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The analysis of energy potential in vine leaves of the 'Regent' cultivar as bio-waste depending on the year of cultivation and the type of rootstock used

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Abstract: The study evaluated physicochemical properties of bio-waste as a potential biofuel in the form of leaves from 'Regent' grapevines grown on six different rootstocks and a control grown on its own roots for three years of cultivation. An elemental analysis was carried out, determining the content of carbon, hydrogen, nitrogen, and sulphur in the leaves tested. A technical analysis of the biofuel was also carried out to determine the content of moisture, volatile matter, and ash. The calorimetric method was used to determine the higher heating value for the material. Fixed carbon and oxygen carbon was calculated based on the elemental and technical analyses. The study showed that the type of rootstock and the year of cultivation influence the amount of leaves obtained from the cultivation area. Leaf entrustment per hectare ranged from 1,140,868.02 in rootstock 161-49 to 1,265,286.7 Mg·ha⁻¹ in rootstock SO4. Regardless of the year of the study, shrubs grafted on 125AA rootstock and the control had the highest combustion heat of 17.5 MJ·kg⁻¹ and 17.6 MJ·kg⁻¹ respectively, while 5BB rootstock had the lowest combustion heat (16.4 MJ·kg⁻¹). Statistical analysis showed no significant effect of test year on the elemental and technical parameters evaluated. It was observed that regardless of the evaluated parameter and the type of rootstock in most parameters, the values in 2022 were the highest, while in 2021 they were the lowest.

Keywords: biomass utilisation, energetic properties, plant biomass, renewable energy, rootstock

INTRODUCTION

In most of the world's grape-growing regions, grapevines (*Vitis vinifera* L.) are grafted onto rootstocks resistant to phylloxera (*Daktulosphaira vitifoliae*). This insect appeared in European vineyards growing its own roots in the 1850s. It devastated significant areas and then spread to other regions around the world. French researchers discovered that the problem could be solved by grafting European vines onto American species (Pouget, 1990). This gave rise to the establishment of vineyards using grafted vines. Initially, single American species were used for this purpose. Today, rootstocks are derived from crossing two or more *Vitis* species.

When choosing the type of rootstock, it is important to consider the location of the plantation (sunshine, amount of

rainfall, type of soil and the presence of various organisms in it, such as insects, fungi and nematodes). The selection of a suitable rootstock also depends on the characteristics of the interaction between rootstock, scion and the environment. In addition, the purpose of production should also be considered. These characteristics can produce different responses to vegetative growth, grape yield size and quality, and grape composition and sensory attributes. In fact, each factor, and interaction between them, can unevenly induce nutrient assimilation by roots, sap translocation in the xylem system and accumulation in grapevine tissues. This leads to the biosynthesis of a wide range of compounds, different biochemical reactions and consequently grapevine physiology (Miele and Rizzon, 2017). In fact, there is a wide cultivar of rootstocks, each with characteristics sought by Magdalena Kapłan, Grzegorz Maj, Kamila E. Klimek

grape growers for specific growing conditions and purposes. The most important of these are related to soil parasites, climatic adversity, adaptability to soil nutrient excess or deficiency, and vine vigour.

There are papers covering various aspects of rootstock effects on grapevines, such as those related to physiology (Virgona, Smith and Holzapfel, 2003; Cookson *et al.*, 2012), biochemistry (Somkuwar *et al.*, 2014; Souza de *et al.*, 2015), mineral nutrition (Miele, Rizzon and Giovannini, 2009; Kodur *et al.*, 2011), yield (Terra *et al.*, 2003; Keller, Mills and Harbertson, 2012), water deficiency or excess (Heralde de *et al.*, 2006), salinity (Walker *et al.*, 2007), fungal diseases (Brown *et al.*, 2013; Wallis, Wallingford and Chen, 2013), viruses (Rosa *et al.*, 2011) and nematodes (Ferris, Zheng and Walker, 2012).

