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EFFICIENCY OF EXTRACTION OF BIOMOLECULES FROM PLANT SPECIES WITH ETHANOL AND ACETONE THROUGH ULTRASOUND ASSISTED EXTRACTION

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**Summary:** Plant species synthesize biomolecules that combine with each other and form complex mixtures that produce antimicrobial biological activity derived from their secondary metabolism (Rodríguez et al., 2012; and Adirano et al., 2018). The extraction efficiency of secondary metabolites depends to a large extent on the type of solvent and technique to be used (Hussam et al., 2013; Zulkafli et al., 2014; Setyaningsih et al., 2015). The objective of the article was to estimate the efficiency of extraction of secondary metabolisms of 10 plant species with ethanol and acetone by ultrasound-assisted extraction in Oaxaca, Mexico. The methodology used was adapted from Guerrero et al, (2007). Approximately 10 kilograms in fresh weight were collected from each species and placed in paper bags, disinfected and dehydrated for 30 days. They were ground in a Krups brand coffee grinder. Branson model 3800 ultrasonic equipment was used, distilled water was applied at the indicated level. Three Erlenmeyer flasks filled with 350 mL of solvent (acetone or ethanol depending on the extract) and 25 g of dehydrated and crushed plant material were placed, obtaining a concentration of 71.43 mg mL-1. This mixture was processed with ultrasonic waves for 3 hours. The extract was placed in a pear flask, for the separation of the solvent-extract in a Buchi model B-100 rotary evaporator, they were deposited in labeled Petri dishes, exposed in a Novatech brand extraction hood, model: CE- 120BE. Finally, the resulting extract was collected with a spatula, weighed, placed in amber flasks and stored in a refrigerator. The results obtained in this investigation show a difference between treatments due to the technique and solvents evaluated, the extraction efficiency varies according to the plant and the solvent used. Keywords: coffee, spices, plant extract.

# INTRODUCTION

It is clear that the industrial-conventional agricultural model and its questionable biotechnological derivations is exhausted and will not be able to provide answers to the current challenge. Given this circumstance, a process of conversion from conventional systems to diversified systems with ethnoecological rationality and transitional character is necessary (Nicholls and Altieri, 2012).

Plant extracts have become an ecologically sustainable and economically viable alternative in the control of pests and diseases (Rodríguez et al., 2012). This is due to the fact that various plant species synthesize, as part of their secondary metabolism, a large number of biomolecules that, combined, form complex mixtures that have demonstrated antimicrobial biological activity (Adirano et al., 2018; Rodríguez et al., 2012), carvacrol, eugenol, thymol, borneol and phytol are some of them (Garcia et al., 2010; Kalemba and Kunicka, 2003). However, the yield and efficiency of metabolite extraction is a question when trying to implement this technique in the field.

The efficiency and yield of extraction of secondary metabolites depends to a large extent on the type of solvent and technique to be used (Bruneton, 2001; Hussam et al., 2013; Luque-García and Luque de Castro, 2004; Setyaningsih et al., 2015; Zulkafli et al., 2014). The variation can also be determined by the amount of compounds or by the difference between them (Sefidkon et al., 2006).

Ultrasound-assisted extraction is, together with Soxhlet, the most accepted conventional leaching technique (Luque de Castro and García-Ayuso, 1998). Although they are two different techniques, their efficiency has been shown to be very similar (Dunnivant and Elzerman, 1988). However, the process is faster in ultrasound-assisted extraction (Luque de Castro and García-Ayuso, 1998). In many cases the ultrasound technique is advisable for thermolabile analytes that are altered when Soxhlet is used (Jenkins and Walsh, 1994).

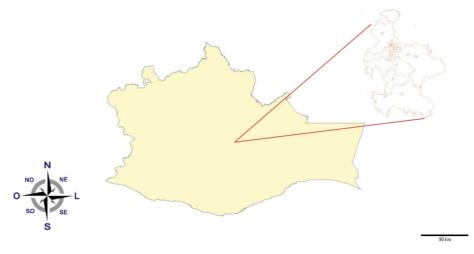
For this reason, the objective of this research is to estimate the efficiency of extraction of secondary metabolites of 10 plant species with ethanol and acetone by ultrasound-assisted extraction, in Oaxaca, Mexico. The work is presented in two sections. The first presents the materials and methods used during the development of the research, the second shows the results obtained accompanied by the discussion, the conclusions derived from the analysis of the results found and finally the cited literature are presented.

# MATERIALS AND METHODS

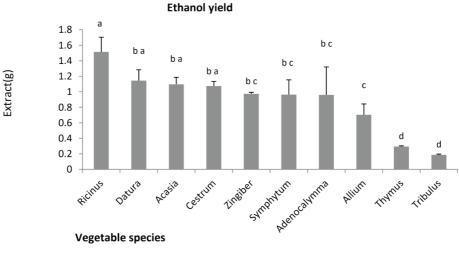
## **STUDY AREA**

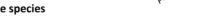
The collections were taken in the Valles Centrales region of Oaxaca (map 1). The experimental species were: garlic (Allium sativum L.), solder with solder (Anredera vesicaria Lam.), thyme (Thymus vulgaris L.), Toloache (Datura ferox L.), yellow caltrop (Tribulus terrestris L.), Garlic (Adenocalymma alliaceum Lam.), Ginger (Zingiber officinale Rosc.), Poton xihuite (Cestrum sp L.), Castor (Ricinus communis L.) and yellow aroma (Acacia farnesiana L.).

