Journal of Soil Science and Plant Nutrition, 2013, 13(1), 67-78

Influence of genotype and harvest year on polyphenol content and antioxidant activity in murtilla (*Ugni molinae* Turcz) fruit

S. Alfaro[1], A. Mutis[2], R. Palma[2], A. Quiroz[2], I. Seguel[3] and E. Scheuermann[4,5*]

*[1]Programa de Doctorado en Ciencias de Recursos Naturales, Universidad de La Frontera, Casilla 54-D, Temuco, Chile. [2]Laboratorio de Ecología Química, Departamento de Ciencias Químicas y Recursos Naturales, Universidad de La Frontera, Casilla 54-D, Temuco, Chile. [3]Instituto de Investigaciones Agropecuarias, INIA-Carillanca, Casilla 58-D, Temuco, Chile. [4]Chemical Engineering Department, Universidad de La Frontera, Casilla 54-D, Temuco, Chile. [5]Center of Food Biotechnology and Bioseparations, BIOREN-UFRO, Universidad de La Frontera, Casilla 54-D, Temuco, Chile. *Corresponding author: ericks@ufro.cl*

Abstract

Polyphenol content and antioxidant activity in murtilla (*Ugni molinae* Turcz) fruit from three genotypes (the 14-4 genotype and the Red Pearl-INIA and South Pearl-INIA varieties) were studied over five growing seasons. Our results showed significant differences in total polyphenol content among yearly harvests. The lowest value (283 ± 72 mg GAE/100 g dw) was obtained for the 14-4 genotype in the 2008 harvest, and the highest value (2,152 ± 290 mg GAE/100 g dw) was observed for the variety South Pearl-INIA in 2007. The lowest value for antioxidant activity (2,234 ± 337 µmol TE/100 g dw) was obtained for the Red Pearl-INIA variety in 2008, and the highest value (4,073 ± 76 µmol TE/100 g dw) was observed for the 14-4 genotype in 2007. There was a significant effect of genotype and growing season on polyphenol content, antioxidant activity and dry matter content for the murtilla fruits evaluated in this study, but additional studies examining other abiotic and biotic factors are required to fully explain causality.

Keywords: DPPH, Trolox, rainfall, frost, climate conditions, native berry.

1. Introduction

The native Chilean species *Ugni molinae* Turcz, commonly called "murtilla," "mutilla" or "murta," is a wild perennial shrub that grows in southern Chile. This plant produces a small globoid berry with an equatorial diameter of 0.71-1.31 cm (Seguel *et al*., 2000). Infusions of murtilla leaves are highly valued in Chilean indigenous mapuche folk medicine for the treatment of conditions such as diarrhea, dysentery, and urinary tract pain (Montenegro, 2002). Murtilla is most often consumed as a fresh fruit because of its organoleptic characteristics, but the fruit is also processed commercially to be sold canned or as jam, juice or liquor (Scheuermann *et al*., 2008).

The murtilla fruit is known for its typical and surrounding aroma, which is produced by 24 volatile compounds that have been identified in four ecotypes and range in concentration from 1.2 to 250.5 μg kg^{-1} fresh weight (Scheuermann *et al*., 2008). Additionally, murtilla fruit is considered a valuable source of high quality pectin that has a chemical composition similar to that of commercial citrus pectin (Taboada *et al.*, 2010). Several reports show an association between the antioxidant activity of murtilla fruits and leaves and the levels of polyphenols (Avello and Pastene, 2005; Rubilar *et al.*, 2006; Shene *et al*., 2009; Ruiz *et al.*, 2010; Rubilar *et al.*, 2011).

Antioxidant activity studies using different ecotypes of murtilla have shown that the levels of antioxidant activity in the fruit are comparable to those of blueberry and lower than those of maqui and calafate fruits (Ruiz *et al*., 2010; Arancibia-Avila *et al*., 2011). These results suggest that high levels of flavan-3-ols such as catechin and epicatechin, in addition to the anthocyanins cyanidin-3-glucoside and peonidin-3-glucoside, could be the responsible for the high antioxidant activity observed in murtilla fruits. The concentration of flavonols observed in murtilla fruit was two times higher than the concentrations observed in maqui and calafate fruits, with quercetin derivatives being the most abundant flavonol (Ruiz *et al.*, 2010).

Polyphenols and many other natural antioxidants (vitamins, carotenoids and other endogenous constituents) may promote better health. These antioxidants are capable of fulfilling a number of functional roles, acting as free radical scavengers, peroxide decomposers, singlet and triplet oxygen quenchers, enzyme inhibitors and synergists (Mandal *et al*., 2009). A close relationship has been shown between dietary intake of various antioxidant components and potential health benefits. However, several studies have found that these compounds may be affected by unaccounted factors, such as variety, growing environment, growing season, climate, temperature, light, soil type and other conditions (processing, post-harvest storage), which could affect both antioxidant content and antioxidant activity in the fruit (van der Sluis *et al*., 2001; Bolling *et al*., 2010).