Along with coal and oil, biomass is an important source of primary energy. The main sources of biomass are energy crops, agricultural waste, forestry and organic waste. More recently, biomass has been used in energy production by combined heat and power plants (CHP) to produce heat and electricity (Świerzewski and Kalina, 2020). A CHP provides great potential for significant improvement in energy efficiency, which explains the interest in converting biomass to heat and power (Asomaning *et al.*, 2018). Finding new renewable energy sources to produce solid biofuels is a priority from a climate and environmental perspective, with the aim to mitigate the effects of global warming while producing cleaner energy. Recently, densified solid biofuels have seen rapid growth due to increasing demand for biomass used for heating, electricity and biofuel (Bajwa *et al.*, 2018).

The annual production of a large amount of green matter and pruning in vineyards ensure a large number of vine shoots (Sánchez-Gómez *et al.*, 2017). The use of vineyard pruning waste has received a lot of attention over the past few years. Vineyard pruning waste obtained from agricultural practices can be used as a renewable energy source in accordance with European Waste Directive 2008/98/EC, which focuses on waste management, recycling and conversion to energy (Directive, 2008). Currently, vineyard pruning waste is used for: surfactant production by autohydrolysis, delignification with NaOH, enzymatic hydrolysis and fermentation with *Lactobacillus paracasei* (Vecino *et al.*, 2017), bioactive compounds (Moreira *et al.*, 2018), bioethanol and chemicals (Pachón, Mandade and Gnansounou, 2020), and the production of cellulose nanocrystals for the development of nanocomposite materials (Achaby *et al.*, 2018).

In recent years, with the effort to reduce pollution, renewable energy production has increased in European countries (Muench and Guenther, 2013). Of all renewable energy sources, biomass seems to be the one that stands out for its better performance in power and heat production (Guo, Song and Buhain, 2015). Agricultural residues could become a potential source of biomass for energy production not only in Poland but also in other European countries (Velazquez-Marti et al., 2001; Scarlat, Blukdea and Dallemand, 2011), especially in Italy (Bernetti, Fagarazzi and Fratini, 2004; Beccali, Columba and D'Aleberti, 2009). In fact, new biomass is available every year and is produced in areas accessible to tractors and vehicles (Magagnotti et al., 2013). In addition, the use of agricultural waste has little environmental impact compared to dedicated energy crops (Gonzalez-Garcia et al., 2014). Leftovers in the form of leaves or shoots after pruning in vineyards, whose exhaust emissions are comparable to those obtained from wood chips, can

be a suitable fuel for energy production (Picchi, Silvestri and Cristoforetti, 2013). Unlike orchards, in order to improve the quality and quantity of vine production, vineyards require significant pruning of all plants each year. This results in significant amounts of residue (Blasi di, Tanzi and Lanzetta, 1997). Currently, the residue is mulched in the vineyards or stored outside vineyards and burned (Spinelli et al., 2014). Both solutions pose problems in terms of time consumption, economic sustainability and environmental impact. Mulching, contributes to maintaining organic matter, nutrient and moisture content in soil, but it is very dangerous as it may spread disease (Scarlat, Blukdea and Dallemand, 2011). Burning, in addition to being cheap, it is labour-intensive (Magagnotti et al., 2009) and produces significant particulate emissions to the atmosphere (Keshtkar and Ashbaugh, 2007). Alternatively, pruning residues, like other woody biomass from agriculture and forestry, could be used as a fuel to replace fossil oil in electricity generation (Jones et al., 2010) or in small boilers to produce heat (Picchi, Silvestri and Cristoforetti, 2013). In addition, this fuel has a positive energy balance and low emissions, and it is capable of providing great environmental benefits (Gonzalez-Garcia et al., 2014).

