils have demonstrated efficiency in the combination of techniques, especially, GC × GC/TOFMS, showed the presence of 123 compounds belonging, mostly, to the aldehydes and carboxylic acids. Maciel et al. [218] also studied the fast pyrolysis of sugarcane straw by GC × GC/TOFMS, but researching the aqueous phase of this process. They found that this phase is very similar to the bio-oil but enriched more soluble phenols, such as *ortho, meta* and *para* cresols.

The technique using GC × GC with detector quadrupole mass detector (qMS) is growing and showing the efficiency of this detector coupled to two-dimensional gas chromatography. In work carried out by Da Cunha et al. [119] and Schneider et al. [219], using a set of conventional speakers have shown the potential of the technique to evaluate the straw pyrolysis product of sugar cane and forest wood sawdust (lignocel). The bio-oil from pyrolysis of sugarcane straw was fractionated on a silica column with pressurized liquid, being separated hydrocarbons of other polar fractions. In this sample, 166 compounds was to find including carboxylic acids, alde-hydes, ketones, esters, phenols, ethers, alcohols and sugar derivatives in the polar fraction, and, the nonpolar fraction, were formed from aromatic, aliphatic, cyclic and olefinic hydrocarbons [119]. The polar compounds of the bio-oil from lignocel sample (forestry wood sawdust) [219] were extracted with alkaline solution before chromatographic analysis, and 130 compounds were identified by GC × GC/qMS among phenols, ethers, ketones, aldehydes, carboxylic acids, alcohols and aromatic hydrocarbons. This analysis is illustrated in **Figure 6** demonstrating the quality of data generated using a mass spectrometry detector with quadrupole analyzer [119].
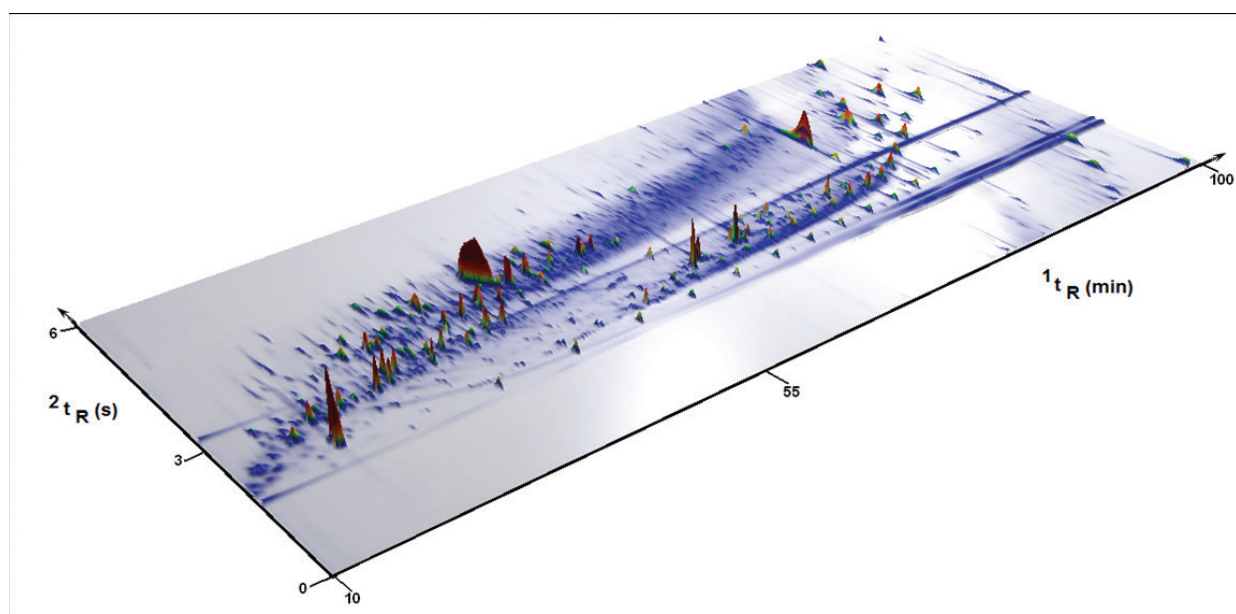
Figure 6. Color diagram (GC × GC/qMS) of the bio-oil from sugarcane straw. Chromatographic conditions: set of columns: OV–5 (60 m × 0.25 mm × 0.25 μm) and DB-17ms (2.15 m × 0.18 mm × 0.18 μm). Heating ramp: 40°C–3°C/min to 120°C, then 2°C/min to 200°C [119].

In recent years, the literature has been publishing a wide range of papers in the pyrolysis of many biomasses with lignocellulosic or residual origin. Eucalyptus [220], mango seed [35], coconut fiber [221], residual cake of crambe seed [37] and castor seed [222], waste of forest industry [36], fruits of palm and pine wood chips [223], sugarcane bagasse [61] sugarcane straw [119] and forest wood sawdust (lignocel) [219] were some of these. These biomasses were submitted to different kinds of pyrolysis (slow [220, 222], fast [35, 37, 177, 221, 223], intermediate [36] and catalyst [34, 218]).

Many other analytical methods can be used to improve the quality of bio-oil and to facilitate its characterization. In this context, extraction methods have been used to isolated fractions and better analyze their components. Some examples of application of extraction techniques are solid phase micro extraction (SPME) [220], liquid-liquid extraction (LLE) [35, 219, 221], mechanical press extraction [37], soxhlet extraction [37], pressurized fluid extraction (PFE) [37] and pressurized liquid fractionation (PLF) [119] among others.

The use of another analytical method beside GC or GC × GC is also a good choice for a more complete characterization of these products. According to reports in the literature GC × GC, especially with TOFMS detector, has been widely used in association with other techniques like GC/qMS [36, 221], FTIR [35], FT-ICR MS [223] and 1H NMR [46] for determining the composition of the biomass pyrolysis products.

The results of the characterization of bio-oil only by GC × GC and the association thereof with other techniques show that different classes of chemical compounds form these samples. The differences in the pyrolysis conditions [14, 224] or in the chemical composition of the biomass can influence the constitution of the composition of bio-oils [178]. The main compounds belonging to the chemical class of phenols, esters, ethers, acids, aldehydes, ketones, alcohols, hydrocarbons (saturates, olefinics and aromatics), sugar derivatives, N-compounds (nitriles, anilines, quinolines, pyridines, indoles, pyrazines, pyrroles, carbazoles and acridines) and sulfur compounds (disulfides and thiophenes) [35–37, 46, 119, 218–223].

As with any other chemical analysis, no single technique is sufficient for complete identification of bio-oil samples. However, the GC × GC has demonstrated its high potential for use combined with other techniques such as infrared spectroscopy, elemental analysis and liquid chromatography with the use of mass detectors among others. The development of rapid chromatographic processes (columns in micro scale) and multidimensional systems (especially comprehensive) allow full characterization of the samples for both constituents in greater proportion as for those at trace levels.

4. Liquid chromatographic methods applied to the analysis of bio-oils

Liquid chromatography (LC) techniques are important tools for the separation and identification of compounds present in bio-oil fractions that are not analyzable by gas chromatography (GC). The high performance liquid chromatography (HPLC) analysis is widely used in different types of samples mainly polar and thermally labile compounds [155]. The main advantage of the LC techniques is the possibility of direct injection of aqueous phases obtained without extraction and sample preparation step. Studies employing such techniques for the characterization of bio-oils and aqueous phases have been recently reported in the literature and are briefly summarized in this chapter.

Similarly as for the GC, the development of LC has been, especially, toward miniaturization and improved chromatographic resolution. Then, one can classify the liquid chromatographic methods in one-dimensional LC and two-dimensional LC.

4.1. One-dimension high performance liquid chromatography

The system can use many detectors, but, the main used for pyrolysis products are UV, RID and MS.

HPLC-UV uses a simple UV detector or diode array detectors (DAD detector or more specifically HPLC PDA detector) especially in the determination of polar compounds containing carbonyl, carboxyl and hydroxyl groups in aqueous samples derived from pyrolysis. The identification of aldehydes in biomass derivatives is the main application of this technique because these compounds are very soluble in water and the recovery of their extraction using conventional techniques such as liquid-liquid extraction or solid phase extraction is very low.

Successful applications of HPLC-UV in the identification of furfural and hydroxymethyl-furfural (HMF) in the aqueous phase of the bio-oil obtained by pyrolysis of agroindustrial biomasses have been described recently [126, 143, 225]. In some cases [225], aldehydes were confirmed and quantified in bio-oil using a GC/MS system.

The HPLC-RID is considered standard for analysis of sugars in aqueous samples. The RID detector is used for detection of compounds that do not absorb in the UV or visible because it is based on measurement of the difference in refractive index between the pure mobile phase and the eluent coming out of the column containing the sample components. As some biomasses (like sugarcane bagasse and sugarcane straw) contain a great amount of sugars, this technique can be important in the characterization of compounds formed during the pyrolysis of these biomasses.

The HPLC-RID has been used, normally, as a complementary technique The bio-oil obtained from red oak fast pyrolysis in a fluidized bed reactor was characterized and sugar derivatives were identified in the water-soluble fraction [148]. In this work, levoglucosan, maltosan celobiosan, xylose and cellobiose were quantified. Johnston and Brown [147] also analyzed glucose and xylose in switchgrass bio-oil samples using HPLC-RID.

HPLC-MS is a technique for the analysis of polar or thermolabile fractions not analyzable by GC on samples of bio-oil and water fraction derived from pyrolysis of biomass. The main difference between the MS in a GC and the MS in a LC system is that LC-MS performs the ionization of analytes in atmospheric pressure (API) with a low-energy fragmentation ("soft"), allowing identification of the molecular ion. The fragmentation can only be done for selected ions and there is no library for identification of compounds. The analyzer must use standard compounds and has to study the entire fragmentation pattern for each peak. HPLC-MS technique is compatible for volatiles and nonvolatiles in a wide range of polarity [226, 227].