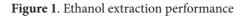
Extraction of secondary metabolites was performed, with acetone and ethanol, on 10 plant species: garlic (Allium sativum), solder with solder (Symphytum officinale), thyme (Thymus vulgaris), toloache (Datura ferox), yellow caltrop (Tribulus terrestrials), garlic vine (Adenocalymma alliaceum), ginger (Zingiber officinale), xihuite squid (Cestrum sp.), castor (Ricinus communis) and yellow aroma (Acacia farnesiana). The collection was made in various sites in the state of Oaxaca. The methodology used was from



Map 1. Central Valleys of Oaxaca, own elaboration on digital map version 6.3.0







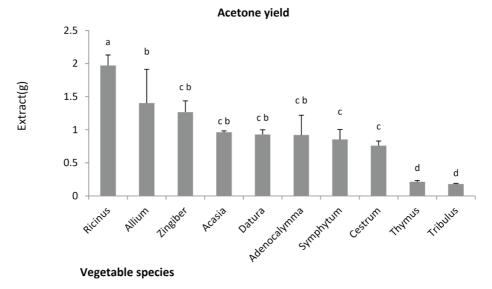


Figure 2. Acetone extraction yield

Guerrero et al, (2007) with modifications. Approximately 10 kilograms in fresh weight of each species were collected and placed in paper bags for transfer to the laboratory. They were washed with drinking water and later with distilled water, for dehydration they were spread on sheets of brown paper under shade for 30 days. They were ground in a Krups brand coffee grinder. Subsequently, Branson model 3800 ultrasonic equipment was used and distilled water was applied at the level marked by the equipment. Three Erlenmeyer flasks previously filled with 350 mL of solvent (acetone or ethanol depending on the extract) and 25 g of dehydrated and crushed plant material were placed, obtaining a concentration of 71.43 mg mL-1. This mixture was processed with ultrasonic waves for 3 hours. The extract was placed in a pear flask, for the separation of the solvent-extract in a Buchi model B-100 rotary evaporator. Once the process was completed, the extract obtained was placed in Petri dishes labeled with the symbology of the corresponding flask, for its total separation, it was exposed in a Novatech brand extraction hood, model: CE-120BE. Finally, the resulting extract was collected with a spatula, weighed, placed in amber flasks and stored in a refrigerator.

Approximately 10 kilograms in fresh weight were collected and placed in paper bags for transfer to the laboratory, dehydrated, and subsequently ground.

## **RESULTS AND DISCUSSION**

The data obtained from the extraction process of the different plant species were analyzed by means tests using the TUKEY method, in order to obtain the extraction efficiency and standard error (Figure 1 and 2), which were analyzed using the program SAS statistician. Statistical Analysis System.

Regarding the extraction efficiency, the best results with ethanol were expressed by

the species of Ricinus communis, Datura ferox and Acasia farnesiana, with 1.51g, 1.14g and 1.09g respectively, being Tribulus terrestrial with 0.18g the lowest result. In the case of extraction with acetone, the best plant species were: Ricinus communis, Allium sativum and Zingiber officinale, with results of 1.97g, 1.40g and 1.26g respectively, Tribulus terrestrial with 0.18g expressed the lowest result.

Species	CEOE	CEOA	EPE %	EPA %
Ricinus	$1.5133 \pm 0.099^{a}$	$1.9700 \pm 0.023^{a}$	6.05	7.88
Datura	$1.1433 \pm 0.070^{ab}$	$\begin{array}{c} 0.9300 \pm \\ 0.012^{\rm bc} \end{array}$	4.57	3.72
Acasia	$1.0967 \pm 0.044^{\rm bc}$	$\begin{array}{c} 0.9336 \pm \\ 0.003^{bc} \end{array}$	4.39	3.85
Cestrum	1.0733 ± 0.029 <sup>bc</sup>	$0.7600 \pm 0.014^{\circ}$	4.29	3.04
Zingiber	$\begin{array}{c} 0.9733 \pm \\ 0.012^{\rm bc} \end{array}$	1.2667 ± 0.028 <sup>bc</sup>	3.89	5.07
Symphytum	$\begin{array}{c} 0.9633 \pm \\ 0.095^{\rm bc} \end{array}$	$\begin{array}{c} 0.8533 \pm \\ 0.028^{c} \end{array}$	3.85	3.41
Adenocalymma	$\begin{array}{c} 0.9600 \pm \\ 0.181^{\rm bc} \end{array}$	$\begin{array}{c} 0.9200 \pm \\ 0.055^{\rm bc} \end{array}$	3.84	3.68
Allium	$\begin{array}{c} 0.7033 \pm \\ 0.072^{c} \end{array}$	${\begin{array}{c} 1.4033 \pm \\ 0.083^{ba} \end{array}}$	2.81	5.61
Thymus	$\begin{array}{c} 0.2933 \pm \\ 0.006^{d} \end{array}$	$\begin{array}{c} 0.2133 \pm \\ 0.005^{d} \end{array}$	1.17	0.85
Tribulus	$\begin{array}{c} 0.1867 \pm \\ 0.003^{d} \end{array}$	$\begin{array}{c} 0.1800 \pm \\ 0.002^{\rm d} \end{array}$	0.75	0.72