MATERIALS AND METHODS

The study examined the effect of rootstock type on the quality and fruit yield of 'Regent' grapevines, as well as leaf area, weight and number of leaves. The grapevines of the varieties studied were grown on seven types of rootstocks, such as 101-14, 125AA, 161-49, 5BB, SO4, SORI, whereas the control was grafted on its own roots.

Figure 1 depicts statistical methods used for the energy and carbon analysis of the raw materials, plant material sampling, and the apparatus used.

RESULTS AND DISCUSSION

The aim of this study was to evaluate physicochemical properties of bio-waste as a potential biofuel in the form of leaves of the 'Regent' grape cultivar grown on six different rootstocks and a control grown on its own roots for three years of cultivation. Table 1 shows effect of rootstock type on selected leaf parameters of 'Regent' grapevines in 3 years of the study.

The number of lateral shoots of 'Regent' grapevines ranged from 17.9 to 18.4 units and did not differ significantly between the rootstock clones evaluated. There was no significant effect of test year on the analysed parameter, as well as the interaction of test year and rootstock type. This trait largely depends on the form of vine management. In the course of research on the evaluation of biomass size of selected grapevine cultivars, no significant effect of cultivar on the number of shoots per plant was shown (Klimek *et. al.*, 2022).

The number of leaves per shoot showed significant differences between the combinations evaluated. Shrubs grafted on 161-49, 101-14 and 5BB rootstocks produced significantly more leaves per shoot than shrubs on 125AA. There was a significant effect of the year of testing on the evaluated parameter. In 2022, plants had significantly more leaves per 1 shoot than in other years. No significant interaction was found



Statistical analysis	Material plants	Energetic properties	
The results obtained in the experiment were statistically analysed using the SAS Enterprise Guide 5.1 software	The Nobilis vineyard (50°39'N; 21°34'E) is located in the south-eastern part of the country in the Sandomierz Upland.	Retsch SM 100 grinder –material pulverisation up to 0.5 mm	
For the obtained results, the normality of the distribution was checked with the Shapiro–Wilk test, then the ANOVA analysis was performed and the significance was assessed with Tukey's HSD test. The analysis of the main components was carried out in order to graphically illustrate the significant differences and similarities of the four grape varieties, regardless of the year of research and energy values	Self-rooted vines of the studied cultivars were planted in the spring of 2010 on loess soil at a spacing of 1.0–2.0 m (5000 pcs. ha ⁻¹).	LECO AC 600 isoperibolic calorimeter –Higher Heating Value (HHV); EN–ISO 1928:2009	
	The plants were grown in the form of a single fixed twine with a trunk 40 cm high and one immobile arm about 0.9 m long, on which.	LECO TGA 701 analyser -Ash content (A); EN-ISO 18122:2016 -Volatile matter content (V); EN-ISO 18123:2016 -Moisture content (M) EN-ISO 18134-3:2015	
	The following parameters were analysed: number and weight of grapes, number and weight of berries and total extract content. The yield and number of grapes per bush were determined by counting and weighing berries from each bush to the nearest 0.001 kg.		
	Average berry weight was determined by weighing and counting berries from five medium- sised clusters, from each repetition.	LECO CHNS 628 analyser -Carbon (C), -Hydrogen (H), -N (nitrogen) content; EN-ISO 16948:2015-07 -Sulfur (S) content EN-ISO 16994:2016-10	
	Fruit extract content was measured using an Abbe refractometer to determine the percentage extract content by squeezing the juice from 20 representative berries from each plant. The experiment was conducted in a randomised block design and included three combinations with five repetitions.		
	The repetitions were plots in which 10 plants were grown.	-Fixed carbon index (FC) = 100%-M-A-V (Choudhury <i>et al.</i> 2021)	
	All annual shoots (epiphylls) were counted on the shrubs included in the experiment in the fall after fruit harvest. In each set, leaves on 50 representative shoots were counted, and their weight including petioles was determined on an AXIS A250 electronic balance with an accuracy of 0.001 kg.	-Oxygen content (O) = 100%-C-H-N-S-A (Alves <i>et al.</i> 2020)	
	Leaves were randomly collected from three locations on the fruiting shoot, for a total of 30 leaves in each repetition. Each sample consisted of 1/3 of the leaves taken at the bottom, 1/3 in the middle and 1/3 at the top of the canopy. Leaf area was estimated using the Area Meter model 3100 on a sample of 30 leaves from each repetition.		
	Based on the results, the number of annual shoots and leaves per plant, the area of 10 leaves and the area of all leaves per 1 hectare, the weight of 10 leaves with and without petioles, and the weight of 10 petioles were determined; parameters determining the weight per hectare are also presented.		