On Py-HPLC-MS (online system of pyrolysis and liquid chromatography-mass spectrometry) was recently developed [228] and applied to the analysis of lignin isolated from forest waste sample. In MS, compounds were detected with molecular masses in the range of 250–500 Daltons. The major compounds identified were syringol and resorcinol derivatives.

A method based on the pyrolysis online with HPLC-UV was developed for analyzing the bio-oil derived from polymers [229]. The bio-oil was fractionated and the resulting fractions were analyzed by mass spectrometry ionization and laser desorption matrix assisted (MALDI-MS) system and finally HPLC-MS using electrospray ionization (ESI). This system

is mainly appropriate for the measurements of oligomeric products, being tested in polymer samples forming less volatile pyrolyzates such as poly(butylene terephthalate) and poly(2, 6-dimethyl-1, 4-phenylene ether). Another utility of this method was in the characterization of the cross-linking sequences in some polymeric resins.

HPLC-MS technique in combination with GC-FID and GC/MS was used for characterization of bio-oils from different forest residues [149], showing a wide range of compounds with masses of between 100 and 400 Da. The major compounds identified in all bio-oils were cyclo-hexane carboxylic acid, 1, 2, 4-trimethoxy benzene and 2, 6-dimethyl phenol.

4.2. Two-dimensional LC (heart-cutting (LC-LC)) and comprehensive two-dimensional liquid chromatography (LC × LC)

Two-dimensional liquid chromatography techniques involve two distinct separations, which can be classified as heart-cutting (LC-LC) or comprehensive (LC × LC). In heart-cutting 2D-LC, only relevant parts of the effluent, containing the target compounds, are directed to the second dimension. The main applications of LC-LC are the analytes purification, improve-ment of the separation efficiency and the sensitivity of analysis [150]. Bio-oil obtained from pyrolysis of pine sawdust was analyzed by LC-LC after the gel permeation chromatography (which made a lean up of the high molecular weight lignin derivatives) allowing the separa-tion of phenolic fraction [144]. The results were compared with earlier analyzes by GC/MS. Among the phenols quantified in this work, the major compounds were guaiacol, vanillin, o-cresol e catechol. LC-LC technique proved to be a faster analysis, with a minimal sample preparation and with less loss of analytes than GC/MS.

Similarly to GC, LC techniques with a higher resolution power are also required due to the complexity of the bio oil and aqueous phase samples, and therefore the comprehensive two-dimensional liquid chromatography (LC × LC) becomes also important for the charac-terization of this kind of sample. This technique involves the coupling of two independent mechanisms of separation, through a high-pressure switching valve, and it is able to provide a complete separation of the whole sample, since all fractions eluting from the first dimen-sion are subjected to a second separation [150, 230–239]. The use of LC × LC solves co-eluting problems due to increased peak capacity, which results in a larger number of identified com-pounds. This technique represents a great improvement in the analysis of organic samples mainly due to enhanced of the separation power and the resolution. Carr and Stoll [239] wrote an excellent chapter, edited by Agilent, with theory, instrumental and applications of 2D-LC.

Le Masle et al. [151] used LC × LC with detection by photodiode array (PDA) for the separa-tion of compounds from the aqueous phase formed during the pyrolysis of oak. Using a stan-dard solution with 38 compounds (phenols, acids, ketones, aldehydes, alcohols and furans), the authors developed a separation method, evaluating the peak capacity and the orthogo-nality of different sets of columns. However, the compounds in the aqueous phase samples have not been identified, since a more informative detector as MS would be required for this. This work was after compared with the LC × SFC technique (on-line comprehensive liquid chromatography × supercritical fluid chromatography) for the analysis of the aqueous phase samples by the same researchers, with the aim of evaluating the two-dimensional system. The new method showed a larger peak capacity in comparison with the previous method [152].

Tomasini et al. [153] described a method for the characterization of aqueous phases from bio-oils of coconut fiber, sugarcane straw and sugarcane bagasse using comprehensive two-dimensional liquid chromatography with detection by diode array followed by mass spectrometry with atmospheric pressure chemical ionization. Using this system, it was identified 26 compounds belonging to the classes of phenols, ketones, furans and alcohols. Phenol and 2-hydroxy-3-methyl-2-cyclopenten-1-one were found in greater abundance for all samples. Belonging to furans, the furfural was detected in higher concentrations in the aqueous phases of coconut fiber and sugarcane bagasse and the 1-(2-furanyl)-ethanone was detected in higher concentrations in the aqueous phase of the sugarcane straw. Belonging to alcohols, the phenyl propanol was detected in higher concentrations in the aqueous phase of coconut fiber, while for samples of sugarcane (straw and bagasse), the coniferyl alcohol had the highest concentration.

4.3. NanoLC

Among the innovations that LC has been showing in last years, it stands out techniques that substantially reduce the volume of solvent employed [240, 241]. The NanoLC is one of these innovations. It consists in the use of a column with micro-dimensions and low solvent flow, producing separations in short time but with high performance [242]. As the number of columns manufactured for LC techniques using micro to nano flows is very small (compared to conventional HPLC), there is a limited number of studies described in the literature on this subject [242, 243]. However, due to the positive results obtained, the NanoLC has been successfully applied in many fields such as biomedical, pharmaceutical, agrochemical and food [244].

The low flow used due to the nano-dimensions allows better coupling to mass detectors, including those with electron impact ionization, normally used for GC [153, 244]. In the case of coupling a NanoLC with a mass spectrometer by electron impact (EI-MS), there is the advantage of compounds identification be performed by direct comparison with mass spectral libraries [153, 244, 245].

As a complementary part to the analysis by two-dimensional liquid chromatography of aqueous phases obtained from pyrolysis from Brazilian biomasses, Tomasini et al. [153] applied the NanoLC-EI-MS to confirm the identification of compounds through the mass available libraries in the same samples before cited. The analysis showed a similar composition, with compounds belonging to the classes of ketones, phenols, and furans. The aqueous phase (AP) from coconut fiber presented the phenol as major compound, while the AP from sugarcane bagasse presented a higher amount of furfural and AP from sugarcane straw presented a lower concentration of almost all the compounds.

The use of a mass spectrometry detector with ionization by electron impact (EI-MS) shows an enrichment of information for characterization of the samples, since the obtained mass spectra can be identified by comparison to spectra available in libraries of software used. In addition to the advantages presented by the detection of NanoLC system, it is important to emphasize the "green chemistry" approach due to the reduced volume of solvent used in the analysis of this technique.

In general, the LC is necessary for the characterization of polar compounds remain in the aqueous phase, which normally would be discarded. This discard would cause environmental and

economic damage, since the identified compounds can have some kind of industrial application. Furthermore, the injection of aqueous samples is not possible without a prior step of extraction, which may have different yields due to the presence of compounds belonging to different chemical classes or can result in contamination of the sample, besides the higher use of materials and longer time.

Although there is a still reduced number of works applying LC for the analysis of bio-oil compared to GC, it should be considered the importance of using new analytical techniques for a complete characterization of bio-oils and aqueous phases from pyrolysis of biomass.

5. Conclusions

The initially suggested use for bio-oil was as an alternative biofuel to diesel and petroleum. However, this route has proved to be nonviable, due to the high oxygen content of bio-oils and operating cost of deoxygenation. Currently, the major studies indicate the use of bio-oil in the chemical industry, particularly the chemistry of phenols, furfural and levoglucosan. The bio-oil can be considered an alternative for crude oil for fine chemical industry.

For proper use of bio-oil in the chemical industry, it is essential the identification and unambiguous determination of its major constituents. Only then, it is possible to propose a recovery route of some of these components for the development of an industry dedicated to a bio-refinery. For this, chromatographic methods, especially GC × GC/MS, are fundamental because they allow analysis with high sensitivity and accuracy in identifying each constituent of the bio-oil.

Acknowledgements

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References

[1] Mythili R, Venkatachalam P, Subramanian P, Uma D. Characterization of bioresidues for bio-oil production through pyrolysis. Bioresour Technol. 2013, **138**: 71–78.

[2] Thangalazhy-Gopakumar S, Adhikari S, Gupta RB, Fernando SD. Influence of pyrolysis operating conditions on bio-oil components: a microscale study in a pyroprobe. Energy Fuel. 2011, **25**: 1191–1199.

[3] Nzihou A, Flamant G, Stanmore B. Synthetic fuels from biomass using concentrated solar energy—a review. Energy. 2012, **42**: 121–131.

[4] Akhtar J, Amin NAS. A review on process conditions for optimum bio-oil yield in hydro-thermal liquefaction of biomass. Renew Sust Energy Rev. 2011, **15**: 1615–1624.

[5] Van de Velden M, Baeyens J, Brems A, Janssens B, Dewil R. Fundamentals, kinetics and endothermicity of the biomass pyrolysis reaction. Renew Energy. 2010, **35**: 232–242.

[6] Bridgwater AV, Peacocke GVC. Fast pyrolysis processes for biomass. Renew Sust Energy Rev. 2000, **4**: 1–73.

[7] Cao JP, Xiao XB, Zhang SY, Zhao XY, Sato K, Ogawa Y, Wei XY, Takarada T. Preparation and characterization of bio-oils from internally circulating fluidized-bed pyrolysis of municipal, livestock, and wood waste. Bioresour Technol. 2011, **102**: 2009–2015.

[8] Moraes MSA, Georges F, Almeida SR, Damasceno FC, Maciel GPS, Zini CA, Jacques RA, Caramão EB. Analysis of products from pyrolysis of Brazilian sugar cane straw. Fuel Process Technol. 2012, **101**: 35–43.

[9] Akhtar J, Amin NS. A review on operating parameters for optimum liquid oil yield in biomass pyrolysis. Renew Sust Energ Rev. 2012, **16**: 5101–5109.

