

# Crop residues and agro-industrial by-products from the province of La Rioja (Argentina) suitable for oyster mushroom culture

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DOI: 10.31047/1668.298x.v39.n2.36768

## SUMMARY

In La Rioja province (Argentina) large amounts of lignocellulosic residual biomass are generated annually. A survey was conducted to identify crop residues and agro-industrial by-products in the province, which could be suitable for oyster mushroom culture. Their morphological composition was characterized and a proxy for the mass that is generated annually by each of them was estimated. The availability of the biomass was also evaluated by assessing the processing, distribution, seasonality, cost and other common uses that may compete with its application as substrate. Eight residual crops and eight agro-industrial by-products were identified, being olive and grape pomace the most abundant, followed by grape pruning, and olive and jojoba litter. The use as substrate of these last two together with *Prosopis* bran and olive pits, has not been registered in the production of oyster mushrooms. In production assays with commercial strains of *Pleurotus* species, biological efficiency (BE) ranged from 28% to 72%, depending on the substrate and fungal species. In a context of high-quality food production deficit and nutritional problems related to it, this province has an interesting potential to introduce the production and consumption of oyster mushrooms as a feasible solution for food regional supply.

**Keywords:** agro-waste, residual biomass

Fracchia, S., Miranda, V., Sede S., Barbero, I., Barros, J. y Delgado, N. (2022). Residuos de cultivos y subproductos agroindustriales de la provincia de La Rioja (Argentina) aptos para el cultivo de hongos ostra *Agriscientia* 39 (2): 29-43

## RESUMEN

En la provincia de La Rioja se generan anualmente grandes cantidades de biomasa residual lignocelulósica. Se realizó un relevamiento para identificar los

residuos y subproductos agroindustriales de la provincia aptos como sustrato para el cultivo de hongos ostra. Se caracterizó su composición morfológica y estimamos la masa que generan anualmente. También se determinó la disponibilidad de la biomasa, evaluando su procesamiento, distribución, estacionalidad, costo y otros usos que compiten con su aplicación como sustrato. Se identificaron ocho cultivos residuales y ocho subproductos agroindustriales, siendo los más abundantes el olivo y el orujo de uva, seguidos por la poda de uva, y la hojarasca de olivo y jojoba. Estos dos últimos junto con el salvado de *Prosopis* y el hueso de aceituna, no registraron uso como sustrato para la producción de hongos. En ensayos de producción con cepas de *Pleurotus*, registramos valores de eficiencia biológica (BE) en un rango de 28% a 72% según el sustrato y la especie fúngica. En un contexto de déficit en la producción de alimentos de alta calidad y problemas nutricionales, la provincia tiene un interesante potencial para la producción y consumo de hongos ostra como una solución factible para el abastecimiento alimentario regional.

**Palabras clave:** residuos agrícolas, biomasa residual

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## INTRODUCTION

Oyster mushroom cultivation has been adopted in many regions around the world, constituting a low-cost nutraceutical food and an alternative for underdeveloped regions where the production and availability of quality food is limited (Kimenju et al., 2009; Ishara et al., 2018). As a food source, *Pleurotus* species provide well-demonstrated benefits that include an excellent source of protein, essential amino acids, dietary fiber, minerals, and vitamins (Jedinak et al., 2011; Khan and Tania, 2012). They also produce significant health benefits by strengthening the immune

system, providing antioxidant and antitumor metabolites, antihyperlipidemic effects, as well as anti-inflammatory effects among several others (Deepalakshmi and Mirunalini, 2008; Yan and Chang, 2012; Adebayo and Oloke, 2017).

A key factor for the introduction of oyster mushroom culture in marginal underdeveloped regions is to have access to suitable biomass generated *in situ*, and available in sufficient quantities at no cost for this purpose (Paulraj and Francois, 1995; de León-Monzón et al., 2004). A huge number of lignocellulosic residues have been tested as substrates for the cultivation of *Pleurotus* species worldwide (de Carvalho et

al., 2010; Stamets, 2011; Mahari et al., 2020). In tropical Latin America, local residues as banana leaves, coffee pulp, maize stubble, sugarcane and agave bagasses are currently used in family farming for the cultivation of different species of the genus *Pleurotus* (Bermúdez et al., 2001; Bernabé-González et al., 2004; García-Oduardo et al., 2006; Vega et al., 2006). Many successful experiences of introducing and adapting oyster mushroom farming in unfavorable or remote areas are well documented also in Africa and South Asian countries (Mabuza et al., 2012; Tesfaw et al., 2015; Bandara et al., 2021).

