

Polyphenols of chuchuhuazo (*Maytenus macrocarpa* bark) as antioxidant and preservative in fresh sausage.


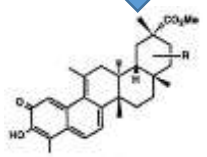
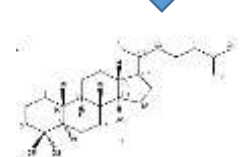

**Authors:** Cristian Correa<sup>1</sup>, Manuel Pérez Quintana<sup>2</sup>, Luis Bravo Sánchez<sup>2</sup>, Reinier Abreu<sup>2</sup>, Darwin Viáfara<sup>3</sup>.

<sup>1</sup>Graduate student in agro-industrial engineering. Amazon State University, Km. 2 ½, vía Puyo a Tena (Paso Lateral). Tel. 032-888-118 / 032-889-118. Postal Code: 160150. Puyo, Ecuador.

<sup>2</sup>Professors-Researchers. Amazon State University, Km. 2 ½, vía Puyo a Tena (Paso Lateral). Tel. 032-888-118 / 032-889-118. Postal Code: 160150. Puyo, Ecuador.

<sup>3</sup>Researching technician. Amazon State University, Km. 2 ½, vía Puyo a Tena (Paso Lateral). Tel. 032-888-118 / 032-889-118. Postal Code: 160150. Puyo, Ecuador.

Author for correspondence: mperez@uea.edu.ec

<p style="text-align: center;"><b>Graphical Abstract</b></p> <p style="text-align: center;">Chuchuhuazo (<i>Maytenus macrocarpa</i>)</p> <div style="text-align: center;">  </div> <div style="text-align: center; border: 1px solid black; padding: 5px; margin: 10px 0;"> <p>Total polyphenols and antioxidant compound from <i>Maytenus macrocarpa</i> are used in sausage as preservative and antioxidant agent.</p> </div> <div style="display: flex; justify-content: space-around; align-items: center;"> <div style="text-align: center;">  <p>Triterpenoid</p> </div> <div style="text-align: center;">  <p>Triterpenoid Dammarane</p> </div> </div> <div style="text-align: center; margin: 10px 0;">  <p><b>Sausage prepared</b></p> </div> <p style="text-align: center;"><b>ANALYSIS</b></p> <div style="display: flex; justify-content: space-around; margin-top: 20px;"> <div style="text-align: center;"> <p>Bromatological Properties</p> </div> <div style="text-align: center;"> <p>Antioxidant Activity</p> </div> </div>	<p><b>Abstract</b></p> <p>The aim was to make sausage with addition of chuchuhuazo polyphenols (<i>Maytenus macrocarpa</i>) as preservative and antioxidant agent. Taking into account the health of the consumer as a priority, as it is a very consumed food in the locality and due to the nutritional properties sausage was processed; polyphenols were added as a chemical preservatives and antioxidants substitute during the mixing of the meat with the seasonings that were used as flavoring agents. As for the bromatological properties, the results showed a protein content that agrees with what is established in the INEN 1338-2012 standard. Everything indicates that the food evidenced was functional and could be conserved and processed using only substances of natural origin. The antioxidant activity was favorable in terms of the results obtained by the analytical methods of determination of total polyphenols and antioxidant activity (FOLIN, FRAP, ABTS), which found that there was a decrease in the oxidative action within the sausage as a function of time.</p> <p><b>Keywords.</b> <i>Maytenus macrocarpa</i>, Polyphenols, antioxidants, preservatives.</p>
---	--

## **Introduction**

In the search for a healthy lifestyle, there are determining factors that lead to functional diets investigation, in such a way that they help in the prevention and cure of diseases. The food study acquires great importance, given the high chronic diseases incidence (Reyes and Atalah 2006). As part of the functional diet, natural foods are used and new methods arise in the processing and elaboration of innovative and functional products, through the use of raw materials from the Amazon region in Ecuador.

In view that meat plays an important role in the human being diet, due to the large amount of proteins, minerals and vitamins it contains, its demand is high and new meat processing methods are being developed, emphasizing in a chemical substances decrease used during the processes for conservation (Yausin & Llallico, 2007).