[10] Venderbosch RH, Prins W. Biofuels. Fast pyrolysis technology development. Bioprod Bioref. 2010, **4**: 178–208.

[11] Tsai WT, Lee MK, Chang YM. Fast pyrolysis of rice husk: product yields and compositions. Bioresour Technol. 2007, **98**: 22–28.

[12] Ferdinand FW, Steene LV, Blaise KK, Siaka T. Prediction of pyrolysis oils higher heating value with gas chromatography–mass spectrometry. Fuel. 2012, **96**: 141–154.

[13] Zhang K, Chang J, Guan Y, Chen H, Yang Y, Jiang J. Renew Energy. 2013, **49**: 175.

[14] Mohan D, Pittman CU, Steele PH. Pyrolysis of wood/biomass for bio-oil:a critical review. Energy Fuel. 2006, **20**: 848–889.

[15] Moraes MSA, Migliorini MV, Damasceno FC, Georges F, Almeida S, Zini CA, Jacques RA, Caramão EB. Qualitative analysis of bio oils of agricultural residues obtained through pyrolysis using comprehensive two dimensional gas chromatography with time-of-flight mass spectrometric detector. J Anal Appl Pyrolysis. 2012, **98**: 51–64.

[16] Jahirul MI, Rasul MG, Chowdhury AA, Ashwath N. Biofuels production through biomass pyrolysis—a technological review. Energies. 2012, **5**: 4952–5001.

[17] Önal PE, Uzun BB, Pütün EA. Steam pyrolysis of an industrial waste for bio-oil production. Fuel Process Technol. 2011, **92**: 879–885.

[18] Duman G, Okutucu C, Ucar S, Stahl R, Yanik J. The slow and fast pyrolysis of cherry seed. Bioresour Technol. 2011, **102**: 1869–1878.

[19] Martins AF, Diniz J, Stahl JA, Cardoso AL. Characterization of liquid products and char from the pyrolysis of eucalyptus sawdust. Quim Nova. 2007, **30**: 873–878.

[20] Weerachanchai P, Tangsathitkulchai C, Tangsathitkulchai M. Characterization of products from slow pyrolysis of palm kernel cake and cassava pulp residue. Korean J Chem Eng. 2011, **28**: 2262–2274.

[21] Pütüm AE, Özbay N, Pönal N, Pütüm E. Fixed-bed pyrolysis of cotton stalk for liquid and solid products. Fuel Process Technol. 2005, **86**: 1207–1219.

[22] Demiral I, Ayan EA. Pyrolysis of grape bagasse: effect of pyrolysis conditions on the product yields and characterization of the liquid product. Bioresour Technol. 2011, **102**: 3946–3951.

[23] Bridgwater AV. Review of fast pyrolysis of biomass and product upgrading. Biomass Bioenergy. 2012, **38**: 68–94.

[24] Butler E, Devlin G, Meier D, McDonnell K. A review of recent laboratory research and commercial developments in fast pyrolysis and upgrading. Renew Sustain Energy Rev. 2011, **15**: 4171–4186.

[25] Bridgwater AV, Meier D, Radlein D. A review of recent laboratory research and commercial developments in fast pyrolysis and upgrading. Org Geochem. 1999, **30**: 1479–1486.

[26] Bridgwater AV. Renewable fuels and chemicals by thermal processing of biomass. Chem Eng J. 2003, **91**: 87–102.

[27] Meier D. Renew sustain. State-of-the-art of fast pyrolysis in IEA bioenergy member countries. Energy Rev. 2013, **20**: 619–641.

[28] Kunii P, Levenspiel O. Fluidization engineering. In: Entrainment and Elutriation from Fluidized Beds, Chapter 7, 2nd edition. Robert E Krieger Publishing Company Inc, Huntington, New York, pp. 165–193. 1977. ISBN 0-409-90233-0.

[29] Bridgwater AV. Biomass fast pyrolysis. Thermal Sci. 2004, **8**: 21–49.

[30] Rozainee M. Effect of fluidising velocity on the combustion of rice husk in a bench-scale fluidised bed combustor for the production of amorphous rice husk ash. Bioresour Technol. 2008, **99**: 703–713.

[31] Penlidis A, McGregor JF, Hamielec AE. Continuous emulsion polymerization: design and control of CSTR trains. Chem Eng Sci. 1989, **44**: 273–281.

[32] Nogueira AL, Lona LMF, Machado RAF. Continuous polymerization in tubular reactors with prepolymerization: analysis using two-dimensional phenomenological model and hybrid model with neural networks. J Appl Polym Sci. 2004, **91**: 871–882.

[33] Palma M, Giudici R. Analysis of axial dispersion in an oscillatory-flow continuous reactor. Chem Eng J. 2003, **94**: 189–198.

[34] Almeida TM, Bispo MD, Cardoso ART, Migliorini MV, Schena T, de Campos MCV, Machado ME, López JA, Krause LC, Caramão EB. Analysis of products from pyrolysis of Brazilian sugar cane straw. J Agric Food Chem. 2013, **61**: 6812–6821.

[35] Lazzari E, Schena T, Primaz CT, Maciel GPS, Machado ME, Cardoso CAL, Jacques RA, Caramão EB. Production and chromatographic characterization of bio-oil from the pyrolysis of mango seed waste. Ind Crop Prod. 2016, **83**: 529–536.

[36] Torri IDV, Paasikallio V, Faccini CS, Huff R, Caramão EB, Sacon V, Oasmaa A, Zini CA. Bio-oil production of softwood and hardwood forest industry residues through fast and intermediate pyrolysis and its chromatographic characterization. Bioresour Technol. 2016, **200**: 680–690.

[37] Onorevoli B, Machado ME, Dariva C, Franceschi E, Krause LC, Jacques RA, Caramão EB. A one-dimensional and comprehensive two-dimensional gas chromatography study of the oil and the bio-oil of the residualcakes from the seeds of *Crambe abyssinica*. Ind Crop Prod. 2014, **52**: 8–16.

[38] Doumer ME, Arízaga GGC, da Silva DA, Yamamoto CI, Novotny EH, Santos JM, dos Santos LO, Wisniewski Jr A, de Andrade JB, Mangrich AS. Slow pyrolysis of different Brazilian waste biomasses as sources of soil conditioners and energy, and for environmental protection. J Anal Appl Pyrolysis. 2015, **113**: 434–443.

[39] Santos RM, Santos AO, Sussuchi EM, Nascimento JS, Lima ÁS, Freitas LS. Pyrolysis of mangaba seed: production and characterization of bio-oil. Bioresour Technol. 2015, **196**: 43–48.

[40] Figueiredo MKK, Romeiro GA, d'Avila LA, Damasceno RN, Franco AP. The isolation of pyrolysis oil from castor seeds via a low temperature conversion (LTC) process and its use in a pyrolysis oil-diesel blend. Fuel. 2009, **88**: 2193–2198.

[41] Czernik S, Bridgwater AV. Overview of applications of biomass fast pyrolysis oil. Energy Fuel. 2004, **18**: 590–598.

[42] Tanneru SK, Steele PH. Pretreating bio-oil to increase yield and reduce char during hydrodeoxygenation to produce hydrocarbons. Fuel. 2014, **133**: 326–331.

[43] Ospina V, Buitrago-Sierra R, López D. HDO of guaiacol over NiMo catalyst supported on activated carbon derived from castor de-oiled cake. Ingeniería InvestIgación. 2015, **35**: 49–55. doi:10.15446/ing. investig.v35n2.49838

[44] Mortensen PM, Grunwaldt JD, Jensen PA, Jensen AD. Influence on nickel particle size on the hydrodeoxygenation of phenol over Ni/SiO_2. Catal Today. 2016, **259**: 277–284.

[45] Vitolo S, Seggiani M, Frediani P, Ambrosini G, Politi L. Catalytic upgrading of pyrolytic oils to fuel over different zeolites. Fuel. 1999, **78**: 1147–1159.

[46] Tessarolo NS, Silva RVS, Vanini G, Casilli A, Ximenes VL, Mendes FL, Pinho AR, Romão W, de Castro EVR, Kaiser CR, Azevedo DA. Characterization of thermal and catalytic pyrolysis bio-oils by high-resolution techniques: 1 H NMR, GC× GC-TOFMS and FT-ICR MS. J Anal Appl Pyrolysis. 2016, **117**: 257–267.

[47] Shetty M, Murugappan K, Prasomsri T, Green WH, Leshkov YR. Reactivity and stability investigation of supported molybdenum oxide catalysts for the hydrodeoxygenation (HDO) of m-cresol. J Catal. 2015, **331**: 86–97.

[48] Persson NE, Blass SD, Rosenthal C, Bhan A, Schmidt LD. On-line deoxygenation of cellulose pyrolysis vapors in a staged autothermal reactor. RSC Adv. 2013, **3**: 20163–20170.

[49] Furimsky E. Hydroprocessing challenges in biofuels production. Catal Today. 2013, **217**: 13–56.

[50] Huber GW, Iborra S, Corma A. Synthesis of transportation fuels from biomass: chemistry, catalysts, and engineering. Chem Rev. 2006, **106**: 4044–4098.

[51] Wang K, Johnston PA, Brown RC. Comparison of *in-situ* and *ex-situ* catalytic pyrolysis in a micro-reactor system. Bioresour Technol. 2014, **173**: 124–131.

[52] Dorado C, Mullen CA, Boateng AA. Origin of carbon in aromatic and olefin products derived from HZSM-5 catalyzed co-pyrolysis of cellulose and plastics via isotopic labeling. Appl Catal B: Environ. 2015, **162**: 338–345.

[53] Stefanidis SD, Kalogiannis KG, Iliopoulou EF, Lappas AA, Pilavachi PA. In-situ upgrading of biomass pyrolysis vapors: catalyst screening on a fixed bed reactor. Bioresour Technol. 2011, **102**: 8261–8267.