In Argentina, oyster mushroom cultivation was introduced in the 1980s, growing steadily in the following decades, being today a well-established activity (Lechner and Albertó, 2011; Rugolo et al., 2020). Diverse agro wastes are commonly used as substrates, both crop residues and agro-industrial by-products generated mainly from extensive annual crops in the Pampas region; some of them are wheat, peanuts, corn and sunflower (Curvetto et al., 2002; Ravera et al., 2008; Mejía and Albertó, 2013). Actually, in our country most commercial culture of oyster mushroom is carried out on wheat straw (a substrate that is available in the Pampas region), and it is concentrated near big cities to have direct access to the markets. However, a large amount of lignocellulosic residual biomass is also produced annually in other regions of the country, such as those generated from crops located in the central and northwest Andean provinces. These provinces are characterized by an agricultural production dominated by perennial crops as vine, olive, walnut and fruit trees, jojoba, and the recently introduced pistachio (Secretaría de Agricultura, Ganadería y Pesca, 2021). Annual crops such as corn, tomato and chili are grown mostly on a small scale by family farmers. All these crops annually generate a big amount of crop residues and agro-industrial by-products which are not incorporated into any industrially profitable process (Rodríguez et al., 2018). Although there are attempts to incorporate this biomass in technological processes (Marco et al., 2010; Sardella et al., 2015; Giménez et al., 2020), to date only a small proportion of grape and olive pomace are composted, or exploited as food for goats, cows and pigs by local farmers. Some experimental trials have been carried out to incorporate them into the cultivation of other edible mushrooms than the *Pleurotus* species (Rugolo et al., 2016). Therefore, a significant volume of residual biomass is discarded annually in landfills or burned, generating greenhouse gases.

The identification and characterization of crop residues and agro-industrial by-products, suitable

to use as oyster mushroom substrate in the Andean province of La Rioja, were included. The spatial distribution where the residual biomass is generated was surveyed, a proxy for the annually generated mass was estimated, and the availability, accessibility and management for its use were studied. In the case of specific residual biomass without reference as substrate for oyster mushroom culture, an evaluation of the biological efficiency (BE) in experimental conditions was carried out. Our work is a first step to introduce oyster mushroom culture in a region that has growing obstacles related to food production, such as a sharp decrease in access to water for irrigation, reduction of arable land due to an increase of desertification, and high rates of morbidity in the population related mainly to deficiencies in the access to quality food (Observatorio Nacional de Degradación de Tierras y Desertificación).

## MATERIALS AND METHODS

### Identification and characterization of crop residues and agro-industrial by-products from La Rioja province

For the identification of the residual biomass generated in the province, information was sought from the Secretaría de Agricultura de La Rioja (Agriculture Department of La Rioja) and the Ministry of Agriculture, Livestock and Fisheries of the Nation (MAGyP). These institutions have no statistical information available on the nature and amounts of residual biomass generated in the province, although there is data on the crops and by-products generated from their processing (Instituto Nacional de Estadística y Censos [INDEC], 2018; Secretaría de Agricultura, Ganadería y Pesca, 2021). From these sources were collected all the information on the crops currently grown in the province, evaluating the kind of crop residue or agro-industrial by-product generated in each of the 18 provincial departments. Additionally, field trips to all provincial departments enabled the direct *in situ* collection of data to confirm the nature and amount of the residual biomass that is generated locally. There are specific regions where there is no data at all, or it is highly limited, confusing, or outdated; therefore, much information was collected directly from personal producers and processing plants.

The morphological composition of each residual biomass was evaluated by measuring the percent dry weight of plant morphological structures included in the sample. For this purpose, the different structures were separated with the help

of a binocular loupe and a tweezer, and then weighed separately. When possible, an average of several samples from different origins were taken, considering that the management and harvest of the crops may vary, as well as the processing in industrial plants (i.e. olive mills, jojoba mills). A proxy for the physicochemical characterization of each residue was compiled from available online updated bibliography. This criterion was taken because there is a great variety within each residual biomass, taking into account that there are a high number of varieties of each crop in the province, as well as diversity in the processes from which agro-industrial residues are obtained.

### Residual biomass production and spatial distribution at a provincial scale

To have a proxy for each annual residual biomass suitable for oyster mushroom culture generated in the province, an estimation was made based on the local crops and processing plants throughout the provincial territory. Crop residues were calculated from the cultivated area, considering the formula  $P_b = a \cdot R$ , where  $P_b$  is the potential biomass in tons produced annually for a given crop residue,  $a$  is the cultivation areas in hectares, and  $R$  is the residues biomass generated in tons per unit of area (hectare) for each specific crop (Martin, 2014). The cultivation area was compiled from official information in most cases, considering the annual average of the planted hectares in the last 5 years for annual crops, when the information was available. Specific  $R$  values for each crop were obtained from the bibliography in most cases, and calculated for quinoa stubble and jojoba litter by weighing samples per square meter (quinoa stubble) or weighing the litter per plant (jojoba), extrapolating the data to the density of plants per hectare.  $R$  value for each crop residue is showed in Table 2 with their references. In the case of agro-industrial by-products, information was collected from websites of official institutions, published data, or directly from processing plants in the province (i.e. wineries, olive and jojoba mills). An estimated range between maximum and minimum values produced in the last 5 years was calculated, when information was available (Varas et al., 2014; Vuksinic and Miguel, 2018). Crop residues and agro-industrial by-products distribution are shown in the provincial map (Figure 1), displaying their geographical distribution in the 18 political departments of La Rioja.