The objective of this study was to determine potential seasonal differences in polyphenol content and antioxidant activity in murtilla fruits from three genotypes (the 14-4 genotype and the Red Pearl-INIA and South Pearl-INIA varieties) harvested in five different years.

2. Materials and methods

2.1. Sampling and experimental design

Fruits were obtained from three murtilla genotypes (the 14-4 genotype and the Red Pearl-INIA and South Pearl-INIA varieties) belonging to a germplasm collection developed by INIA-Chile through its murtilla research project. The plants were grown in an experimental field near Puerto Saavedra (38°45' S in latitude, 73°21' W in longitude). The genotypes used in this research were selected for their agronomical and organoleptic characteristics (Seguel *et al*., 2000; Scheuermann *et al.*, 2008). The fruits were harvested in the same state and at the same stage in the plants' life cycle in April of 2006, 2007, 2008, 2009 and 2011 and were transported immediately after harvest to the Food Science Laboratory at the Universidad de La Frontera.

Total polyphenol content, antioxidant activity and dry matter of the murtilla fruit were determined for the three genotypes and the five harvest years (2006, 2007, 2008, 2009 and 2011). However, lack of financial support made it impossible to adequately manage the crops. Thus, the data from the fruit harvested in 2010 were excluded from the study due to low reliability.

2.2. Extraction

A solvent extraction of the murtilla fruit was used to measure polyphenol levels and antioxidant activity according to the methodology described by Scheuermann

(2009). Approximately 6 g of fresh fruit was weighed with a semi-analytical balance (+/- 0.001 g) and ground with a mortar. The crushed fruit was subsequently transferred to a 100 mL bottle and 20 mL of methanol (99.9%) that had been previously conditioned to a temperature e of 30 °C was added. The solvent fruit extraction was performed in an oven (GFL-3032, Germany) under agitation (170 rpm) for 20 min at 30 °C. After the extraction step, the fruit extract was separated from solid matter by vacuum filtration using Advantec 232 filter paper (Toyo, Japan). The extracts were stored in 100mL flasks protected with aluminum foil and were analyzed immediately.

2.3. Total polyphenol content

Total polyphenol content was determined according to the method described by Wong *et al*. (2006). Using this method, 3.16 mL of distilled water were added to 40 µL of the sample and mixed with 200 µL of Folin-Ciocalteau reagent. After 5 minutes, 600 µL of 20% Na_2CO_3 were added to the reaction mixture and the mixture was allowed to stand for 120 min. The absorbance of each sample was measured at 765 nm and expressed in mg of gallic acid equivalents (GAE) per 100 g of dry weight (dw) by calibrating the optical density of each sample to a standard curve that was previously established using varying concentrations of gallic acid.

2.4. DPPH antioxidant activity

The antioxidant capacity of the murtilla fruit extracts was determined by measuring the radical 2,2-diphenyl-1-picrylhydrazy (DPPH) scavenging of the antioxidant compounds present in the extracts. The degree of discoloration of the solution indicated the scavenging efficiency of the added substance. In its radical form, DPPH has an absorption band of 515 nm, which disappears following reduction by an antiradical compound (von Gadow *et al.*, 1997; Atoui *et al*., 2005). The initial absorbance of the DPPH in methanol was measured at 515 nm and had a range of 0.630 to 0.640. An aliquot (50 µL) of the murtilla fruit extract was added to 1,950 µL of the metanolic DPPH solution. The change in absorbance at 515 nm was measured at 30 min. The antioxidant capacity, based on the DPPH free radical scavenging ability of the extract, was expressed in µmol Trolox equivalents (TE) per 100 g of dry weight (dw).

2.5. Dry matter

Dry matter was determined using fruit moisture content according to the Instituto de Salud Pública de Chile (1998). Five grams of halved murtilla fruits were oven-dried at 105 ºC for five hours and then weighed. This process was repeated until the difference between two successive weighings was less than 5 mg.

2.6. Climate condition data

Climate records for rainfall and number of frosts were obtained from the closest meteorological station, located in Tranapuente (38°41' S, 73°21' W). The first growing season (2006) ran from May 2005 to April 2006 when the fruit was harvested. The growing season was the same during subsequent years, up through the final harvest in April 2011.

2.7. Data analysis

For all experiments, three to five replicates were analyzed per treatment. The data were analyzed using a mixed-model ANOVA with genotype and season, followed by Tukey's Honestly Significant Difference tests as pos-hoc tests (Sokal and Rohlf, 1995). Alternatively, Student's t-tests were used to analyze pairs of independent samples (Zar, 1999). Prior to performing the statistical tests, the data for each variable were checked for normality and homogeneity of variance. A canonical discriminant analysis was performed to evaluate murtilla genotype performance using 95% confidence ellipses. Total polyphenol content, antioxidant activity and dry matter were correlated with rainfall and number of frosts (measured in the field) using the nonparametric

Spearman's rank method. JMP v.8 software (SAS Institute Inc., Cary, NC) was used for all statistical analyses and differences were considered significant at $p \leq 0.05$.