Taking into account, as a main point, the health of the consumer, they are intended to replace the chemical preservatives, used in the process of making sausages, by natural substances, because the consumer looks for products that contain less chemical additives and more natural products, that are innovative with affordable costs that contribute to the health care. Chuchuhuazo polyphenols (*Maytenus macrocarpa*) as alternative to improve the quality of the sausages, replacing the chemical preservatives with natural antioxidants without altering the physical-chemical, organoleptic and bromatological properties of the meat product (Quiñones, 2012).

Antioxidants have different action mechanisms, some prevent the formation of free radicals or reactive oxygen species (prevention system), others inhibit the action of free radicals and others favor the repair and reconstruction of damaged biological structures (system repair); the two antioxidant groups, as they are traditionally known, are the primary antioxidants and secondary antioxidants (Armenteros et al., 2012).

The present work analyze the effect of the addition of three polyphenols percentages of chuchuhuazo (*Maytenus macrocarpa*) in the bromatological and antioxidant characteristics of pork sausage.

## Materials and methods.

Location and duration of the experiment.

The present research was carried out in the Agroindustry, Bromatology, Chemistry and Biology Laboratories of the Amazon State University, belonging to the Department of Earth Sciences, located at km 21/2 via Tena, Canton Puyo, province of Pastaza with an altitude of 940 meters above sea level, with a latitude of 0 ° 59 '-1 "S and a length of 77 ° 49' 0" W. The investigation was carried out in a time of 65 days. The treatments consisted in the addition of polyphenols of chuchuhuazo (*Maytenus macrocarpa*) with a concentration of 0.2; 0.4 and 0.6% added to the meat base of sausage with polifenols of chuchuhuazo (*Maytenus Macrocarpa*).

Table 1 shows the pork sausage formulation without chemical additives, with the addition of chuchuhuazo polyphenols (*Maytenus macrocarpa*) in different percentages (0.2- 0.4-6.6%).

Table 1: Addition of chuchuhuazo polyphenols (*Maytenus macrocarpa*) to sausage.

Description	Quantity	Percentages
Pork Meat	0,81 kg	81 %
Pork loin Fat	0,19 kg	19 %
Ice	0,23 kg	11,5 %
Polifenles of <i>Maytenus M...</i>	-	0,2; 0,4 y 0,5 %
Common salt	32 g	1,6 %
Sugar	3,16 g	0,158 %
Condiments for sausage	30 g	1,5 %
Milk powder	2 g	0,1 %
Peppers	3 g	0,15 %
Black pepper	3,33 g	0,167
Liquid smoke	1 ml	
Red vegetable coloring	2 ml	

## Evaluation of the bromatological features.

The evaluation of the bromatological characteristics of the sausage were protein (Cruz, 2017), ash (AOAC, 1995), fat (Cobos et al., 2014) and humidity (Navarro, 2007) and they were carried out, in order to determine the nutritional value of the meat food.

## Extraction of polyphenols from the bark of chuchuhuazo (*Maytenus macrocarpa*).

The polyphenolic extracts were obtained by ultrasound extraction methods. 5 g of samples and 25 ml of absolute ethanol were placed in a 100 ml glass beaker. After sonication, the mixtures were filtered through the Whatman No. 4 paper under vacuum conditions and stored in an amber bottle (Abreu et al, 2017).

## Antioxidant activity spectrophotometric determination.

The polyphenol content determination was made according Folin-Ciocolteau test (Proestos and Varzakas, 2017). The total antioxidant activity was carried out by two methods: ABTS (2,2-azinobis (3-ethylbenzthiazolin)-6-sulfonic acid) according to Baqueiro-Peña and Guerrero-Beltrán (2017) and FRAP (Ferric ion reducing antioxidant Power) by Thaipong et al. (2006) technique.

## Results and Discussion.

### Bromatological analyzes.

The sausage nutrient contents are shown in table 2. An increase in protein content, total fat, sodium, dry mater and total carbohydrates was observed in the treatment with 0.6% polyphenolic extract. In the same way, the contents of total fat, sodium, dry matter and total carbohydrates in 0.4% of extract are higher.