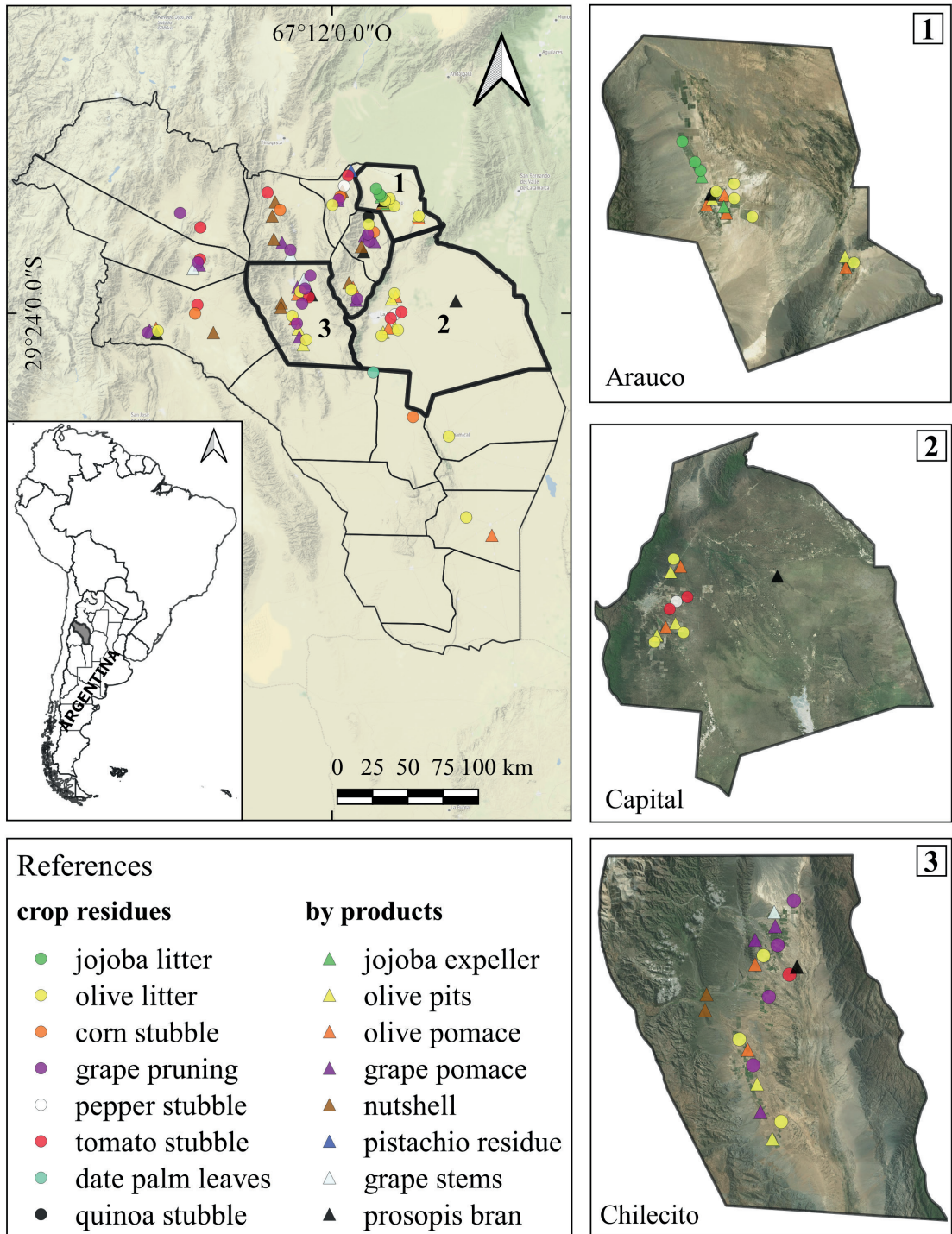
### Assessment of residual biomass according to

#### its availability

To evaluate the availability of each of the residual biomass identified in the province, four main variables were considered: 1) processing and stowage, according to whether biomass requires processing before storage and posterior application or it is directly collected from the field without further processing; 2) availability over time, taking into account that there is biomass that is generated seasonally; thus, it can be used for other purposes or degrades and loses its ability to be used as a substrate; 3) monetary cost, whether biomass is free of charge or not; and 4) other common uses that compete for the availability of the residual biomass. This qualitative information was collected mainly from oral communications from government agencies (INTA, SENASA) and field trips and direct observation.