3. Results and Discussion

3.1. Total polyphenol content, DPPH antioxidant activity and dry matter of murtilla fruit from different growing seasons

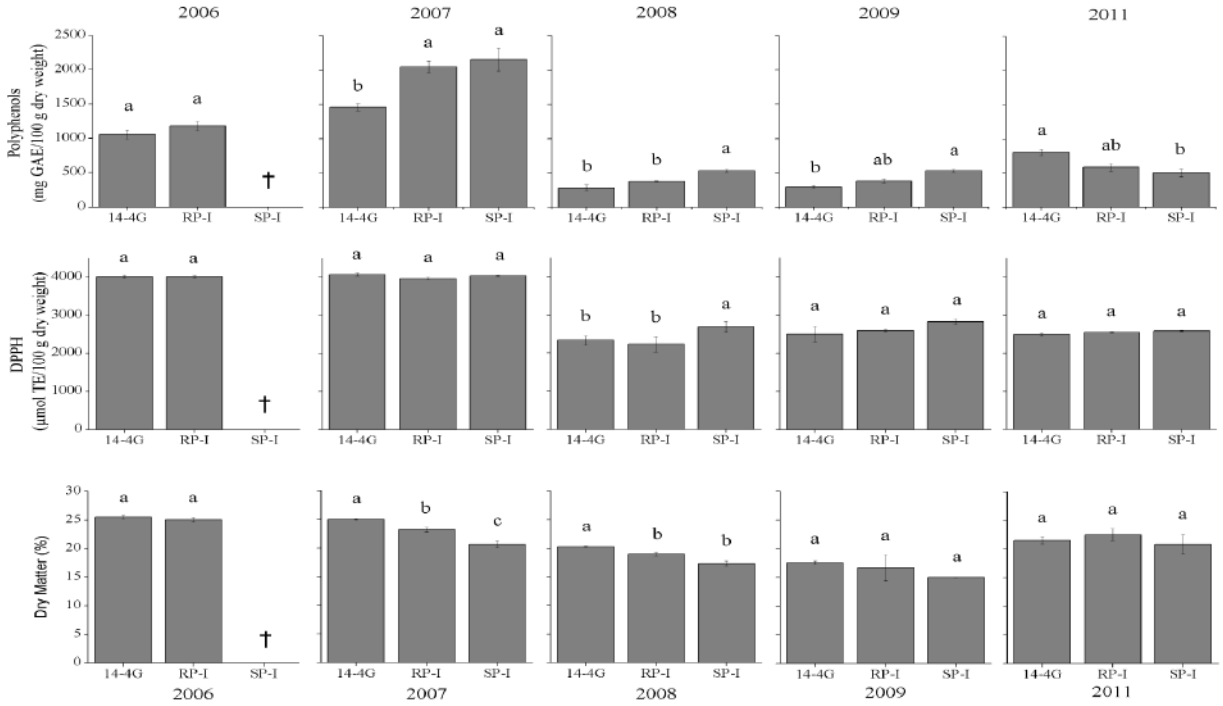
The effect of harvest year on total polyphenol content, DPPH antioxidant activity and dry matter for the three genotypes studied was significant (Tukey's test, $p<0.05$) (Figure 1). Large differences in total polyphenol content were observed among harvest years, with polyphenol content ranging from 283 ± 72 mg GAE/100 g dw for the 14-4 genotype in 2008 to 2,152 ± 290 mg GAE/100 g dw for the South Pearl-INIA variety in 2007. DPPH antioxidant activity ranged from 2,234 ± 337 µmol TE/100 g dw in the Red Pearl-INIA variety in 2008 to 4,073 ± 76 µmol TE/100 g dw for the 14-4 genotype in 2007.

These differences could be explained by variation in weather conditions among the growing seasons, such as differences in rainfall and the number of frosts. The highest correlations were between total polyphenol content and rainfall, dry matter and rainfall, dry matter and number of frosts and DPPH antioxidant activity and rainfall (Table 1). These abiotic factors could also be responsible for the changes in dry matter. For instance, notable changes in fruit dry matter between the 2006 and 2009 harvest years were observed for the 14-4 genotype (30.9% decrease) and the Red Pearl-INIA variety (33.2% decrease) (Figure 1). This result is corroborated by the significant correlations between dry matter and rainfall (0.82) and dry matter and number of frosts (0.75).