Fig. 1. Test procedure; source: own elaboration

between the year of testing and the type of rootstock. Klimek et al. (2022) showed that this parameter is significantly modified by cultivar and year of testing.

The number of leaves per hectare of 'Regent' grapevines varied from 1,253,383.3 to 1,408,300.0 units, depending on the rootstock type. All rootstock types differed significantly among themselves. The highest number of leaves was in shrubs grafted

on 101-14 rootstock, while the lowest in 125AA. It was not confirmed that the biological factor had a significant effect on the number of leaves per hectare when assessing the number of leaves in grapevines of the 'Regent', 'Seyval Blanc' and 'Solaris' cultivars (Klimek et. al., 2022).

The number of leaves per hectare ranged from 1,140,868.02 m^2 in the 161-49 rootstock to 1,265,286.7 m^2 in the

Factor		Number of shoots per shrub	Number of leaves per shoot	Number of leaves per 1 ha area	Leaf surface per $1 \ln (m^2)$	Leaf mass with petioles per 1 ha area
					I na area (m)	(Mg⋅ha ⁻¹)
Rootstock type (A)	101-14	18.4 ±0.3 ^A	15.3 ±0.8 ^A	1,408,300.0 ±93,773.6 ^A	1,213,498.0 ±1,362,17.2 ^C	$7.6 \pm 0.8^{\mathrm{D}}$
	125AA	18.3 ±0.2 ^A	13.7 ±0.5 ^B	1,253,383.3 ±40,472.7 ^G	1,234,909.6 ±55,571.6 ^B	8.4 ±0.2 ^B
	161-49	18.1 ±0.2 ^A	15.5 ±0.6 ^A	1,397,233.3 ±544,65.6 ^B	1,140,868.0 ±61,324.2 ^G	7.1 ±0.6 ^G
	5 BB	17.9 ±0.2 ^A	15.1 ±0.5 ^A	$1,352,000.0 \pm 46,207.4^{\rm C}$	1,196,330.8 ±52,811.4 ^D	7.2 ±0.5 ^F
	S04	18.1 ±0.1 ^A	14.6 ± 0.7^{AB}	1,318,833.3 ±58,502.8 ^E	1,265,286.7 ±54,183.7 ^A	9.0 ±0.6 ^A
	SORI	18.1 ±0.2 ^A	14.6 ± 0.5^{AB}	1,319,216.7 ±62,096.0 ^D	1,146,907.4 ±11,707.6 ^F	$7.4 \pm 0.5^{\rm E}$
	control	18.0 ±0.2 ^A	14.5 ± 0.5^{AB}	1,302,216.7 ±32,736.8 ^F	1,181,713.7 ±47,101.9	7.9 $\pm 0.4^{\rm C}$
	<i>p</i> -value	0.0527	0.0031	0.0041	0.0051	0.0041
Year (B)	2020	18.1 ±0.2 ^A	14.2 ±0.6 ^B	$1,287,764.3 \pm 49,505.0^{\circ}$	1,152,935.8 ±53,646.8 ^C	7.3 ±0.7 ^C
	2021	18.1 ±0.3 ^A	14.7 ±0.6 ^B	1,326,500.0 ±43,091.2 ^B	1,179,239.5 ±45,117.8 ^B	7.7 ± 0.7^{B}
	2022	18.2 ± 0.3^{A}	15.3 ±0.7 ^A	1,393,385.7 ±76,679.8 ^A	1,259,045.1 ±72,503.5 ^A	8.3 ±0.6 ^A
	<i>p</i> -value	0.7978	0.0021	0.0011	0.0010	0.0010
A×B	<i>p</i> -value	0.9951	0.9967	0.0009	0.0007	0.0101