#### Mushroom culture

From the residual biomass identified in the province, information was compiled about its previous utilization as substrate for the cultivation of oyster mushrooms. Data of *Pleurotus* species involved, BE values, and the use of pure or mixed substrates were included (Table 4). For those crop residues and agro-industrial by-products without record as substrates (jojoba litter, olive litter, olive pits, *prosopis* bran), experimental cultures were performed with two commercial strains *Pleurotus ostreatus* (BAFC1723) and *Pleurotus djamor* (BAFC1765) (Buenos Aires Fungal Collection). The selection of these two species of *Pleurotus* for experimentation is due to their different adaptation to temperature, *P. djamor* strain being more tolerant to high temperatures and *P. ostreatus* strain more efficient at lower temperatures, which allows an alternate production according to the seasons of the year. The biological efficiency assay was carried out, cultivating both species in 5-liter plastic containers (Figure 2F, 2G and 2H). The substrates were obtained from local farmers (olive litter and *Prosopis* bran), and from jojoba and olive agricultural establishments (jojoba litter, olive pits) in La Rioja province. Each substrate was hydrated overnight in a plastic container, washed with tap water, and then pasteurized in polypropylene bags at 80 °C for 2 hours. After cooling at room temperature, each substrate was mixed with the inoculum of each fungus formulated in millet seeds at 5 % V/V and added with 2 % of CaCO<sub>3</sub>. The plastic containers sealed with the caps, but not hermetically, were incubated at 28 °C in the dark for 21 days, and then exposed to light (800 lux) at room



**Figure 1.** Map of the province of La Rioja (Argentina) showing the location of eight crop residues and eight agroindustrial by-products, with detail of the three departments where the highest amount of residual biomass is produced



**Figure 2.** Crop residues, agro-industrial by-products and mushroom culture system. A, grape pomace accumulated in a winery during the mill season; B, jojoba expeller mounds left at the edge of the provincial roads; C, olive pomace in landfills constructed exclusively to discard this agro-industrial by-product; D, grape pruning left at the side of the vineyards; E, jojoba leaves mounds close to the crop fields; F, substrate hydration in plastic containers to evaluate oyster mushroom production; G-H, *P. ostreatus* and *P. djamor* growing in jojoba litter as substrate

temperature (22-25 °C) and 80 % humidity, allowing successive fructifications up to 53 days. The BE was calculated as: weight fresh basidiomes x 100/ weight dry substrate, considering all production flushes throughout the production period. Finally, a qualitative assessment of the size, aroma, taste and general appearance of the basidiomes was carried out. Five replicates for each substrate and fungal treatment were performed.

## RESULTS AND DISCUSSION

### Identification and characterization of crop residues and agro-industrial by-products from La Rioja province

A total of 16 residues of lignocellulosic nature suitable for oyster mushroom production were identified in the province of La Rioja (Table 1): eight crop residues and eight agro-industrial by-products. An estimated mass of 75,000 tons comprising

these 16 types of residual biomass is generated annually, which is not incorporated into any type of biotechnological process. The identification and quantification of residual biomass is still a pending issue in Argentina. Regarding the proportions of plant structures included in each residue, large variations among samples collected from different origins was not observed. Most of the residual biomass is a mixture of structures, mostly stem and leaves in crop residues, and fruit/seeds structures in agro-industrial by-products. On the other hand, all the residues are generated by a single crop species, except for the *Prosopis* bran, which is generated by pod structures of two species of the genus *Prosopis*. Particularly in La Rioja province, the most exploited species for the production of flour are *Prosopis flexuosa* and *Prosopis chilensis* (Biurru et al., 2007). From a total of 16 identified residues, it was possible to gather information regarding the physicochemical characterization of 14 of them (Table 1). Only *Prosopis* bran and jojoba litter lacked any references in the literature.

**Table 1.** Crop residues and agroindustrial by-products identified in La Rioja province, the crop and plant species that originate them, morphological structures involved and references to their physico-chemical characterization