[54] Patel MA, Baldanza MAS, Da Silva VT, Bridgwater AV. In situ catalytic upgrading of bio-oil using supported molybdenum carbide. Appl Catal A: General. 2013, **458**: 48–54.

[55] Wang L, Lei H, Bu Q, Ren S, Wei Y, Zhu L, Zhang X, Liu Y, Yadavalli G, Lee J, Chen S, Tang J. Aromatic hydrocarbons production from ex situ catalysis of pyrolysis vapor over Zinc modified ZSM-5 in a packed-bed catalysis coupled with microwave pyrolysis reactor. Fuel. 2014, **129**: 78–85.

[56] Ludlow-Palafox C, Chase HA. Microware pyrolysis of plastic wastes. In: Scheirs J, Kaminsky W, editors. Feedstock Recycling and Pyrolysis of Waste Plastics, Chapter 21. John Wiley & Sons Ltd, pp. 569–594. 2006. ISBN 9780470021545.

[57] Holland KM, US pat 5084141, 1992.

[58] Holland KM, US pat 5387321, 1995.

[59] Fernández Y, Arenillas A, Menéndez JA. Microwave heating applied to pyrolysis. In: Grundas S, editor. Advances in Induction and Microwave Heating of Mineral and Organic Materials, Chapter 31, p. 723. 2011. doi:10.5772/562

[60] Luque R, Menéndez JA, Arenillas A, Cot J. Microwave-assisted pyrolysis of biomass feedstocks: the way forward? Energy Environ Sci. 2012, **5**: 5481–5488.

[61] Budarin VL, Clark JH, Lanigan BA, Shuttleworth P, Breeden SW, Wilson AJ, Macquarrie DJ, Milkowski K, Jones J, Bridgeman T, Ross A. In-situ upgrading of biomass pyrolysis vapors: catalyst screening on a fixed bed reactor. Bioresour Technol. 2009, **100**: 6064–6068.

[62] Mehdizadeh M. Microwave/RF Applicators and Probes for Material Heating Sensing and Plasma Generation. William Andrew Publishing, Boston, p. 400. 2010. ISBN-10 0815515928.

[63] Javadi HHS. Microwave materials. In: Ishii T, editor. Handbook of Microwave Technology: Components and Devices, Chapter 19. Elsevier, Milwaukee, Wisconsin, pp. 605–643. 1995. ISBN 9780123746955.

[64] Thostenson ET, Chou TW. Microwave processing: fundamentals and applications. Compos Part A: App Sci Manuf. 1999, **30**: 1055–1071.

[65] Makibar J, Akarregi ARF, Amutio M, Lopez G, Olazar M. Performance of a conical spouted bed pilot plant for bio-oil production by poplar flash pyrolysis. Fuel Process Technol. 2015, **137**: 283–289.

[66] http://wwwpyrolysisoilnet/PRODUCTS_SOLUTIONS/Tire_Pyrolysis_Plant/ (accessed September 10, 2016).

[67] Pichini S, Rotolo MC, Bellotti P, Minutilloa A, Mastrobattistaa L, Pacifici R. Quali-quantitative analysis of best selling drugs from pharmacy, street market and traditional herbal medicine: a pilot study of market surveillance in Senegal. J Pharm Biomed. 2015, **104**: 62–66.

[68] Amutio M, Lopez G, Alvarez J, Olazar M, Bilba J. In-situ upgrading of biomass pyrolysis vapors: catalyst screening on a fixed bed reactor. Bioresour Technol. 2015, **194**: 225–232.

[69] Toor SS, Rosendahl L, Nielsen MP, Glasius M, Rudolf A, Iversen SB. Continuous production of bio-oil by catalytic liquefaction from wet distiller's grain with solubles (WDGS) from bio-ethanol production. Biomass Bioenerg. 2012, **36**: 327–332.

[70] Pérez JMM, Rocha JD, Cortez LAB, Medina MP, Luengo CA, Cascarosa E. Fast oxidative pyrolysis of sugar cane straw in a fluidized bed reactor. Appl Therm Eng. 2013, **56**: 167–175.

[71] Shemfe MB, Gu S, Ranganathan P. Techno-economic performance analysis of biofuel production and miniature electric power generation from biomass fast pyrolysis and bio-oil upgrading. Fuel. 2015, **143**: 361–372.

[72] https://patentesgooglecom (accessed September 10, 2016).

[73] https://patentscopewipoint (accessed September 10, 2016).

[74] Gómez N, Rosas JG, Cara J, Martínez O, Alburquerque JA, Sánchez ME. Slow pyrolysis of relevant biomasses in the Mediterranean basin. Part 1. Effect of temperature on process performance on a pilot scale. J Clean Prod. 2016, **120**: 181–190.

[75] Moncada JB, Aristizábal VM, Cardona A. Design strategies for sustainable biorefineries. J Biochem Eng. 2016. doi:101016/jbej201606009 (in press)

[76] Sadhukhan J, Lloyd JR, Scott K, Premier GC, Yu HE, Curtis T, Head IM. A critical review of integration analysis of microbial electrosynthesis (MES) systems with waste biorefineries for the production of biofuel and chemical from reuse of CO_2. Renew Sust Energ Rev. 2016, **56**: 116–132.

[77] Kokossis AC, Tsakalova M, Pyrgakis K. Design of integrated biorefineries. Comput Chem Eng. 2015, **81**: 40–56.

[78] Mota FAS, Viegas RAA, Lima AS, Santos FP, Caselli FTR. Pyrolysis of lignocellulose biomass: a review. Revista GEINTEC. 2015, **5**: 2511–2525. ISSN: 2237-0722. doi: 107198/S2237-0722201500040003

[79] Roy C, Blanchette D, Korving L, Yang J, Caumia B. Development of a novel vacuum pyrolysis reactor with improved heat transfer potential. In: Bridgewater AV, Boocock DGB, editors. Thermochem Biomass Convers. Blackie Academic and Professional, London, UK, p. 351. 1997.

[80] Alvarez J, Lopez G, Amutio M, Bilbao J. Bio-oil production from rice husk fast pyrolysis in a conical spouted bed reactor. Fuel. 2014, **128**: 162–169.

[81] Tang L, Huang H. Plasma pyrolysis of biomass for production of syngas and carbon adsorbent. Energy Fuels. 2005, **19**: 1174–1178.

[82] Huang X, Cheng D, Chen F, Zhan X. Reaction pathways of hemicellulose and mechanism of biomass pyrolysis in hydrogen plasma: a density functional theory study. Renew Energy. 2016, **96**: 490–497.

[83] Tang L, Huang H, Hao H, Zhao K. Development of plasma pyrolysis/gasification systems for energy efficient and environmentally sound waste disposal. J Electrostat. 2013, **71**: 839–847.

[84] Yan B, Xu P, Guo CYYJ, Cheng Y. Experimental study on coal pyrolysis to acetylene in thermal plasma reactors. J Chem Eng. 2012, **207**: 109–116.

[85] Yan B, Cheng Y, Jin Y, Guo CY. Performance of a conical spouted bed pilot plant for bio-oil production by poplar flash pyrolysis. Fuel Process Technol. 2012, **100**: 1–10.

[86] Punčochář M, Ruj B, Chatter PK. Development of process for disposal of plastic waste using plasma pyrolysis technology and option for energy recovery. Procedia Eng. 2012, **42**: 420–430.

[87] Fernández Y, Menéndez JA. Influence of feed characteristics on the microwave-assisted pyrolysis used to produce syngas from biomass wastes. J Anal App Pyrolysis. 2011, **91**: 316–322.

[88] Lam SS, Russell AD, Chase HA. Microwave pyrolysis, a novel process for recycling waste automotive engine oil. Energy. 2010, **35**: 2985–2991.

[89] Domínguez A, Fernández Y, Fidalgo B, Pis JJ, Menéndez JA. Bio-syngas production with low concentrations of CO_2 and CH_4 from microwave-induced pyrolysis of wet and dried sewage sludge. Chemosphere. 2008, **70**: 397–403.

[90] Domínguez A, Menéndez JA, Fernández Y, Pis JJ, Valente Nabais JM, Carrott PJM, Ribeiro Carrott MML. Conventional and microwave induced pyrolysis of coffee hulls for the production of a hydrogen rich fuel gas. J Anal App Pyrolysis. 2007, **79**: 128–135.

[91] Huang YF, Kuan WH, Lo SL, Lin CF. Total recovery of resources and energy from rice straw using microwave-induced pyrolysis. Bioresour Technol. 2008, **99**: 8252–8258.

[92] Zhao X, Song Z, Liu H, Li Z, Li L, Ma C. Microwave pyrolysis of corn stalk bale: a promising method for direct utilization of large-sized biomass and syngas production. J Anal App Pyrolysis. 2010, **89**: 87–94.

[93] Hofmann L, Antal MJ. Numerical simulations of the performance of solar fired flash pyrolysis reactors. Sol Energy. 1984, **33**: 427–440.

[94] Shakya BD. PhD Thesis School of Chemical and Biomolecular Engineering. University of Sydney, Sydney, Australia, p. 98. 2007.

[95] Li R, Zeng K, Soria JGM, Gauthier D, Rodriguez R, Flamant G. Product distribution from solar pyrolysis of agricultural and forestry biomass residues. Renew Energy. 2016, **89**: 27–35.

[96] Zeng K, Gauthier D, Li R, Flamant G. Solar pyrolysis of beech wood: effects of pyrolysis parameters on the product distribution and gas product composition. Energy. 2015, **93**: 1648–1657.

[97] Zeng K, Gauthier D, Lu J, Flamant G. Parametric study and process optimization for solar pyrolysis of beech wood. Energy Convers Manage. 2015, **106**: 987–998.