Table 1. Effect of rootstock type on selected leaf parameters of 'Regent' grapevines in 2020-2022

Explanations: A, B, ..., F in the columns show significant differences at $\alpha = 0.05$, *p*-values in italic = significant values. Source: own study.

SO4 rootstock. All types of rootstocks differed significantly among themselves in leaf area. A significant effect of cultivar on the evaluated parameter was shown in the study of Klimek *et al.* (2022).

The analysed mass of petioled leaves per 1 ha differed significantly among all assessed rootstock types. It was shown that shrubs grafted on 161-49 rootstock (7.1 Mg·ha⁻¹) produced significantly the lowest leaf mass, while those on SO4 rootstock (9.0 Mg·ha⁻¹) produced significantly the highest among all evaluated combinations. A significant effect of cultivar on the evaluated parameter was shown in the study of Klimek *et al.* (2022).

Considering the green weight of leaves (number, area and weight of leaves with petioles per ha), a significant influence of the year of the study was shown. In 2022, all green leaf mass parameters were the highest, while in 2020 they were significantly the lowest. A significant interaction of the year of testing and rootstock type was observed. The influence of the year of testing on the aforementioned parameters was demonstrated by evaluating three grapevine varieties under Polish conditions (Klimek *et al.*, 2022).

Results of higher heating value are presented on Figure 2.

The analyses of the higher heating value (HHV) showed major differences in the rootstocks used in particular years of the study. However, the difference between average values was statistically insignificant. It was found that regardless of the type of rootstock used, the heat from leaf combustion was the highest in 2021. It was shown that there was no clear trend between the



Fig. 2. Results of higher heating value (*HHV*) measurements for the tested leaves of the 'Regent' cultivar; 101-14, 125AA, 161-49, 5BB, SO4, SORI = rootstocks; source: own study

higher heating value in the first and last years of the study. Regardless of the year of the study, vines grafted on 125AA rootstock and control were characterised by the highest heating value, while the 5BB rootstock had the lowest value. Comparing the higher heating values for the leaves from 101-14, 125AA, SO4, SORI rootstocks and the control, it should be noted that there were similarities in the second year of cultivation with the values obtained for pruning vine and wood chips (Torreiro *et al.*, 2020), *Eucalyptus globulus* (Enes *et al.*, 2019), black poplar leaves, oak

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tree leaves, peach tree leaves or pinecone leaf (Güleç *et al.*, 2022). For the first and third year of cultivation regardless of rootstock type, similar results were achieved for barley grain, eucalyptus chips, orange tree leaves (Güleç *et al.*, 2022), sugarcane bagasse, or tea waste (Rahimi, Anand and Gautam, 2022). It can be noted that the levels of higher heating value do not differ from those typically achieved for agricultural or forest biomass.

Results of technical and elemental analysis for the tested leaves of the 'Regent' cultivar are shown Table S1.

The statistical analysis showed no significant effect of the year of the study on the elementary and technical parameters. It was observed that regardless of the parameter and rootstock type the values in 2022 were the highest, while in 2021 they were the lowest. There was no significant effect of rootstock type on V and H parameters. It was observed that the level of the parameters M and A did not depend on the year of the study and it was significantly the highest in leaves from shrubs grafted on 5BB rootstock, while significantly the lowest in the control. An inverse relationship was found for the C content. In the case of the N content, it was observed that it did not depend on the year of testing and the highest values occurred in the case of the 125AA