Therefore, our results showed a trend in increasing total polyphenol content, DPPH antioxidant activity and dry matter with increasing rainfall and number of frosts. Several researchers attribute differences in the level of phenolic compounds and antioxidant activity in fruits and vegetables of the same genotype grown in different locations or different years to changes in environmental conditions, such temperature, water availability (drought or precipitation), light intensity, salinity and pollination (van der Sluis *et al*., 2001; Connor *et al*., 2002; Howard *et al*., 2003; Temime *et al*., 2006; Ksouri *et al.*, 2008; Bolling *et al*., 2010; Kevers *et al*., 2011; Sun *et al.*, 2011). However, the mechanism behind the effects of low temperature (number of frosts) and water availability (rainfall) on phenolic content and other secondary metabolites in plants is not clearly understood. Normally, the mechanism considered the biotic or abiotic stresses that result in the formation of reactive oxygen species (ROS) followed by activation of the enzyme and hormone systems of the plant, producing antioxidants that include enzymatic and non-enzymatic components (Xin and Browse, 2000; Janda *et al*., 2003; Ksouri *et al.*, 2008). The effects of water stress (e.g., waterlogging or drought) and temperature (low v. high, constant v. fluctuating) on the medicinal herb *Hypericum brasiliense* included an increase in the levels of some phenolic compounds (quercetin, rutin, 1,5-dihydroxyxanthone and isouliginosin B). However, changes in phenolic content depended on the type of stress and the compound analyzed and were likely a response to the generation of ROS (de Abreu and Mazzafera, 2005).

Figure 2 shows the values for monthly total rainfall and minimum accumulated temperature used to correlate each growing season (Table 1) with total polyphenol content, DPPH antioxidant activity and dry matter for the three murtilla fruit genotypes. As shown in Figure 2A, the accumulated rainfall values for the month of August in the 2005-2006 and 2010-2011 growing seasons were similar to the total rainfall in the 2007-2008 and 2008-2009 growing seasons.

Figure 1. Total polyphenol content (mg GAE/100 g dry weight), DPPH antioxidant capacity (µmolTE/100 g dry weight) and dry matter (%) in murtilla fruit for the genotype and the two varieties over the five study years.



14-4 G: Genotype 14-4; RP-I: Red Pearl-INIA variety; SP-I: South Pearl-INIA variety. † Missed treatment. Letters for each combination of parameter/year indicate significant differences according to the HSD-Tukey test (Student's t-test was used to compare treatments within the 2006 harvest year) (p<0.05).

Table 1. Precipitation and number of frosts by growing season and their degree of correlation with the parameters measured in murtilla fruit.

	Growing Seasons							
	2005-2006	**2006-2007[a]**	**2007-2008**	**2008-2009**	**2010-2011**	**Total polyphenols [b]**	**DPPH AA [b]**	**Dry matter [b]**
Rainfall (mm)	1713	---	935	1014	1646	0.83 (39)	0.75 (36)	0.82 (36)
Number of frosts	9	---	8	2	7	0.62 (39)	0.56 (36)	0.75 (36)

a Missing data. **b.** All correlation coefficients (Spearman's rho) are significant at the p<0.001 level. Numbers in parentheses indicate sample size. DPPH AA: DPPH antioxidant activity.

There were differences in rainfall distribution and quantitative rainfall between April 2005 and April 2011, confirming that there was variance in this agronomic parameter over the course of the study period at the location of the experimental field is located (38°45' S latitude; 73°21'W longitude). A similar pattern occurred for minimum accumulated temperature (Figure 2B), with the 2008-2009 growing seasons showing the highest temperature (44 °C) and the lowest the number of frosts (Table 1) for the study period.

Year-to-year variations in certain phenolic compounds that are important sources of secondary plant metabolites have been observed in other types of fruit (van der Sluis *et al*., 2001; Kevers *et al*., 2011). Connor *et al*. (2002) reported that the phenolic content of several highbush and interspecific hybrid blueberry cultivars grown at three locations varied considerably over two growing seasons. Howard *et al*. (2003) suggested that this effect was not surprising because abiotic and biotic factors such as temperature, irradiation, herbivory and pathogenic infection are known to induce the protective antioxidant mechanisms of plants. The correlation coefficients determined by Sun *et al*. (2011) showed that altitude and annual precipitation had a significant effect on sour jujube fruit (*Ziziphus jujuba* Mill.). Fruits grown in harsh, arid and high-altitude areas can produce a larger amount of natural antioxidants and therefore may exhibit higher antioxidant activities than fruits grown in other areas.

3.2. Genotypic and seasonal effects on total polyphenol content, DPPH antioxidant activity and dry matter of murtilla fruit

The analysis of variance showed that the effects of genotype, season and genotype x season were significant for all variables, with the exception of genotype and genotype x season for DPPH antioxidant activity (Table 2).