| Residual biomass  | Crop residue/ by product | Crop        | Species                         | Crop Dynamic/ Habit | Morphological structures* | Physico-chemical references |                               |
|-------------------|--------------------------|-------------|---------------------------------|---------------------|---------------------------|-----------------------------|-------------------------------|
| tomato            | crop residue             | tomato      | <i>Lycopersicon</i>             | annual              | stems                     | 90-95                       | Sánchez et al. (2008)         |
| stubble           |                          |             | <i>esculentum</i>               |                     | leaves                    | 5-10                        |                               |
| pepper            | crop residue             | chilli      | <i>Capsicum</i>                 | annual              | stems                     | 90-95                       | Forero et al. (2008)          |
| stubble           |                          |             | <i>annuum</i>                   |                     | leaves                    | 5-10                        |                               |
| corn stubble      | crop residue             | corn        | <i>Zea mays</i>                 | annual              | stems                     | 80-90                       | Zárate-salazar et al., (2020) |
| quinoa            | crop residue             | quinoa      | <i>Chenopodium</i>              | annual              | leaves                    | 10-20                       | Toledo Alvarez (2010))        |
| stubble           |                          |             | <i>quinoa</i>                   |                     | stems                     | 80-90                       |                               |
| grape pruning     | crop residue             | grapevine   | <i>Vitis vinifera</i>           | perennial           | leaves                    | 10-20                       | Dang et al. (2015)            |
| olive litter      | crop residue             | olive       | <i>Olea europaea</i>            | perennial           | Branches                  | 100                         |                               |
|                   |                          |             |                                 |                     | leaves                    | 80-90                       | Aydino lu and Sargin (2013)   |
|                   |                          |             |                                 |                     | stems                     | 10-20                       |                               |
|                   |                          |             |                                 |                     | leaves                    | 60-80                       |                               |
| jojoba litter     | crop residue             | jojoba      | <i>Simmondsia</i>               | perennial           | stems                     | 15-30                       | not available                 |
|                   |                          |             | <i>chinensis</i>                |                     | pericarp                  | 5-20                        |                               |
|                   |                          |             |                                 |                     | seeds                     | 3-10                        |                               |
| date palm leaves  | crop residue             | date        | <i>Phoenix</i>                  | perennial           | leaves                    | 100                         | Kabirifard et al (2012)       |
|                   |                          |             | <i>dactylifera</i>              |                     | pericarp                  | 30-50                       |                               |
| grape pomace      | by product               | grapevine   | <i>Vitis vinifera</i>           | perennial           | seeds                     | 45-60                       | Stamets (2011)                |
|                   |                          |             |                                 |                     | stems/leaves              | 3-10                        |                               |
|                   |                          |             |                                 |                     | seeds                     | 40-60                       |                               |
| olive pomace      | by product               | olive       | <i>Olea europaea</i>            | perennial           | pericarp                  | 40-70                       | Stamets (2011)                |
|                   |                          |             |                                 |                     | stems/leaves              | 3-5                         |                               |
| jojoba expeller   | by product               | jojoba      | <i>Simmondsia</i>               | perennial           | seeds                     | 100                         | Gayol et al. (2009)           |
| olive pits        | by product               | olive       | <i>Olea europaea</i>            | perennial           | seeds/endocarp            | 100                         | Ghanbari et al. (2012)        |
| pistachio residue | by product               | pistachio   | <i>Pistacia vera</i>            | perennial           | epicarp/mesocarp          | 100                         | Demiral et al. (2008)         |
| grape stems       | by product               | grapevine   | <i>Vitis vinifera</i>           | perennial           | stems                     | 95-100                      | Pardo et al. (2007)           |
|                   |                          |             |                                 |                     | pericarp                  | 3-10                        |                               |
| prosopis bran     | by product               | algarrobo** | <i>P. flexuosa/P. chilensis</i> | perennial           | pericarp                  | 70-85                       | Díaz-Batalla et al. (2018)    |
|                   |                          |             |                                 |                     | seeds                     | 15-20                       |                               |
| nutshell          | by product               | walnut      | <i>Juglans regia</i>            | perennial           | endocarp                  | 100                         | Zhu and Xiong (2007)          |

\*(perennial/annual) \* % dry weight, \*\* natural populations

Basic and reliable information regarding nature, amounts, geographical or temporal availability of residual biomass is not generally accessible for most provinces (Roberts et al., 2015; Fermanelli et al., 2020). It is clear that it is not an easy task, since the residual biomass is dispersed in public and private farms, mills and other industries from provincial and national organizations. However, other countries have made accurate estimates of residual biomass, mainly focused on its use for renewable energy generation (Ferreira-Leitao et al., 2010; Lozano-García et al., 2020).

### Residual biomass production and spatial distribution at a provincial scale

A proxy mass of 75,000 tons of potential residual biomass suitable for oyster mushroom culture is produced annually in La Rioja province (Table 2). Crop residues of grape pruning and olive litter, and the agro-industrial by-products of olive and grape pomace, accounted for more than 75 % of the total residual biomass. The olive pomace is the largest residue produced in the province with an average of 20,000 tons per year (Vuksinic and Miguel, 2018).

**Table 2.** Estimated annual amounts of crop residues and agroindustrial by-products suitable for oyster mushroom culture in La Rioja province

| Residual biomass  | R value* (tn/ha/year) | Potential biomass (tn/year) * | References                     |
|-------------------|-----------------------|-------------------------------|--------------------------------|
| tomato stubble    | 1.4                   | 560                           | Ali Hamamre et al., (2017)     |
| pepper stubble    | 1.7                   | 680                           | Roberts et al., (2015)         |
| corn stubble      | 2.2                   | 2200                          | Torma et al., (2018)           |
| quinoa stubble    | 2.5                   | 25                            | This work                      |
| grape pruning     | 2.0                   | 16000                         | Velázquez Martí et al., (2011) |
| olive litter      | 0.8                   | 19200                         | García-Maraver et al. (2012)   |
| jojoba litter     | 1.2                   | 3360                          | This work                      |
| date palm leaves  | 2.0                   | 40                            | Ali, (2010)                    |
| grape pomace      | -                     | 5000-8000                     | -                              |
| olive pomace      | -                     | 15000-25000                   | -                              |
| jojoba expeller   | -                     | 3000-5000                     | -                              |
| olive pit         | -                     | <50                           | -                              |
| pistachio residue | -                     | <25                           | -                              |
| grape stems       | -                     | <25                           | -                              |
| prosopis bran     | -                     | <25                           | -                              |
| Nutshell          | -                     | <25                           | -                              |