[98] Zeng K, Flamant G, Gauthier D, Guillot E. Solar pyrolysis of wood in a lab-scale solar reactor: influence of temperature and sweep gas flow rate on products distribution. Energy Procedia. 2015, **69**: 1849–1858.

[99] Zeng K, Minh DP, Gauthier D, Hortala EW, Nzihou A, Flamant G. The effect of temperature and heating rate on char properties obtained from solar pyrolysis of beech wood. Bioresour Technol. 2015, **182**: 114–119. doi:10.1016/j.biortech.2015.01.112

[100] Chopra S, Jain AK. A review of fixed bed gasification systems for biomass. Agric Eng Int CIGR J. 2007, **5**: 1–23.

[101] Rao MS, Singha SP, Sodha MS, Dubey AK, Shyam M. Stoichiometric, mass, energy and exergy balance analysis of countercurrent fixed-bed gasification of post-consumer residues. Biomass Bioenery. 2004, **27**: 155–171. doi:10.1016/j.biombioe.2003.11.003

[102] Lv PM, Xiong ZH, Chang J, Wu CZ, Chen Y, Zhu JX. An experimental study on biomass air–steam gasification in a fluidized bed. Bioresour Technol. 2004, **95**: 95–101. doi:10.1016/j.biortech.2004.02.003

[103] Li XT, Grace R, Lim CJ, Watkinson AP, Chen HP, Kim JR. Biomass gasification in a circulating fluidized bed. Biomass Bioenergy. 2004, **26**: 171–193. doi:10. 1016/ S0961-9534(03)00084-9

[104] Joubert JE, Carrier M, Dahmen N, Stahl R, Knoetze JH. Inherent process variations between fast pyrolysis technologies: a case study on *Eucalyptus grandis*. Fuel Process Technol. 2015, **131**: 389–395. doi:10.1016/j.fuproc.2014.12.012

[105] Kook JW, Choi HM, Kim BH, Ra HW, Yoon SJ, Mun TY, Kim JH, Kim YK, Lee JG, Seo MW. Gasification and tar removal characteristics of rice husk in a bubbling fluidized bed reactor. Fuel. 2016, **181**: 942–950. doi:10. 1016/j. fuel. 2016. 05. 027

[106] Wagenaar BM, Venderbosch RH, Carrasco J, Strenziok R, Van der BJA. Rotating cone bio-oil production and applications. In: Bridgwater AV, editor. Progress in Thermochemical Biomass Conversion. Blackwell Science, Oxford, pp. 4952–5001. 2001. doi:10.1002/9780470694954.ch105

[107] Garcia-Perez M, Chaala A, Pakdel H, Kretschmer D, Roy C. Characterization of bio-oils in chemical families. Biomass Bioenergy. 2007, **31**: 222–242. doi:10.1016/j.biom-bioe.2006.02. 006

[108] Kim J-S. Production, separation and applications of phenolic-rich bio-oil—a review. Bioresour Technol. 2015, **178**: 90–98. doi:10.1016/j.biortech.2014.08.121

[109] Kim T-S, Kim J-Y, Kim KH, Lee S, Choi D, Choi I-G, Choi JW. The effect of storage duration on bio-oil properties. J Anal Appl Pyrolysis. 2012, **95**: 118–125. doi:10.1016/j. jaap.2012.01.015

[110] Shen DK, Gu S. The mechanism for thermal decomposition of cellulose and its main products. Bioresour Technol. 2009, **100**: 6496–6504. doi:10.1016/j.biortech.2009.06.095

[111] Shen DK, Gu S, Luo KH, Wang SR, Fang MX. The pyrolytic degradation of wood-derived lignin from pulping process. Bioresour Technol. 2010, **101**: 6136–6146. doi:10.1016/j.biortech.2010.02.078

[112] Kanaujia PK, Sharma YK, Garg MO, Tripathi D, Singh RJ. Review of analytical strategies in the production and upgrading of bio-oils derived from lignocellulosic biomass. J Anal Appl Pyrolysis. 2014, **105**: 55–74. doi:10.1016/j.jaap.2013.10.004

[113] Mortensen PM, Grunwaldt JD, Jensen PA, Knudsen KG, Jensen DA. A review of catalytic upgrading of bio-oil to engine fuels. Appl Catal A General. 2011, **407**: 1–19. doi:10.1016/j. apcata.2011.08.046

[114] Zhang L, Liu R, Yin R, Mei Y. Upgrading of bio-oil from biomass fast pyrolysis in China: a review. Renew Sustain Energy Rev. 2013, **24**: 66–72. doi:10.1016/j.rser.2013.03.027

[115] Lehto J, Oasmaa A, Solantausta Y, Kytö M, Chiaramonti D. Review of fuel oil quality and combustion of fast pyrolysis bio-oils from lignocellulosic biomass. Appl Energy. 2014, **116**: 178–190. doi:10.1016/j.apenergy.2013.11.040

[116] Chiaramonti D, Oasmaa A, Solantausta Y. Power generation using fast pyrolysis liquids from biomass. Renew Sustain Energy Rev. 2007, **11**: 1056–1086. doi:10.1016/j.rser.2005.07.008

[117] Yang Z, Kumar A, Huhnke RL. Review of recent developments to improve storage and transportation stability of bio-oil. Renew Sustain Energy Rev. 2015, **50**: 859–870. doi:10.1016/j.rser.2015.05.025

[118] Cao JP, Zhao XY, Morishita K, Wei XY, Takarada T. Fractionation and identification of organic nitrogen species from bio-oil produced by fast pyrolysis of sewage sludge. Bioresour Technol. 2010, **101**: 7648–7652. doi:10.1016/j.biortech.2010.04.073

[119] Cunha ME, Schneider JK, Brasil MC, Cardoso CA, Monteiro LR, Mendes FL, Pinho A, Jacques RA, Machado ME, Freitas LS, Caramão EB. Analysis of fractions and bio-oil of sugar cane straw by one-dimensional and two-dimensional gas chromatography with quadrupole mass spectrometry (GC × GC/qMS). Microchem J. 2013, **110**: 113–119. doi:10.1016/j.microc.2013.03.004

[120] Sukhbaatar B, Steele PH, Ingram LL, Kim MG. An exploratory study on the removal of acetic and formic acids from bio-oil. Bioresources. 2009, **4**: 1319–1329.

[121] Bertero M, Gorostegui HA, Orrabalis CJ, Guzmán CA, Calandri EL, Sedran U. Characterization of the liquid products in the pyrolysis of residual chañar and palm fruit biomasses. Fuel. 2014, **116**: 409–414. doi:10.1016/j.fuel.2013.08.027

[122] Sukhbaatar B, Steele PH, Kim MG. Use of lignin separated from bio-oil in oriented strand board binder phenol-formaldehyde resins. Bioresources. 2009, **4**: 789–804.

[123] Mourant D, Wang Z, He M, Wang XS, Garcia-Perez M, Ling K, Li C-Z. Mallee wood fast pyrolysis: effects of alkali and alkaline earth metallic species on the yield and composition of bio-oil. Fuel. 2011, **90**: 2915–2922. doi:10.1016/j.fuel.2011.04.033

[124] Isahak WNRW, Hisham MWM, Yarmo MA, Hin TY. A review on bio-oil production from biomass by using pyrolysis method. Renew Sustain Energy Rev. 2012, **16**: 5910–5923. doi:10.1016/j.rser.2012.05.039

[125] Vitasari CR, Meindersma GW, de Haan AB. Water extraction of pyrolysis oil: the first step for the recovery of renewable chemicals. Bioresour Technol. 2011, **102**: 7204–7210. doi:10.1016/j.biortech.2011.04.079

[126] Abou-Yousef H, Hassan EB. Efficient utilization of aqueous phase bio-oil to furan derivatives through extraction and sugars conversion in acid-catalyzed biphasic system. Fuel. 2014, **137**: 115–121. doi:10.1016/j.fuel.2014.07.067

[127] Teella A, Huber GW, Ford DMJ. Separation of acetic acid from the aqueous fraction of fast pyrolysis bio-oils using nanofiltration and reverse osmosis membranes. J Membrane Sci. 2011, **378**: 495–502. doi:10.1016/j.memsci.2011.05.036

[128]   Bharti A, Banerjee T. Enhancement of bio-oil derived chemicals in aqueous phase using ionic liquids: experimental and COSMO-SAC predictions using a modified hydrogen bonding expression. Fluid Phase Equil. 2015, **400**: 27–37. doi:10.1016/j.fluid.2015.04.029

[129]   Das DD, Schnitzer MI, Monreal CM, Mayer P. Chemical composition of acid-base fractions separated from biooil derived by fast pyrolysis of chicken manure. Bioresour Technol. 2009, **100**: 6524–6532. doi:10.1016/j.biortech.2009.06.104

[130]   Wang S, Wang Y, Cai Q, Wang X, Jin H, Luo Z. Multi-step separation of monophenols and pyrolytic lignins from the water-insoluble phase of bio-oil. Sep Pur Technol. 2014, **122**: 248–255. doi:10.1016/j.seppur.2013.11.017

[131]   de Oliveira AM, Biajoli AFP, Fidelis CHV, Silva RGC, Augusto F. Trends in sample preparation. In: Arruda MAZ, editor. Nova Science, New York, p. 142. 2006.