Howard *et al*. (2003) reported similar results in blueberries and attributed these results to genetic and environmental factors. The significant main effects of growing season, genotype and genotype x growing season on antioxidant activity measure by the Oxygen Radical Absorbance Capacity (ORAC) test, phenolic content and fruit weight demonstrated that environmental growing conditions can impact phenolic and ORAC levels in blueberry and that genotypes vary in their capacity to synthesize phenolic compounds under different growing conditions. In contrast, Connor *et al.* (2002) found that the antioxidant activity and phenolic content of blueberry cultivars were more strongly affected by genotype than by harvest year.

Correlation does not imply causation, but a correlation does indicate that changes in one variable are related to changes in another variable. New studies that include other abiotic and biotic factors are necessary to determinate all of the causes that could fully explain year-to-year changes in total polyphenol content, DPPH antioxidant activity and dry matter in the murtilla fruit varieties evaluated in this study. It is the first whose purpose is to relate some environmental and chemical variables for this fruit.

3.3. Importance of genotype

The multivariate discriminant analysis of murtilla genotypes resulted in two canonical variables accounting for 94.2% and 5.7% of the variation between genotypes. Reduced space plots of the two canonical variables distinguished among the genotypes based on 95% confidence ellipses (Figure 3). The analysis showed that the responses of the Red Pearl-INIA variety (RP-I) and the 14-4 genotype (14-4G) were indistinguishable from each other under the conditions of this study, while the South Pearl-INIA (SP-I) variety appeared to have a significantly different effect.

In this study we demonstrated the effect of genotype and growing season on total polyphenol content, DPPH antioxidant activity and dry matter in murtilla fruit. There are reports showing

that antioxidant activity in the fruits and leaves of blackberry, raspberry, and strawberry plants varies with cultivar and developmental stage (Wang and Lin, 2000). In one study, apple polyphenol content varied by at least 5.2-fold among 67 cultivars (Wojdylo *et al*., 2008). The effect of genotype on antioxidant activity has been shown in grapes (Lee and Talcott, 2004), plums (Gil *et al.*, 2002), apples (Wolfe *et al*., 2003), citrus fruits (Bocco *et al*., 1998), guavas (Jiménez-Escrig *et al*., 2001), nectarines and peaches (Gil *et al.*, 2002). Bolling *et al.* (2010) found seasonal differences in total phenol and antioxidant constituents in almonds, reporting a 13% greater polyphenol content between different cultivars in 2005 compared to 2007. Because the qualitative and quantitative composition of phenolic compounds in fruits is unique to individual species and genotypes, it is possible to determine similarities and differences among standardized and confirmed genotypes and predict the response of new varieties to specific production and cultivation conditions (Sochor *et al*., 2010).

In this study, high correlations were found between climatic factors (rainfall and number of frosts) and the fruit characteristics evaluated (Table 1). Similar results were observed by Temime *et al*. (2006), who reported a positive linear relationship between precipitation and phenol content in Chétoui variety virgin olive oils. Moreover, the results of Janda *et al.* (2003) suggest that plants grown at low temperatures have higher levels of both antioxidants and antioxidant enzymes, and Xin and Browse (2000) suggest that low temperatures induce changes in plants, such as increased levels of antioxidants and reduced water content.

Table 2. F-values and significance of the main effects and interaction effect of the variables analyzed by multifactorial ANOVA.

	Genotype	**Season**	**Genotype x Season**
Total polyphenols content	11.17**	186.46**	7.83**
DPPH antioxidant activity	3.25ns	39.83**	0.67ns
Dry matter	10.77**	54.00**	2.31*

* $p<0.05$; ** P<0.01

ns: non-significant difference.