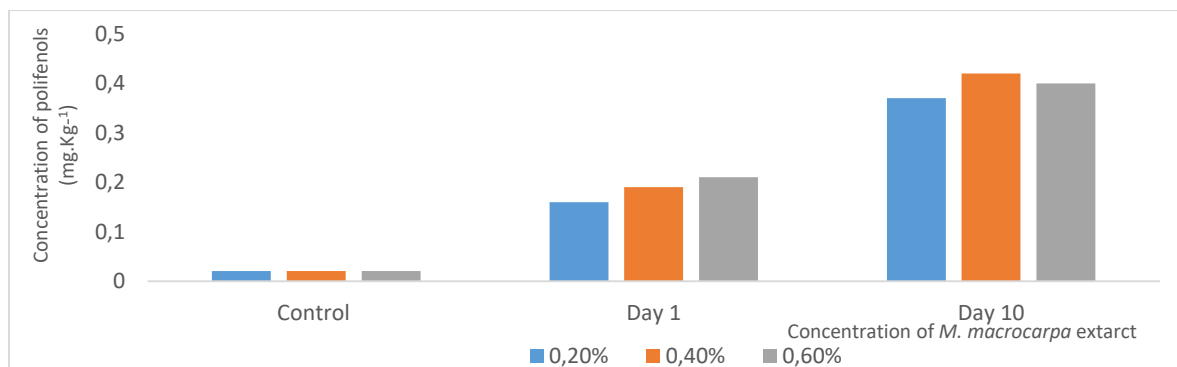
The values of the sausage bromatological analysis stored during 10 days were similarity that exists in the INEN 1338-2012 standard (Navarro, 2007).

**Table 2: Determination of the nutritional content in sausage after 10 days of storage.**

Analysis	control	0,2 %	0,4 %	0,6 %
Crude protein g/100g	21,72 <sup>b</sup>	22,23 <sup>b</sup>	21,57 <sup>b</sup>	23,94 <sup>a</sup>
Total fat g/100g	29,27 <sup>a</sup>	23,1 <sup>b</sup>	26,27 <sup>b</sup>	25,92 <sup>b</sup>
Water g/100g	39,67	38,72	33,82	33,27
Minerals g/100g	4,96	4,63	4,1	4,34
Salt (Na Cl) g/100g	3,52	3,36	3,35	3,18
Sodium mg/100g	1386,58 <sup>b</sup>	1323,4 <sup>b</sup>	1666,91 <sup>a</sup>	1252,09 <sup>b</sup>
Dry mater g/100g	60,33 <sup>b</sup>	61,28 <sup>b</sup>	66,18 <sup>a</sup>	66,73 <sup>a</sup>
Total carbohydrates g/100g	8,37 <sup>b</sup>	11,32 <sup>a</sup>	14,24 <sup>a</sup>	12,53 <sup>a</sup>

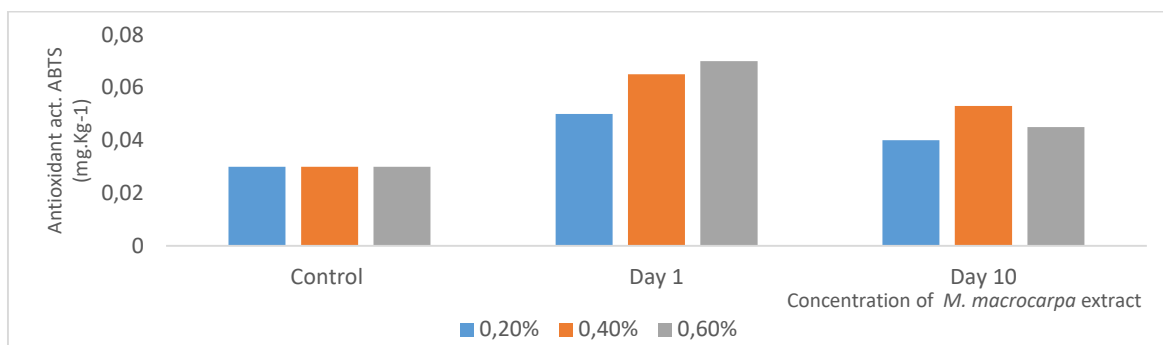
### Polyphenols activity in sausage in relation to the time.

Figure 1 shows the results in polyphenols activity in sausage and the time during storage. It is observed that both time, concentration and their interaction have a significant effect. The independent variable of greatest influence was time, followed by interaction and finally concentration.