[132]   Xu R, Ferrante L, Briens C, Berruti FJ. Flash pyrolysis of grape residues into biofuel in a bubbling fluid bed. J Anal Appl Pyrolysis. 2009, **86**: 58–65. doi:10.1016/j.jaap.2009.04.005

[133]   Xu R, Ferrante L, Briens C, Berruti FJ. Bio-oil production by flash pyrolysis of sugarcane residues and post treatments of the aqueous phase. J Anal Appl Pyrolysis. 2011, **91**: 263–272. doi:10.1016/j.jaap.2011.03.001

[134]   Xie J, Su D, Yin X, Wu C, Zhu J. Thermodynamic analysis of aqueous phase reforming of three model compounds in bio-oil for hydrogen production. Int J Hydro Energy. 2011, **36**: 15561–15572. doi:10.1016/j.ijhydene.2011.08.103

[135]   Sanna A, Vispute TP, Huber GW. Hydrodeoxygenation of the aqueous fraction of bio-oil with Ru/C and Pt/C catalysts. Appl Catal B Environ. 2015, **165**: 446–456. doi:10.1016/j.apcatb.2014.10.013

[136]   Vispute TP, Huber GW. Production of hydrogen, alkanes and polyols by aqueous phase processing of wood-derived pyrolysis oils. Green Chem. 2009, **11**: 1433–1445. doi:10.1039/B912522C

[137]   Huber GW, Dumesic JA. An overview of aqueous-phase catalytic processes for production of hydrogen and alkanes in a biorefinery. Catal Today. 2006, **111**: 119–132. doi:10.1016/j.cattod.2005.10.010

[138]   Gerdes C, Simon CM, Ollesch T, Kaminsky W. Design, construction, and operation of a fast pyrolysis plant for biomass. Eng Life Sci. 2002, **2**: 167–174. doi:10.1002/1618-2863(200206)2:6<167::AID-ELSC167>3. 0. CO, 2-F

[139]   Bayerbach R, Meier D. Characterization of the water-insoluble fraction from fast pyrolysis liquids (pyrolytic lignin). Part IV: structure elucidation of oligomeric molecules. J Anal Appl Pyrolysis. 2009, **85**: 98–107. doi:10.1016/j.jaap.2008.10.021

[140] Djokic MR, Dijkmans T, Yildiz G, Prins W, Van Geem KM. Quantitative analysis of crude and stabilized bio-oils by comprehensive two-dimensional gas-chromatography. J Chromatogr A. 2012, **1257**: 131–140. doi:10.1016/j.chroma.2012.07.035

[141] von Muhlen C, Zini CA, Caramao EB, Marriott PJ. Applications of comprehensive two-dimensional gas chromatography to the characterization of petrochemical and related samples. J Chromatogr A. 2006, **1105**: 39–50. doi:10.1016/j.chroma.2005.09.036

[142] Marriott PJ, Shellie R. Principles and applications of comprehensive two-dimensional gas chromatography. TrAC Trends Anal Chem. 2002, **21**: 573–583. doi:10.1016/S0165-9936(02)00814-2

[143] Tessini C, Müller N, Mardones C, Meier D, Berg A, von Baer D. Chromatographic approaches for determination of low-molecular mass aldehydes in bio-oil. J Chromatogr A. 2012, **1219**: 154–160. doi:10.1016/j.chroma.2011.10.093

[144] Andersson T, Hyotylainen T, Riekkola M-L. Analysis of phenols in pyrolysis oils by gel permeation chromatography and multidimensional liquid chromatography. J Chromatogr A. 2000, **896**: 343–349. doi:10.1016/S0021-9673(00)00678-6

[145] Tienpont B, David F, Pereira A, Sandra P. In-vial pyrolysis (PyroVial) with pre- and post-sample treatment combined with different chromatographic techniques. J Chromatogr A. 2011, **1218**: 8303–8312. doi:10.1016/j.chroma.2011.09.053

[146] Yanik J, Stahl R, Troeger N, Sinag A. Pyrolysis of algal biomass. J Anal Appl Pyrolysis. 2013, **103**: 134–141. doi:10.1016/j. jaap.2012.08.016

[147] Johnston PA, Brown RC. Quantitation of sugar content in pyrolysis liquids after acid hydrolysis using high-performance liquid chromatography without neutralization. J Agric Food Chem. 2014, **62**: 8129–8133. doi:10. 1021/jf502250n

[148] Choi YS, Johnston PA, Brown RC, Shanks BH, Lee K-H. Detailed characterization of red oak-derived pyrolysis oil: integrated use of GC, HPLC, IC, GPC and Karl-Fischer. J Anal Appl Pyrolysis. 2014, **110**: 147–154. doi:10.1016/j.jaap.2014.08.016

[149] Undri A, Abou-Zaid M, Briens C, Berruti F, Rosi L, Bartoli M, Frediani M, Frediani P. J Anal Appl Pyrolysis. 2015, **114**: 208–221. doi:10.1016/j.jaap.2015.05.019

[150] Jacques RA, Caramão EB. Two dimensional liquid chromatography. Sci Chromatogr. 2009, **1**: 73–82.

[151] Le Masle A, Angot D, Gouin C, D'Attoma A, Ponthus J, Quignard A, Heinisch S. Development of on-line comprehensive two-dimensional liquid chromatography method for the separation of biomass compounds. J Chromatogr A. 2014, **1340**: 90–98.

[152] Sarrut M, Corgier A, Crétier G, Le Masle A, Dubant S, Heinisch S. Potential and limitations of on-line comprehensive reversed phase liquid chromatography × supercritical fluid chromatography for the separation of neutral compounds: an approach to separate an aqueous extract of bio-oil. J Chromatogr A. 2015, **1402**: 124–133.

[153] Tomasini D, Cacciola F, Rigano F, Sciarrone D, Donato P, Beccaria M, Caramão EB, Dugo P, Mondello L. Complementary analytical liquid chromatography methods for the characterization of aqueous phase from pyrolysis of lignocellulosic biomasses. Anal Chem. 2014, **86**: 11255–11262.

[154] Zhang Q, Chang J, Wang T, Xu Y. Review of biomass pyrolysis oil properties and upgrading research. Energy Convers Manag. 2007, **48**: 87–92.

[155] Kanaujia PK, Sharma YK, Agrawal UC, Garg MO. Comprehensive two dimensional gas chromatography review. Trends Anal Chem. 2013, **42**: 125–136.

[156] Pighinelli ALMT, Boateng AA, Mullen CA, Elkasabi Y. Evaluation of Brazilian biomasses as feedstocks for fuel production via fast pyrolysis. Energy Sustain Dev. 2014, **21**: 42–50.

[157] Lira CS, Berruti FM, Palmisano P, Berruti F, Briens C, Pécora AAB. Fast pyrolysis of Amazon *tucumã* (*Astrocaryum aculeatum*) seeds in a bubbling fluidized bed reactor. J Anal Appl Pyrolysis. 2013, **99**: 23–31.

[158] Tessarolo NS, dos Santos LRM, Silva RSF, Azevedo DA. Chemical characterization of bio-oils using comprehensive two-dimensional gas chromatography with time-of-flight mass spectrometry. J Cromatogr A. 2013, **1279**: 68–75.

[159] Spilã K, Kuoppala E, Fagernãs L, Oasmaa A. Characterization of biomass-based flash pyrolysis oils. Biomass Bioenergy. 1998, **14**: 103–113.

[160] Oasmaa A, Kuoppala E, Solantausta Y. Fast pyrolysis of forestry residue. 2. Physicochemical composition of product liquid. Energy Fuels. 2003, **17**: 433–443.

[161] Fagernäs L, Kuoppala E, Tiilikkala K, Oasmaa A. Chemical composition of birch wood slow pyrolysis products. Energy Fuels. 2012, **26**: 1275–1283.

[162] Wiggers VR, Meier HF, Wisniewski AJ, Barros AAC, Maciel MRW. In-situ upgrading of biomass pyrolysis vapors: catalyst screening on a fixed bed reactor. Bioresour Technol. 2009, **100**: 6570–6577.

[163] Kuddusi M, Karagöz S. Analytical pyrolysis of biomass using gas chromatography coupled to mass spectrometry. Trends Anal Chem. 2014, **61**: 11–16.

[164] Mullen CA, Boateng AA. Catalytic pyrolysis-GC/MS of lignin from several sources. Fuel Process Technol. 2010, **91**: 1446–1458.

[165] Liaw SS, Perez VH, Zhou S, Justo OR, Perez MG. Py-GC/MS studies and principal component analysis to evaluate the impact of feedstock and temperature on the distribution of products during fast pyrolysis. J Anal Appl Pyrolysis. 2014, **109**: 140–151.

[166] Ralph J, Hatfield RD. Pyrolysis-GC-MS characterization of forage materials. J Agric Food Chem. 1991, **39**: 1426–1437.

[167] Del Río JC, Gutiérrez A, Hernando M, Landín P, Romero J, Martínez AT. Determining the influence of eucalypt lignin composition in paper pulp yield using Py-GC/MS. J Anal Appl Pyrolysis. 2005, **74**: 110–115.

[168] Barbosa LCA, Maltha CRA, Silva VL, Colodette JL. Determination of the siringyl/guaiacyl ratio in eucalyptus wood by pyrolysis-gas chromatography/mass spectrometry (PY-GC/MS). Quim Nova. 2008, **31**: 2035–2041.

[169] Fortes ICP, Baugh PJ. Study of analytical on-line pyrolysis of oils from macauba fruit (Acrocomia sclerocarpa M) via GC/MS. J Braz Chem Soc. 1999, **10**: 469–477.

[170] Vinhal JO, Lima CF, Barbosa LCA. Analytical pyrolysis of the kernel and oil of babassu palm (*Orbignya phalerata*). J Anal Appl Pyrol. 2014, **107**: 73–81.

[171] Lu Q, Dong C-K, Zhang X-M, Tian H-Y, Yang Y-P, Zhu Xi-F. Selective fast pyrolysis of biomass impregnated with ZnCl$_2$ to produce furfural: analytical Py-GC/MS study. J Anal Appl Pyrol. 2011, **90**: 204–212.

[172] Carvalho WS, Cunha IF, Pereira MS, Ataíde CH. Thermal decomposition profile and product selectivity of analytical pyrolysis of sweet sorghum bagasse: effect of addition of inorganic salts. Ind Crop Prod. 2015, **74**: 372–380.

[173] Cardoso CR, Ataíde CH. Analytical pyrolysis of tobacco residue: effect of temperature and inorganic additives. J Anal Appl Pyrolysis. 2013, **99**: 49–57.