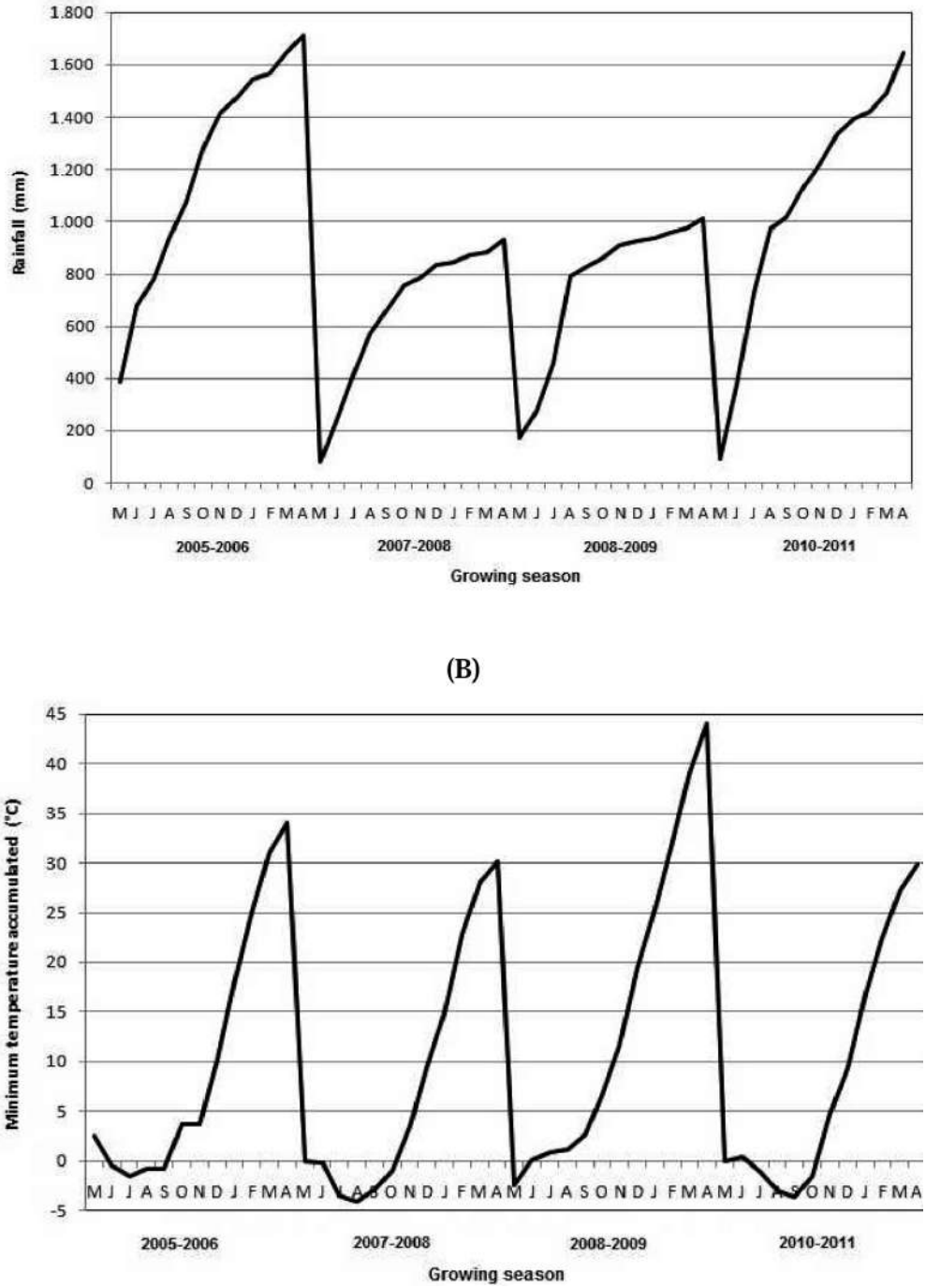


Figure 2. Changes in monthly total rainfall (A) and minimum accumulated temperature (B) for each growing season. Correlations with total polyphenol content, DPPH antioxidant activity and dry matter of the three murtilla fruit genotypes (the 14-4 genotype and the Red Pearl-INIA and South Pearl-INIA varieties) are shown in Table 1.