\*(perennial/annual) \*dry weight basis

In the case of annual crops, the cultivated area is highly variable over the years, mainly consisting of tomato, pepper, and corn crop. However, these crops generate only a minor fraction compared with the residues registered for vine, olive or jojoba. The largest biomass is concentrated in three departments: Arauco, Chilecito and Capital (Figure 1). The first department concentrates the production of jojoba, the second the production of vine, while the olive growing area is distributed equally on the three ones. The departments of Felipe Varela, San Blas de los Sauces and Famatina, also generate significant amounts of olive and grape biomass residues, including crop residues and agro-industrial by-products. Other residues such as corn stubble or nutshell, although with much smaller production quantities, are distributed in several departments (Figure 1). While *Prosopis* species are well distributed in all the province, their harvest and processing are carried out in only a few locations by family farmers. Pistachio and date palm residues are produced in a single location, meanwhile quinoa has been recently introduced in the province. Currently, family farmers cultivate this seed on a few hectares in two departments with the aim of evaluating its productivity. The southern departments have extreme climatic conditions with high temperatures and low precipitation rates, which allows the cultivation of corn on a family scale, goat pasture with buffel grass, and small-surface olive groves. The Andean departments are sparsely populated and are the only provincial departments with permanent rivers. They are characterized by a reduced agricultural production: they generate a small amount of crop residues from corn, pepper and tomato, in addition to crop residues from vineyards and olive orchards. To have a theoretical approximation of residual biomass generated in the province, R values from published sources were considered and when the information was not available, they were estimated (Roberts et al., 2015). However, these values must be adjusted to local conditions to have a more accurate data on the residual biomass that is generated by each crop. For example, the mass of vine pruning produced per hectare over a year depends on the type of vine, age, way of harvesting, configuration of terrain, edaphoclimatic variables and crop management among others. Overall, the R values can range between 1 and 5 tons per hectare (Puglia et al., 2017). Thus, these values should be adjusted for a particular region and its variation should be projected according to multiple factors in order to have an accurate residual biomass estimation. On the other hand, the generation of specific residual biomass could



be highly oscillating over the seasons (Breunig et al., 2018; Honorato-Salazar and Sadhukhan, 2020). An exceptional case is the olive tree, followed by pistachio, which has years of high production alternating with years of low, generating fluctuation in the amounts of agro-industrial by-products (Lavee, 2007). Moreover, annual and perennial crops vary according to annual climatic conditions, and also to market values, having years when harvests are not achieved due to drops in the commercialization values of the commodities. In the same way, the cultivated area of annual species can change between years, as in the case of tomato and pepper in La Rioja, with years that can quadruple the area of previous seasons. In this context, our proxy values can be highly variable, taking into account crop dynamics and the fluctuating productive and commercial conditions in the province. However, there are residual biomasses such as jojoba and olive litter, grape pruning or *Prosopis* bran, which

are generated annually in abundance and are not subjected to the variables mentioned above. On the other hand, although olive and grape pomace as well as olive pits can vary significantly, there is a basal production that ensures that this biomass is always available.

### Assessment of residual biomass according to its availability

Two crop residues (jojoba and olive litter) and two agro-industrial by-products (*Prosopis* bran and grape stems) do not need any kind of processing to be used as substrate (Table 3). In general terms, crop residues need to be collected from the field and chopped prior to be used for mushroom culture, while agro-industrial by-products are generally concentrated in the processing factories (jojoba, olive, grapes) and must be dried after its collection for long term storage. Nutshell is the only by

**Table 3.** Crop residues and agroindustrial by-products from La Rioja province, main variables that determine its availability as a lignocellulosic substrate for oyster mushroom culture

| Residual biomass  | Processing and stowage                   | Seasonality (months) | Cost*    | Alternative uses                             |
|-------------------|--|----------------------|----------|--|
| tomato stubble    | field collection<br>chopped up<br>drying | December-March       | free     | none   |
| pepper stubble    | field collection<br>chopped up<br>drying | February, March      | free     | none   |
| corn stubble      | field collection<br>chopped up           | January-May          | free     | none   |
| quinoa stubble    | field collection<br>chopped up<br>drying | March, April         | free     | none   |
| grape pruning     | field collection<br>chopped up<br>drying | July, August         | variable | home fuel                                    |
| olive litter      | field collection                         | all year             | free     | cattle feed,<br>compost                      |
| jojoba litter     | field collection                         | all year             | free     | compost                                      |
| date palm leaves  | field collection<br>chopped up           | all year             | free     | none   |
| grape pomace      | drying                                   | February-April       | free     | cattle feed,<br>compost                      |
| olive pomace      | drying                                   | all year             | free     | cattle feed,<br>compost,<br>rural road cover |
| jojoba expeller   | drying                                   | all year             | variable | fuel<br>industrial use                       |
| olive pit         | washing<br>drying                        | all year             | variable | home fuel,<br>rural road cover               |
| pistachio residue | drying                                   | April                | free     | none   |
| grape stems       | none                                     | all year             | free     | none   |
| prosopis bran     | none                                     | January-April        | free     | cattle feed                                  |
| nutshell          | chopped up                               | March-May            | variable | home fuel                                    |