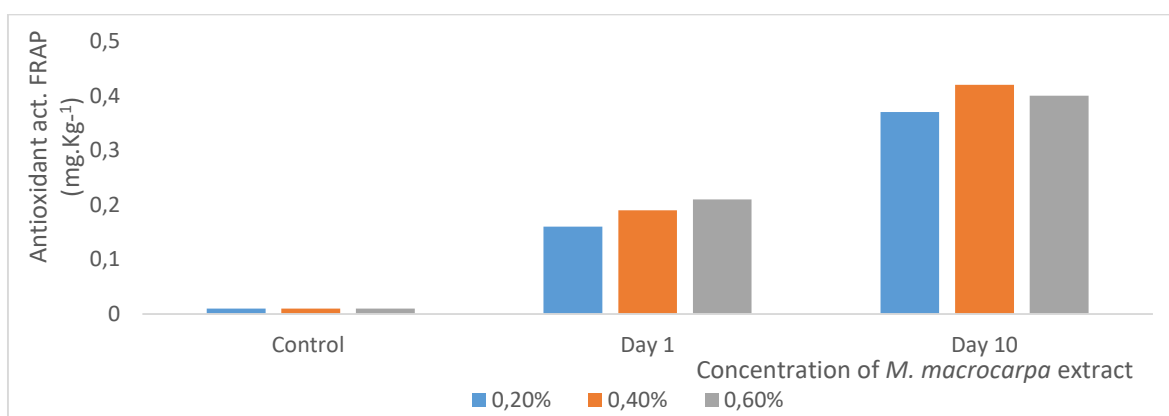


**Figure 1.** Polyphenol activity by folin in sausage and during storage.

In the figures 2 and 3 the results of the antioxidant activity are presented with the data obtained from the means, result of analysis with the both methods: FRAP-ABTS. It is demonstrated that the factors of study (time-concentration) and their interaction during sausage storage are also significant in relation to the concentration of polyphenols into sausage, however, the variable time is the highest incidence, followed by interaction and concentration.



**Figure 2:** Antioxidants activity in chorizo by the ABTS method.



**Figure 3.** Antioxidant activity analysis in sausage determined by the FRAP method.

Several studies have shown a high relationship between these compounds and the antioxidant potential when using different antioxidant analysis techniques (Kuskoski et al., 2005, Muhtadi et al., 2015).

During the experiment with the application of chuchuhuazo (*Maytenus macrocarpa*) polyphenols it was demonstrated that the substance used is favorable for the sausage, decreasing the oxidative activity of the lipids and fats contained in the product, the shelf life of the food was completed 10 days of conservation and subject to temperatures not higher than 18 °C, without showing changes in color, taste, smell, texture and nutritional value which were demonstrated with the application of polyphenol and antioxidant analysis methodologies such as FOLIN, FRAP, ABTS; adding to them the bromatological analyzes

that demonstrated the feasibility of the food as a functional, nutritious and alternative for the human being.

The use of these polyphenols, as antioxidants in meat products, has shown a considerable increase in the preservation and nutritional value in the meat sausage. This makes it a relevant, effective alternative for the processing of healthy, nutritious foods with positive organoleptic characteristics before the human sight and taste of the consumer.

## Conclusions

The pork sausage with the addition of *Maytenus macrocarpa* polyphenols showed antioxidant activity and favorable conservation characteristics for quality according to the analysis of antioxidant activity according to Folin, Frap and Abts. After 10 days of storage showed nutritional values and a favorable antioxidant activity and protein content in a 22.23 g/100 g during that period of time.