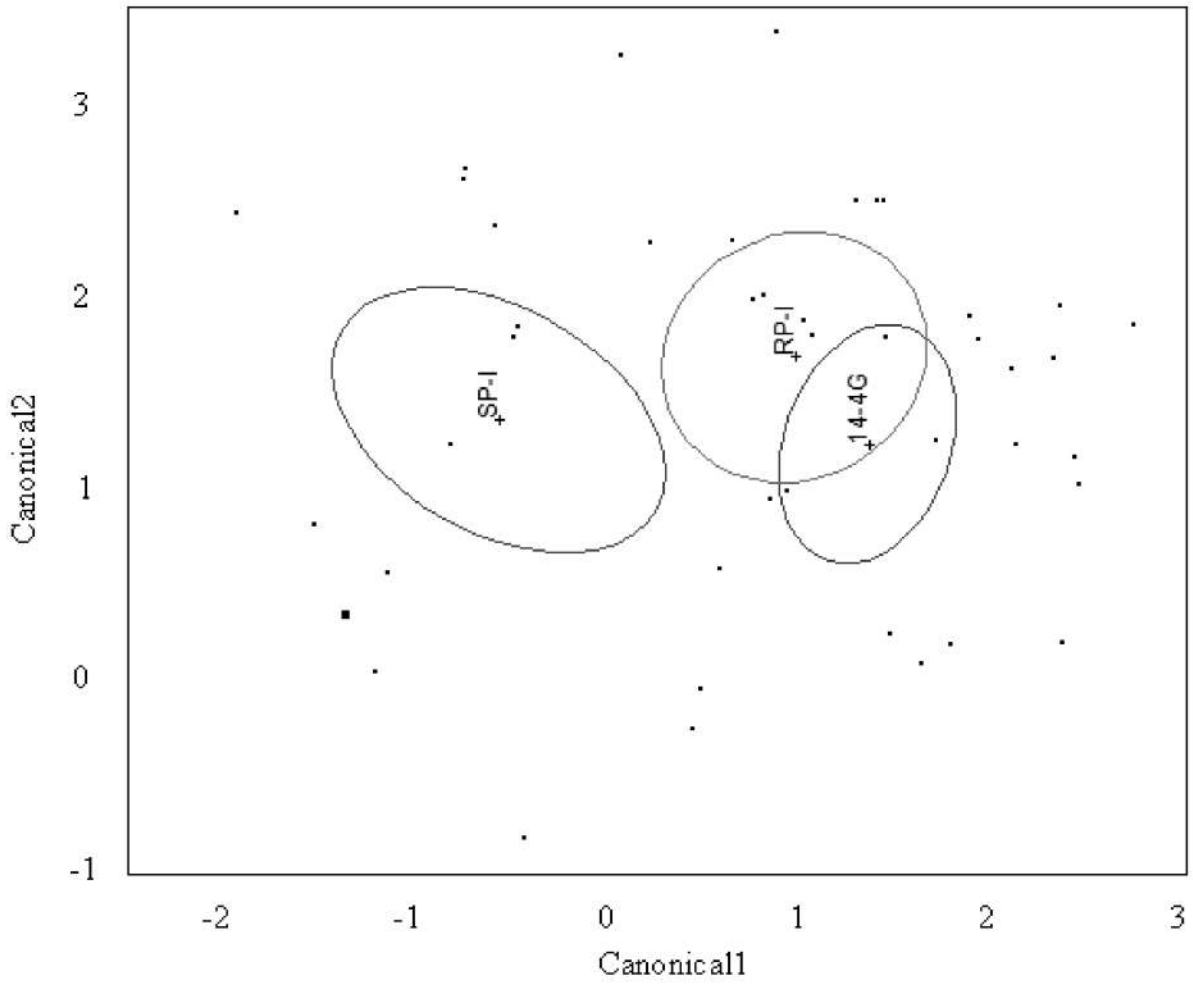


Figure 3. Canonical discriminant analysis of murtilla varieties based on polyphenol content, DPPH antioxidant activity and dry matter. Labels: SP-I: South Pearl-INIA variety, RP-I: Red Pearl-INIA variety, 14-4G: 14-4 genotype. Data represent the first and second canonical variables (Canonical 1 and Canonical 2) for the murtilla sample by variety (95% confidence ellipses).

4. Conclusions

In summary, in this study, there was a statistically significant effect of growing season on polyphenol content, antioxidant activity and dry matter of murtilla fruits from three genotypes (the 14-4 genotype and the Red Pearl-INIA and South Pearl-INIA varieties). These characteristics trend to increase with higher rainfall and number of frosts. In addition, polyphenol content and dry matter were affected by genotype. A canonical discriminant analysis of seasonal differences in the Red Pearl-INIA, South Pearl-INIA and 14-4 genotypes showed that the South Pearl-INIA variety varied the most from the other genotypes. Additional studies that examine other biotic and abiotic factors that affect secondary metabolites are needed to fully explain the causes of year-to-year variation in polyphenolic compounds and antioxidant activity in murtilla fruit.

Acknowledgements

The authors are grateful for the financial support provided by Project DIUFRO 120622 of the Universidad de La Frontera and Project FONDEF D05I10086 of the Instituto de Investigaciones Agropecuarias INIA-Carillanca, the Universidad de La Frontera and the Universidad Austral de Chile.

References

Arancibia-Avila, P., Toledo, F., Werner, E., Suhaj, M., Leontowicz, H., Leontowicz, M., Martinez-Ayala, A., Paśko, P., Gorinstein, S. 2011. Partial characterization of a new kind of chilean murtilla-like berries. Food Research International. 44, 2054-2062.

Atoui, A.K., Mansouri, A., Boskou, G., Kefalas, P. 2005. Tea and herbal infusions: their antioxidant activity and phenolic profile. Food Chemistry. 89, 27-36.

Avello, M., Pastene, E. 2005. Actividad antioxidante de infusos de *Ugni molina*e Turcz ("Murtilla"). Boletín Latinoamericano y del Caribe de Plantas Medicinales y Aromáticas. 4, 33-39.

Bocco, A., Cuvelier,M-E., Richard, H., Berset, C. 1998. Antioxidant activity and phenolic composition of citrus peel and seed extracts. Journal of Agricultural and Food Chemistry. 46, 2123-2129.

Bolling, B., Dolnikowski, G., Blumberg, J., Chen, C.Y. 2010. Polyphenol content and antioxidant activity of California almonds depend on cultivar and harvest year. Food Chemistry. 122, 819-825.

Connor, A.M., Luby, J., Tong, C., Finn, C., Hancock, J. 2002. Genotype and environmental variation in antioxidant activity, total phenolic content and anthocyanins content among blueberry cultivars Journal of the American Society for Horticultural Science. 127, 89-97.

de Abreu, I.N., Mazzafera, P. 2005. Effect of water and temperature stress on the content of active constituents of Hypericum brasiliense Choisy. Plant Physiology and Biochemistry 43, 241-248.