[174] Sfetsas T, Michailof C, Lappas A, Li Q, Kneale B. Qualitative and quantitative analysis of pyrolysis oil by gas chromatography with flame ionization detection and comprehensive two-dimensional gas chromatography with time-of-flight mass spectrometry. J Chromatogr A. 2011, **1218**: 3317–3325. doi:10.1016/j.chroma.2010.10.034

[175] Case PA, Truong C, Wheeler MC, DeSisto WJ. Calcium-catalyzed pyrolysis of lignocellulosic biomass components. Bioresour Technol. 2015, **192**: 247–252.

[176] Liu Z, Phillips JB. Comprehensive two-dimensional gas chromatography using an on-column thermal modulator interface. J Chromatogr Sci. 1991, **29**: 227–231.

[177] Cortes HJ, Winniford B, Luong J, Pursch M. Comprehensive two dimensional gas chromatography review. J Sep Sci. 2009, **32**: 883–904.

[178] Özbay N, Apaydın-Varol E, Uzun EB, Pütün AE. Characterization of bio-oil obtained from fruit pulp pyrolysis. Energy. 2008, **33**: 1233–1240.

[179] Busetto L, Fabbri D, Mazzoni R, Salmi M, Torri C, Zanotti V. Techno-economic performance analysis of biofuel production and miniature electric power generation from biomass fast pyrolysis and bio-oil upgrading. Fuel. 2011, **90**: 1197–1207.

[180] Wannapeera J, Fungtammasan B, Worasuwannarak N. Qualitative and quantitative analysis of pyrolysis oil by gas chromatography with flame ionization detection and comprehensive two-dimensional gas chromatography with time-of-flight mass spectrometry. J Anal Appl Pyrol. 2011, **92**: 99–105.

[181] Wang Y, Chen Q, Norwood DL, McCaffrey J. Recent development in the applications of comprehensive two-dimensional gas chromatograph. J Liquid Chromatogr Related Technol. 2010, **33**: 1082–1115.

[182] Dallüge J, Beens J, Brinkman UAT. Comprehensive two-dimensional gas chromatography: a powerful and versatile analytical tool. J Chromatogr A. 2003, **1000**: 69–108.

[183] Von Mühlen C, Zini CA, Caramão EB, Marriott PJ. Nomenclature for comprehensive multidimensional chromatography in portuguese language. Quím Nova. 2007, **30**: 682–687.

[184] Adahchour M, Beens J, Vreuls RJJ, Brinkman UAT. Recent developments in comprehensive two-dimensional gas chromatography (GC×GC): IV. Further applications, conclusions and perspectives. Trends Anal Chem. 2006, **25**: 821–840.

[185] Adam F, Bertoncini F, Coupard V, Charon N, Thiébaut D, Espinat D, Hennion M-C. Using comprehensive two-dimensional gas chromatography for the analysis of oxygenates in middle distillates: I. Determination of the nature of biodiesels blend in diesel fuel. J Chromatogr A. 2008, **1186**: 236–244.

[186] Flego C, Gigantiello N, Parker Jr WO, Calemma V. A comprehensive two-dimensional gas chromatography coupled with quadrupole mass spectrometry approach for identification of C_{10} derivatives from decalin. J Chromatogr A. 2009, **1216**: 2891–2899.

[187] Hua R, Li Y, Liu W, Zheng J, Wei H, Wang J, Lu X, Kong H, Xu G. Determination of sulfur-containing compounds in diesel oils by comprehensive two-dimensional gas chromatography with a sulfur chemiluminescence detector. J Chromatogr A. 2003, **1019**: 101–109.

[188] Moraes MSA, Zini CA, Gomes CB, Bortoluzzi JH, von Mühlen C, Caramão EB. Application of comprehensive two-dimensional gas chromatography (GC×GC) in the characterization of biodiesel/diesel blends: application to beef tallow biodiesel. Quím Nova. 2011, **34**: 1188–1192.

[189] Migliorini MV, Moraes MSA, Machado ME, Caramão EB. Caracterization of phenols in bio-oil from the pyrolysis of peach core by GC/MS e GC×GC/TOFMS. Sci Chromatogr. 2013, **5**: 47–65.

[190] Hoh E, Matovska K, Lehotay SJ. Optimization of separation and detection conditions for comprehensive two-dimensional gas chromatography–time-of-flight mass spectrometry analysis of polychlorinated dibenzo-*p*-dioxins and dibenzofurans. J Chromatogr A. 2007, **1145**: 210–221.

[191] Pedroso MP, de Godoy LAF, Fidélis CHV, Ferreira EC, Poppi RJ, Augusto F. Comprehensive two-dimensional gas chromatography (GC × GC). Quím Nova. 2009, **32**: 421–430.

[192] Von Mühlen C, Khummueng W, Zini CA, Caramão EB, Marriott PJ. Detector technologies for comprehensive two-dimensional gas chromatography. J Sep Sci. 2006, **29**: 1909–1921.

[193] Kallio M, Hyotyläianen T, Lehtonen M, Jussila M, Hartonen K, Shimmo M, Riekkola ML. Comprehensive two-dimensional gas chromatography in the analysis of urban aerosols. J Chromatogr A. 2003, **1019**: 251–260.

[194] Adahchour M, Beens J, Vreuls RJJ, Brinkman UAT. Recent developments in comprehensive two-dimensional gas chromatography (GC×GC): II. Modulation and detection. Trends Anal Chem. 2006, **25**: 540–553.

[195] Beens J, Adahchour M, Vreuls RJJ, Van Altena K, Brinkman UAT. Simple, non-moving modulation interface for comprehensive two-dimensional gas chromatography. J Chromatogr A. 2001, **919**: 127–132.

[196] Gaines RB, Frysinger GS. Temperature requirements for thermal modulation in comprehensive two-dimensional gas chromatography. J Sep Sci. 2004, **27**: 380–388.

[197] Adahchour M, Beens J, Brinkman UAT. Recent developments in the application of comprehensive two-dimensional gas chromatography. J Chromatogr A. 2008, **1186**: 67–108.

[198] Barceló D. Comprehensive two dimensional gas chromatography. In: Ramos L, editor. Comprehensive Analytical Chemistry Series, volume 55. Elsevier, Amsterdam. 2009. eBook ISBN 9780080932699.

[199] Bertsch W. Recent developments in the application of comprehensive two-dimensional gas chromatography. J High Resol Chromatogr. 2000, **23**: 167–181.

[200] Godoy LAF, Hantao LW, Pedroso MP, Poppi RJ, Augusto F. Quantitative analysis of essential oils in perfume using multivariate curve resolution combined with comprehensive two-dimensional gas chromatography. Anal Chim Acta. 2011, **699**: 120–125. doi:10.1016/j.aca.2011.05.003

[201] Striebich RC, Contreras J, Balster LM, West Z, Shafer LM, Zabarnick S. Identification of polar species in aviation fuels using multidimensional gas chromatography-time of flight mass spectrometry. Energy Fuels. 2009, **23**: 5474–5482. doi:10.1021/ef900386x

[202] Vendeuvre C, Ruiz-Guerrero R, Bertoncini F, Duval L, Thiebaut D. Comprehensive two-dimensional gas chromatography for detailed characterization of petroleum products. Oil and Gas Sci Technol. 2007, **62**: 43–55. doi:10.2516/ogst:2007004

[203] Von Mühlen C, de Oliveira EC, Zini CA, Caramão EB, Marriott PJ. Characterization of nitrogen-containing compounds in heavy gas oil petroleum fractions using comprehensive two-dimensional gas chromatography coupled to time-of-flight mass spectrometry. Energy Fuels. 2010, **24**: 3572–3580. doi:10.1021/ef1002364

[204] Khummueng W, Trenerry C, Rose G, Marriot P. Application of comprehensive two-dimensional gas chromatography with nitrogen-selective detection for the analysis of fungicide residues in vegetable samples. J Chromatogr A. 2006, **1131**: 203–214. doi:10.1016/j.chroma.2006.07.035

[205] Marsman JH, Wildschut J, Mahfud F, Heeres HJ. Identification of components in fast pyrolysis oil and upgraded products by comprehensive two-dimensional gas chromatography and flame ionization detection. J Chromatogr A. 2007, **1150**: 21–27. doi:10.1016/j.chroma.2006.11.047

[206] Marsman JH, Wildschut J, Evers P, de Koning S, Heeres HJ. Identification and classification of components in flash pyrolysis oil and hydrodeoxygenated oils by two-dimensional gas chromatography and time-of-flight mass spectrometry. J Chromatogr A. 2008, **1188**: 17–25. doi:10.1016/j.chroma.2008.02.034

[207] Windt M, Meier D, Marsman JH, Heeres HJ, de Koning S. Micro-pyrolysis of technical lignins in a new modular rig and product analysis by GC–MS/FID and GC×GC–TOFMS/FID. J Anal Appl Pyr. 2009, **85**: 38–46. doi:10.1016/j.jaap.2008.11.011

[208] Kallio MK. Comprehensive Two-Dimensional Gas Chromatography: Instrumental and Methodological Development [Academic Dissertation]. University of Helsinki, Helsinki. 2008.