## Bibliography

- AOAC, 1995. En: Official method of analysis of A.O.A.C. 10 th Edic. Assoc. Of Official Agricultural Chemists. Washington DC.
- Armenteros, M., Ventanas, S., Morcuende, D., Estevez, M., & Ventanas, J. (2012). *Empleo de antioxidantes naturales en productos carnicos*. eurocarne, N° 207. Junio 2012. [http://www.eurocarne.com/daal/a1/boletin\\_imagenes/a2/20705.pdf](http://www.eurocarne.com/daal/a1/boletin_imagenes/a2/20705.pdf)
- Baqueiro-Peña I, Guerrero-Beltrán JÁ (2017) Physicochemical and antioxidant characterization of Justicia spicigera. Food Chem 218:305–312. <https://doi.org/10.1016/j.foodchem.2016.09.078>
- Cobos Velasco, J. E., Soto Simental, S., & Alfaro Rodriguez, R. H. (2014). Evaluación de parámetros de calidad de chorizos elaborados con carne de conejo, cordero y cerdo, adicionados con fibra de trigo. Evaluation of quality parameters of sausages made with rabbit meat, lamb and pork, added with wheat fiber. 8, 15.
- Cruz Lugo, A. L. (2017). *Correlacion del método kjeldahl con el método dumas automatizado para determinación de proteína en alimento*. Universidad Autonoma del Estado de Hidalgo. <http://dgsa.uaeh.edu.mx:8080/bibliotecadigital/bitstream/handle/231104/555/Correlacion%20del%20metodo%20KJELDAHL.pdf;jsessionid=EFCD9DAAC920F454A729A734F5986E1F?sequence=1>.
- Kuskoski Marta, Asuero Agustín, Troncoso Ana, Mancini-Filho Jorge, Fett Roseane. 2005. Aplicación de diversos métodos químicos para determinar actividad antioxidante en pulpa de frutos. Cienc. Tecnol. Aliment., Campinas, 25(4): 726-732.
- Muhtadi, Haryoto, Tanti Azizah, Peni Indrayudha, Andi Suhendia, Khong Heng Yenb. 2014. International Journal of Indigenous Medicinal Plants, ISSN:2051-4263, Vol.47, Issue 1.
- Navarro Marquez, M. A. (2007). *Analisis de alimento*. colegio de bachilleres del estado de sonora. México. <http://www.etpcba.com.ar/DocumentosDconsulta/ALIMENTOS-PROCESOS%20Y%20QU%20C3%20DMICA/Manual%20de%20pr%20C3%A1cticos%20en%20alimentos.pdf>.
- Proestos and Varzakas. 2017. Aromatic Plants: Antioxidant Capacity and Polyphenol Characterisation. Foods, 6, 28; doi:10.3390/foods6040028
- Quiñones, M. (2012). Los polifenoles, compuestos de origen natural con efectos saludables sobre el sistema cardiovascular. 76-89. doi:10.3305/nh.2012.27.1.5418.

- Reinier Abreu-Naranjo, Yasiel Arteaga-Crespo, Luis Ramón Bravo-Sánchez, Manuel Lázaro Pérez-Quintana, Yudel García-Quintana. 1917. Response surface methodology for optimization of total polyphenol content and antioxidant activity of extracts from *Maytenus macrocarpa* bark by means of ultrasound-assisted extraction. *Wood Science and Technology* <https://doi.org/10.1007/s00226-018-1034-x>.
- Reyes J, Marcela y Atalah S, Eduardo. Intervención nutricional en prevención de enfermedades cardiovasculares: el caso de Noruega. *Rev. chil. nutr.* [online]. 2006, vol.33, n.3 [citado 2018-09-21], pp.464-472. Disponible en: <[https://scielo.conicyt.cl/scielo.php?script=sci\\_arttext&pid=S0717-75182006000500002&lng=es&nrm=iso](https://scielo.conicyt.cl/scielo.php?script=sci_arttext&pid=S0717-75182006000500002&lng=es&nrm=iso)>. ISSN 0717-7518. <http://dx.doi.org/10.4067/S0717-75182006000500002>.
- Thaipong K, Boonprakob U, Crosby K, Cisneros-Zevallos L, Hawkins Byrne D (2006) Comparison of ABTS, DPPH, FRAP, and ORAC assays for estimating antioxidant activity from guava fruit extracts. *J Food Compos Anal* 19:669–675. <https://doi.org/10.1016/j.jfca.2006.01.003>.
- Yausin, & LLallico, C. (2007). *Evaluación de tres tipos de antioxidantes naturales*. Escuela Superior Politecnica de Chimborazo. Repositorio Institucional de la Escuela Superior Politécnica de Chimborazo. <http://dspace.esPOCH.edu.ec/handle/123456789/46/browse?type=author&order=ASC&rpp=20&value=Yausin+Llallico%2C+Carlos>