\* variable: eventual commercialization

product that needs to be processed to obtain a finer texture required for oyster mushroom production. Regarding temporary availability, half of the agro-industrial by-products are available all year round, as they accumulate in landfills where it naturally dries up due to the high temperatures and low humidity in the region. Crop residues such as corn, tomato and pepper stubble, the agro-industrial by-products from grape pruning, *Prosopis* bran, and nutshell are available for a limited time, since they are exploited mostly as cattle feed, or burned *in situ* or used as home bio fuel. Only four residues that are occasionally traded were registered. Nutshell, olive pits and grape pruning are good fuels, and eventually these could be marketed informally at low cost for home fuel or small industrial boilers. Only the jojoba expeller can be marketed for the industrial extraction of residual waxes, leaving this biomass in other provinces or countries. The jojoba litter is a very interesting crop residue that is collected together with the seed harvest and is discarded in large piles in the jojoba wax extraction mills. In the Arauco department, more than 3,000 tons of jojoba litter per annum are estimated to be accumulated in the fields or straightway burned. An interesting point is that this crop can be productive for more than 100 years, warranting larger amounts of lignocellulosic biomass over time (Reddy and Chikara, 2010). By contrast, the palm leaf is a variable and heterogeneous lignocellulosic residue in the province, since there is only one palm farm that was practically abandoned several years ago, leaving today only a fraction of the originally cultivated palms. However, there are ornamental and naturalized plantations of other palm species such as *Phoenix canariensis* and *Washingtonia filifera* in different locations of the province, which could also be used to process their leaves as substrates for oyster mushroom culture. Other palm species have also been used to formulate substrates for the cultivation of other mushrooms; Ali et al. (2018) and Cogorni et al. (2014) have valued these species as an interesting alternative for this purpose. Although our work was focused on La Rioja province, other Andean provinces such as Mendoza, San Juan, Catamarca, and Salta have a similar agricultural production that generate thousands of tons of some crop residues and agro-industrial by-products identified in our study (Marco et al., 2010; Cáceres et al., 2012). More than 90 % of olive orchards, vineyards and walnut cultivated in Argentina are located in these provinces; while jojoba, pistachio and date palm crops are exclusively settled in this region. These five provinces occupy a territory of 585,000 km<sup>2</sup> with a population of approximately 4.5 million,

which highlights the potential of the great amount of residual biomass for oyster mushroom cultivation in this region.

### Mushroom culture

Only four of the identified residues did not register use as substrate for oyster mushroom culture: jojoba litter, olive litter, olive pits and *Prosopis* bran (Table 4). The remaining 12 residues were previously used as substrates, pure or combined with other lignocellulosic ones, mostly for the culture of *P. ostreatus*, *P. djamor* and *P. pulmonarius* species. Regarding production assays of the four substrates without previous references, our results indicated variability among the evaluated residues. *Prosopis* bran reached the highest values of biological efficiency (> 70 %), while olive litter and olive pits showed the lowest ones (< 40 %) for *P. ostreatus* and *P. djamor* species. Intermediate values of BE (near 50 %) were obtained with the jojoba litter for both *Pleurotus* species (Table 4). Two flushes were recorded for olive litter and at least three for the rest of the evaluated residues (jojoba litter, olive pits and *Prosopis* bran). Fungal fructifications with good appearance were harvested in all treatments, with size, consistence, odor and flavor similar to those produced with standard substrates as wheat straw (Figure 2 G, H). In the present work the BE values fluctuated between 28 % and 72 % for the four substrates evaluated. Although pure substrates were used to obtain reference values, it is well recognized that BE can be improved when adding supplements or formulating mixed substrates. Previously, the jojoba expeller had proved to be an efficient supplement to improve the BE of *P. ostreatus* using *Jatropha macrocarpa* residues as substrate (Fracchia et al., 2009). An improvement can also be carried out by supplementing with soy flour, brewing bagasse, wheat bran or rice bran, among others (Jafarpour et al., 2010; Luque et al., 2021).

Reference values of BE for each crop residue and agro-industrial by-product showed contrasting values according to different authors and the methodology adopted (Piña-Guzmán et al., 2017). Variables such as the quality of the residual biomass, fungal strain involved, or culture conditions can significantly modify the BE values. In the present work, mean values were used as a reference for each of the residual crops and agro-industrial by-products identified in La Rioja province. An important step to advance in the oyster mushroom production of the region should focus on trials of mixed substrates, in order to make