[209] Purcaro G, Tranchida PQ, Ragonese C, Conte L, Dugo P, Dugo G, Mondello L. Evaluation of a rapid-scanning quadrupole mass spectrometer in an apolar × ionic-liquid comprehensive two-dimensional gas chromatography system. Anal Chem. 2010, **82**: 8583–8590. doi:10.1021/ac101678r

[210] Korytar P, Haglund P, Deboer J, Brinkman UAT. Comprehensive two-dimensional gas chromatography for the analysis of organohalogenated micro-contaminants. Trends Anal Chem. 2006, **25**: 373–396. doi:10.1016/j.trac.2005.12.003

[211] Mondello L, Casilli A, Tranchida PQ, Dugo G, Dugo P. Comprehensive two-dimensional gas chromatography in combination with rapid scanning quadrupole mass spectrometry in perfume analysis. J Chromatogr A. 2005, **1067**: 235–243. doi:10.1016/j.chroma.2004.09.040

[212] Schmarr H-G, Bernhardt J, Fischer U, Stephan A, Müller P, Durner D. Two-dimensional gas chromatographic profiling as a tool for a rapid screening of the changes in volatile composition occurring due to microoxygenation of red wines. Anal Chim Acta. 2010, **672**: 114–123. doi:10.1016/j.aca.2010.05.002

[213] Song SM, Marriott P, Wynne P. Comprehensive two-dimensional gas chromatography—quadrupole mass spectrometric analysis of drugs. J Chromatogr A. 2004, **1058**: 223–232. doi:10.1016/j.chroma.2004.08.087

[214] Mondello L, Casilli A, Tranchida PQ, Lo Presti M, Dugo P, Dugo G. Comprehensive gas chromatography coupled to mass spectrometry for the separation of pesticides in a very complex matrix. Anal Bioanal Chem. 2007, **389**: 1755–1763. doi:10.1007/s00216-007-1412-y

[215] Frysinger GS, Gaines RB. Comprehensive two-dimensional gas chromatography with mass spectrometric detection (GC×GC/MS) applied to the analysis of petroleum. J Sep Sci. 1999, **22**: 251–255. doi:10.1002/(SICI)1521-4168(19990501)22:5<251::AID-JHRC251>3.0.CO,2-V

[216] Kochman M, Gordin A, Alon T, Amirav A. Flow modulation comprehensive two-dimensional gas chromatography–mass spectrometry with a supersonic molecular beam. J Chromatogr A. 2006, **1129**: 95–104. doi:10.1016/j.chroma.2006.06.079

[217] Moraes MAS, Bortoluzzi JH, Migliorini MV, Zini CA, Caramão EB. Cromatografia gasosa bidimensional abrangente aplicada à análise qualitativa dos componentes majoritários do bio-óleo da pirólise de bagaço de laranja. Sci Chromatogr. 2011, **3**: 301–314. doi:10.4322/sc.2011.018

[218] Maciel GPS, Machado ME, Barbara JA, Dal Molin D, Caramão EB, Jacques RA. GC × GC/TOFMS analysis concerning the identification of organic compounds extracted from the aqueous phase of sugarcane straw fast pyrolysis oil. Biomass Bioenergy. 2016, **85**: 198–206. doi:10.1016/j.biombioe.2015.11.009

[219] Schneider JK, Cunha ME, Santos AL, Maciel GPS, Brasil MC, Pinho AR, Mendes FL, Jacques RA, Caramão EB. Comprehensive two dimensional gas chromatography with fast-quadrupole mass spectrometry detector analysis of polar compounds extracted from the bio-oil from the pyrolysis of sawdust. J Chromatogr A. 2014, **1356**: 236–240. doi:10.1016/j.chroma.2014.06.053

[220] Araujo RC, Pasa VDM, Marriot PJ, Cardeal ZL. Analysis of volatile organic compounds in polyurethane coatings based on Eucalyptus sp. bio-oil pitch using comprehensive two-dimensional gas chromatography (GC×GC). J Anal Appl Pyrolysis. 2010, **88**: 91–97. doi:10.1016/j.jaap.2010.02.012

[221] Almeida TM, Bispo MD, Cardoso ART, Migliorini MV, Schena T, Campos MCV, Machado ME, López JA, Krause LC, Caramão EB. Preliminary studies of bio-oil from fast pyrolysis of coconut fibers. 2013, **61**: 6812–6821. doi:10.1021/jf401379s

[222] Silva RVS, Casilli A, Sampaio AL, Ávila BMF, Veloso MCC, Azevedo DA, Romeiro GA. The analytical characterization of castor seed cake pyrolysis bio-oils by using comprehensive GC coupled to time of flight mass Spectrometry. J Anal Appl Pyrolysis. 2014, **106**: 152–159. doi:10.1016/j.jaap.2014.01.013

[223] Tessarolo NS, Silva RC, Vanini G, Pinho A, Romão W, Castro EVR, Azevedo DA. Assessing the chemical composition of bio-oils using FT-ICR mass spectrometry and comprehensive two-dimensional gas chromatography with time-of-flight mass Spectrometry. Microchem J. 2014, **117**: 68–76. doi:10.1016/j.microc.2014.06.006

[224] Salehi E, Abedi J, Harding T. Bio-oil from sawdust: pyrolysis of sawdust in a fixed-bed system. Energy Fuels. 2009, **23**: 3767–3772. doi:10.1021/ef900112b

[225] Martinez A, Rodriguez ME, York SW, Preston JF, Ingram LO. Use of UV absorbance to monitor furans in dilute acid hydrolysates of biomass. Biotechnol Progr. 2000, **16**: 637–641. doi:10.1021/bp0000508

[226] Lanças FM. A cromatografia líquida moderna e a espectrometria de massas: finalmente "compatíveis"? Sci Chromatogr. 2009, **1**: 35–61.

[227] Chiaradia MC, Collins CH, Jardim ICSF. O estado da arte da cromatografia acoplada à espectrometria de massas na análise de compostos tóxicos em alimentos. Quim Nova. 2008, **31**: 623–636. doi:10.1590/S0100-40422008000300030

[228] van der Hage ERE, Boon JJ. On-line Curie-point pyrolysis-high-performance liquid chromatographic-mass spectrometric analysis of lignin polymers. J Chromatogr A. 1996, **736**: 61–75. doi:10.1016/0021-9673(95)01345-8

[229] Kano H, Okamoto T, Kitagawa S, Iiguni Y, Ohtani H, Ito H, Iwaia K, Kuno M. A novel analytical pyrolysis device applicable for measurements of less volatile pyrolyzates. J Anal Appl Pyrolysis. 2015, **113**: 165–173. doi:10.1016/j.jaap.2014.12.008

[230] Dugo P, Cacciola F, Kumm T, Dugo G, Mondello L. Comprehensive multidimensional liquid chromatography: theory and applications. J Chromatogr A. 2008, **1184**: 353–368. doi:10.1016/j.chroma.2007.06.074

[231] Francois I, Sandra K, Sandra P. Comprehensive liquid chromatography: fundamental aspects and practical considerations—a review. Anal Chim Acta. 2009, **641**: 14–31. doi:10.1016/j.aca.2009.03.041

[232] Guiochon G, Marchetti N, Mriziq K, Shalliker R. Implementations of two-dimensional liquid chromatography. J Chromatogr A. 2008, **1189**: 109–168. doi:10.1016/j.chroma.2008.01.086

[233] Bedani F, Schoenmakers PJ, Janssen H. Theories to support method development in comprehensive two-dimensional liquid chromatography—a review. Sep Sci. 2012, **35**: 1697–1711. doi:10.1002/jssc.201200070

[234] Jandera P, Hájek T, Česla P. Comparison of various second-dimension gradient types in comprehensive two-dimensional liquid chromatography. Sep Sci. 2010, **33**: 1382–1397. doi:10.1002/jssc.200900808

[235] Camenzuli M, Schoenmakers PJ. A new measure of orthogonality for multi-dimensional chromatography. Anal Chim Acta. 2014, **838**: 93–101. doi:10.1016/j.aca.2014.05.048

[236] Elsner V, Laun S, Melchior D, Koehler M, Schmitz OJ. Analysis of fatty alcohol derivatives with comprehensive two-dimensional liquid chromatography coupled with mass spectrometry. J Chromatogr A. 2012, **1268**: 22–28. doi:10.1016/j.chroma.2012.09.072

[237] Blumberg L, Klee MS. A critical look at the definition of multidimensional separations. J Chromatogr A. 2010, **1217**: 99–103. doi:10.1016/j.chroma.2009.08.081

[238] Carr PW, Stoll DR. Two-Dimensional Liquid Chromatography—Principles, Practical Implementation and Applications. Agilent Technologies Inc, Germany, p. 182. 2015. ISBN 5991-2359EN.

[239] François I, de Villiers A, Tienpont B, David F, Sandra P. Comprehensive two-dimensional liquid chromatography applying two parallel columns in the second dimension. J Chromatogr A. 2008, **1178**: 33–42. doi:10.1016/j.chroma.2007.11.032

[240] Rieux L, Lubda D, Niederlander HAG, Verpoorte E, Bischoff R. Fast, high-efficiency peptide separations on a 50-μm reversed-phase silica monolith in a nanoLC–MS set-up. J Chromatogr A. 2006, **1120**: 165–172. doi:10.1016/j.chroma.2006.01.038

[241] Rigano F, Albergamo A, Sciarrone D, Beccaria M, Purcaro G, Mondello L. Nano liquid chromatography directly coupled to electron ionization mass spectrometry for free fatty acid elucidation in mussel. Anal Chem. 2016, **88**: 4021–4028. doi:10.1021/acs.analchem.6b00328

[242] Lanças FM. Vantagens e limitações da miniaturização em cromatografia líquida. Sci Chromatogr. 2009, **1**: 51–60.

[243] Silva RGC, Collins CH, Bottoli CBG. Capillary liquid chromatography: state of the art and current applications. Quim Nova. 2011, **34**: 841–849. doi:10.1590/S0100-40422011000500020

[244] Fanali C, Dugo L, Dugo P, Mondello L. Capillary-liquid chromatography (CLC) and nano-LC in food analysis. Trends Anal Chem. 2013, **52**: 226–238. doi:10.1016/j.trac.2013.05.021

[245] Cappiello A, Famiglini G, Palma P, Trufelli H. Advanced liquid chromatography-mass spectrometry interface based on electron ionization. Anal Chem. 2007, **79**: 5364–5372. doi:10.1021/ac070468l