**Table 1.** Yield obtained from 25 g of dehydratedmatter and 350 mL of solvent.

Letters in different columns indicate statistically significant differences (Tukey 0.05), ¶transformed variable (SQRT) CEOE = amount of extract obtained with ethanol, CEOA = amount of extract obtained with acetone, EPE = percentage extraction efficiency, EPA = percentage extraction efficiency. The mean ± standard error is included.

# CONCLUSIONS

The results obtained show that with the technique and the solvents evaluated in this investigation there was a difference between treatments, the extraction efficiency varies according to the plant and the solvent used. The highest biomolecule extraction efficiency was recorded for *Ricinus communis, Datura* 

ferox and Acacia farnesiana with 6.5%, 4.5% and 4.3% respectively with ethanol and acetone, *Ricinus communis, Allium sativum y Zingiber officinale* were the most efficient with 7.9%, 5.6% and 5.0% respectively. The highest extract yield with ethanol was 1.51 g and with acetone 1.97 g per 25 g of dehydrated plant.

# REFERENCES

Adirano, A. L., J. Mejia, I. Ovando, V. Albores, M. Salvador. 2018. Effect of alcoholic extracts of garlic (*Allium sativum*) and clove (*Syzygium aromaticum*) on the development of *Mycosphaerella fijiensis* Morelet. Revista Mexicana de Fitopatología. 36(3): 379-393.

Bruneton, J. 2001. Farmacognosia. Fitoquimica Plantas Medicinales. 2da edición. Acribia, S. A. 1073 p.

Dunnivant, F. M. and A. W. Elzerman. 1988. Determination of polychlorinated biphenyls in sediments, using sonication extraction and capillary column gas chromatography-electron capture detection with internal standard calibration. J Assoc Off Anal Chem. 71(3):551-6.

Garcia, L. G., A. Martínez, J. Ortega, F. castro. 2010. Componentes químicos y su relación con las actividades biológicas de algunos extractos vegetales. Revista QuímicaViva. 2: 86 – 96.

Guerrero-Rodríguez, E., S. Solís-Gaona, F. D. Hernández-Castillo, A. Flores-Olivas, V. Sandoval-López. 2007. Actividad Biológica in vitro de Extractos de Flourensia cernua D.C. en Patógenos de Postcosecha: Alternaria alternata (Fr.:Fr.) Keissl., Colletotrichum gloeosporioides (Penz.) Penz. y Sacc. y Penicillium digitatum (Pers.:Fr.) Sacc. Revista Mexicana de FITOPATOLOGIA. 25: 48-53.

Jenkins, T. F., and M. E. Walsh. 1994. Instability of tetryl to Soxhlet extraction. Journal of Chromarogruphy. 178-184.

Hussam, A. Q., J. Cánovas, E. Barrajón, V. Micol, J. A. Cárcel, y J. V. García. 2013. Kinetic and compositional study of phenolic extraction from olive leaves (var. Serrana) by using power ultrasound. Innov. Food Sci. Emerg. 17: 120-129.

Kalemba, D. y A. Kunicka (2003). Antibacterial and antifungal properties of essential oils. Curr Med Chem 10 (10): 13-29.

Luque-García, J. L. and Luque de Castro, M. D. 2004. Ultrasound-assisted Soxhlet extraction: an expeditive approach for solid sample treatment Application to the extraction of total fat from oleaginous sedes. Journal of Chromatography. 237–242.

Nicholls, C. I. and Altieri, M. A. 2012. Modelos ecológicos y resilientes de producción agrícola para el siglo xxi. Agroecología 6: 28-37.

Rodríguez, P. A., M. Ramirez, S. Bautista, A. Cruz, D. Rivera. 2012. Actividad antifúngica de extractos de Acacia farnesiana sobre el crecimiento *in vitro* de *Fusarium oxysporum* f. sp. *Lycopersici*. Revista Científica UDO Agrícola 12 (1): 91-96. 2012

Sefidkon, F., K. Abbasi, and G. B. Khaniki. 2006. **Influence of drying and extraction methods on ngibre (Zingiber officinale)**. Revista Amazónica de Investigacion Alimentaria 1(1):38-42.

Setyaningsih, W., I. E. Saputro, M. Palma, and C. G. Barroso. 2015. **Optimization and validation of the microwave-assisted extraction of phenolic compounds from rice grains**. Food Chem. 169: 141-149.

Zulkafli, Z. D., H. Wang, F. Miyashita, N. Utsumi, and K. Tamura. 2014. Cosolvent-modified supercritical carbon dioxide extraction of phenolic compounds from bamboo leaves (*Sasa palmata*). J. Supercrit Fluid. 94: 123-129.