**Table 4.** Crop residues and agroindustrial by products from La Rioja province and references to their use as substrate for oyster mushroom culture

| Residual biomass  | Mushroom culture | Pleurotus species     | BE*         | References                     |
|-------------------|------------------|-----------------------|-------------|--------------------------------|
| tomato stubble    | yes              | <i>P. ostreatus</i>   | 11 % P      | Sanchez et al., (2008)         |
|                   |                  | <i>P. pulmonarius</i> | 139 % P     |                                |
| pepper stubble    | yes              | <i>P. ostreatus</i>   | 31-45 % M   | Forero et al., (2008)          |
| corn stubble      | yes              | <i>P. ostreatus</i>   | 85-105 % P  | Zárate-Salazar et al., (2020)  |
| quinoa stubble    | yes              | <i>P. ostreatus</i>   | 88-97 % P   | Rodríguez Caba, (2020)         |
| grape pruning     | yes              | <i>P. ostreatus</i>   | 55 % M      | Dang et al., 2015              |
|                   |                  | <i>P. ostreatus</i>   | 32 % P      |                                |
| olive litter      | no               | <i>P. djamor</i>      | 28 % P      | this work                      |
|                   |                  | <i>P. ostreatus</i>   | 58 % P      |                                |
| jojoba litter     | no               | <i>P. ostreatus</i>   | 45 % P      | this work                      |
|                   |                  | <i>P. djamor</i>      | 45 % P      |                                |
| date palm leaves  | yes              | <i>P. ostreatus</i>   | 12-51 % M   | Owaid et al., (2015)           |
| grape pomace      | yes              | <i>P. ostreatus</i>   | 16 % P      | Papadaki et al., (2019)        |
|                   |                  | <i>P.pulmonarius</i>  | 31 % P      |                                |
| olive pomace      | yes              | <i>P. ostreatus</i>   | 33 % P      | Mansour–Benamar et al., (2013) |
| jojoba expeller   | yes              | <i>P. ostreatus</i>   | 90 % M      | Fracchia et al., (2009)        |
| olive pits        | no               | <i>P. ostreatus</i>   | 38 % P      | this work                      |
|                   |                  | <i>P. djamor</i>      | 30 % P      |                                |
| pistachio residue | yes              | <i>P. ostreatus</i>   | 112-129 % M | Pardo-Giménez et al., (2016)   |
| grape stems       | yes              | <i>P. ostreatus</i>   | 45 % P      | Pardo et al., (2005)           |
| prosopis bran     | no               | <i>P. ostreatus</i>   | 72 % P      | this work                      |
|                   |                  | <i>P. djamor</i>      | 50 % P      |                                |
| nutshell          | yes              | <i>P. ostreatus</i>   | 8 % P       | Sözbir et al., (2015)          |

\*P: used as pure substrate, M: mixed substrates

the production more efficient, improving BE values. The sixteen types of residual biomass allowed us to project a range of possibilities for each region of the province, taking into account the distribution and availability of each one of them in the different provincial departments. Currently, other substrate suitable for oyster mushroom culture available in La Rioja province is the forage crop buffelgrass (*Cenchrus ciliaris*). This grass species was introduced in the 1980s as fodder for goats, it later spread beyond the farms, invading large areas of the region (Blanco et al., 2005; Guevara et al., 2009). Its suitability for oyster mushroom culture was assessed, and it proved to be a suitable substrate with a BE above 65 % for *P. ostreatus* and *P. djamor* (personal communication with Delgado N.). Khare et al. (2015) obtained higher BE values (>80%) with this substrate supplemented with cowpea pods or wheat bran, meanwhile Gaitán-Hernández (1993) recorded lower BE values using buffelgrass straw with other supplements. However, this crop does not leave an agricultural residue or generate an agro-industrial by-product, thus, in the present work this residual crop has not been considered as available biomass for mushroom culture. Other substrates also available in small quantities are sawdust and shavings of mixed woods that accumulate in small commercial sawmills. However, there is no forestry production in the province.

A relevant final consideration is that the introduction of mushroom cultivation in agricultural communities presents complexities associated not only to the availability of residual biomass, economic costs or logistics of its production, but also to cultural issues such as gastronomy and traditional practices of food production (Carrera et al., 2007). In this scenario it is essential to plan a strategy for the introduction of fungal culture in this region. Recently oyster mushroom production was incorporated working together with secondary schools and small cooperatives, using a mixed substrate formulated with jojoba litter and grape and olive pomace. In a society without a habitual consumption of edible mushrooms and with a negative general attitude towards fungi, it is essential to lead promotion activities with educational institutions, in order to ensure that mushroom production is welcomed into the community.

## CONCLUSIONS

In La Rioja province, an estimated mass of 75,000 tons of crop residues and agro-industrial by-products are generated annually. Our preliminary results of BE suggest the potential incorporation of this biomass into food production systems. In a context of poverty, poor availability of healthy food, and emerging environmental issues, the utilization

of this biomass as a substrate for mushroom cultivation is a feasible solution to be carried out in this region. Potentially in a single process, the generation of harmful gases can be reduced, nutraceutical food can be produced, and small farmers can incorporate a profitable activity.

## ACKNOWLEDGEMENTS

This work was supported by the grant PUE (Proyecto Unidad Ejecutora) No. 125, awarded by CONICET (National Scientific and Technical Research Council- Argentina).

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