A principal elemental parameter analysis (Fig. 3a) of the 'Regent' grapevine leaves oculised on six types of rootstocks and own-root vines (control) allowed to separate three clusters. The first cluster consists of rootstock 101-14 and control, and subcluster 125AA. The next two clusters consist of leaf biomass of vines grafted on SO4 and SORI rootstocks, as well as 5BB and 161-49. The next dendogram of technical principal component analysis (Fig. 3b) of 'Regent' grapevine leaves allowed two separate clusters to be distinguished. The first cluster included the leaf biomass of 101-14 rootstocks and controls, while the next cluster consisted of the other rootstock types. When considering elemental and technical analyses, similarity was observed in the first cluster consisting of leaf biomass of 101-14 rootstock and control (ownrooted shrubs). Despite the fact that the number of leaves per shrub in the above-mentioned plants differed significantly, to the extent that shrubs grafted on 101-14 rootstock produced significantly the largest volume of leaves, while the own-rooted ones produced significantly the least volume of leaves among all the combinations evaluated. For the other parameters determining biomass, i.e. leaf mass and leaf area, no clear relationship was observed with elemental and technical parameters (Fig. 3).



Fig. 3. Principal component analysis of leaves of 'Regent' grapevines oculised on six types of rootstocks (101-14, 125AA, 161-49, 5BB, SO4, SORI) and own-rooted vines (control): a) elemental (elementary parameters: carbon content (C), hydrogen content (H), nitrogen content (N) and sulphur content (S)), b) technical parameters: moisture content (M), volatile matter content (V), and ash content (A); source: own study

rootstock, while the lowest in the case of the 5BB rootstock. Regardless of the year of study, significantly the smallest value of the S content was obtained from leaves derived from the 101-14 rootstock. In the case of 125AA, SORI and 161-49 rootstocks, the values were significantly the highest not depending on the year of the study, while in the case of the SO4 rootstock in the first and last year, and in the case of 5BB in 2022. The ash content for the tested raw materials was at a fairly high level. Similar results were achieved for apple tree leaves, peach tree leaves (Güleç et al., 2022), grapevine leaves, lemon leaves, plum leaves or raspberry leaves (Vassilev et al., 2017). As regards agrobiomass, the same levels were recorded for corn stover, sunflower pressed bagasse, and wheat husk (Rahimi, Anand and Gautam, 2022). Carbon, nitrogen and hydrogen contents were similar for pruning vine, pruning kiwi (Torreiro et al., 2020) apple tree leaves, cherry tree leaves, hazelnut tree leaves (Güleç et al., 2022), but significantly higher levels were recorded for sulphur than those obtained for the aforementioned biomass. Figure 3 shows results of the principal component analysis for leaves of 'Regent' grapevines.

CONCLUSIONS

In most of the studied parameters, tests carried out for the leaves of grapes of the 'Regent' cultivar showed no significant differences depending on the year of cultivation. Hence, when considering the possibility of obtaining additional raw material as biomass for energy purposes, the year of cultivation does not play a role in shaping the energy potential, no less it affects the amount of raw material obtained. Instead, the energy potential is influenced by the type of rootstock used in cultivation. The analysis of results obtained showed that the 101-14 rootstock, 125AA and the control have the highest leaf energy potential considering higher heating value (HHV), while the lowest value applies to cultivation on the 5BB rootstock. Hence, the energy bio-waste management should be based on both the amount of available biomass and its energy potential. In addition to yield, the optimal choice would be to recommend cultivation on the SO4 rootstock, for which both the highest weight of leaves with petioles from the growing area $(9.0 \text{ Mg} \cdot \text{ha}^{-1})$ and high heat of combustion of $18.4 \text{ MJ} \cdot \text{kg}^{-1}$ have been observed for leaves of the 'Regent' cultivar.

SUPPLEMENTARY MATERIAL

Supplementary material to this article can be found online at: https://www.jwld.pl/files/Supplementary_material_Kaplan.pdf.

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