Gil, M.I., Tomás-Barberan, F.A., Hess-Pierce, B., Kader, A.A. 2002. Antioxidant capacities, phenolic compounds, carotenoids, and vitamin C content of nectarine, peach, and plum cultivars from California. Journal of Agricultural and Food Chemistry. 50, 4976-4982.

Howard, L., Clark, J., Brownmiller, C. 2003. Antioxidant capacity and phenolic content in blueberries as affected by genotype and growing season. Journal of the Science of Food and Agriculture. 83, 1238-1247.

Instituto de Salud Pública de Chile. 1998. Manual Métodos de Análisis Físico-Químicos de Alimentos, Aguas y Suelo. Andros Ltda., Santiago de Chile, Chile. 187p.

Janda, T., Szalai, G., Rios-Gonzalez, K., Veisz, O., Páldi, E. 2003. Comparative study of frost tolerance and antioxidant activity in cereals. Plant Science. 164, 301-306.

Jiménez-Escrig, A., Rincón, M., Pulido, R., Saura-Calixto, F. 2001. Guava fruit (*Psidium guajava* L.) as a new source of antioxidant dietary fiber. Journal of Agricultural and Food Chemistry. 49, 5489-5493.

Kevers, C., Pincemail, J., Tabart, J., Defraigne, J-O. Dommes, J. 2011. Influence of cultivar, harvest time, storage conditions and peeling on the antioxidant capacity and phenolic and ascorbic acid contents of apples and pears. Journal of Agricultural and Food Chemistry. 59, 6165-6171.

Ksouri, R., Megdiche, W., Falleh, H., Trabelsi, N., Boulaaba, M., Smaoui, A., Abdelly, C. 2008. Influence of biological, environmental and technical factors on phenolic content and antioxidant activities of *Tunisian halophytes* Comptes Rendus Biologies. 331, 865-873.

Lee, J-H., Talcott, S. 2004. Fruit maturity and juice extraction influences ellagic acid derivatives and other antioxidant polyphenolics in muscadine grapes. Journal of Agricultural and Food Chemistry. 52, 361-366.

Mandal, S., Yadav, S., Yadav, S., Nema, R.K. 2009. Antioxidants: a review. Journal of Chemical and Pharmaceutical Research. 1, 102-104.

Montenegro, G. 2002. Chile Nuestra Flora Útil. Universidad Católica de Chile, Chile. 267p.

Rubilar, M., Pinelo, M., Ihl, M., Scheuermann, E., Sineiro, J., Nuñez, M.J. 2006. Murta leaves (*Ugni molinae* Turcz*)* as a source of antioxidant polyphenols. Journal of Agricultural and Food Chemistry. 54, 59-64.

Rubilar, M., Jara, C., Poo, Y., Acevedo, F., Gutierrez, C., Sineiro, J., Shene, C. 2011. Extracts of Maqui (Aristotelia chilensis) and Murta (*Ugni molinae* Turcz): sources of antioxidant compounds and α-Glucosidase/α-Amylase inhibitors. Journal Agriculture Food Chemistry. 59, 1630-1637.

Ruiz, A., Hermosin-Gutierrez, I., Mardones, C., Vergara, C., Herlitz, E., Dorau, C., Winterhalter, P., von Baer, D. 2010. Polyphenols and antioxidant activity of calafate (*Berberis microphylla)* fruits and other native berries from southern Chile. Journal Agriculture Food Chemistry. 58, 6081-6089.

Scheuermann, E. 2009. Estudio de componentes volátiles y capacidad antioxidante de frutos de murta (*Ugni molinae* Turcz) y evaluación de sus estabilidad en almacenamiento refrigerado, congelación y tratamiento térmico de pasteurización. Tesis para optar al grado académico de Doctor en Ciencias y Tecnología de Alimentos, Universidad de Santiago de Chile, Chile. 200p.

Scheuermann, E., Seguel, I., Montenegro, A., Bustos, R., Hormazábal, E., Quiroz, A. 2008. Evolution of aroma compounds of murtilla fruits (*Ugni molinae* Turcz) during storage. Journal of the Science of Food and Agriculture. 88, 485-492.

Seguel, I., Peñaloza, E., Gaete, N., Montenegro, A., Torres, A. 2000. Colecta y caracterización molecular de germoplasma de murta (*Ugni molinae* Turcz.) en Chile. Agro Sur. 28, 32-41.

Shene, C., Reyes, A., Villarroel, M., Sineiro, J., Pinelo, M., Rubilar, M. 2009. Plant location and extraction procedure strongly alter the antimicrobial activity of murta extracts. European Food Research and Technology. 228, 467-475.

Sochor, J., Zitka, O., Skutkova, H., Pavlik, D., Babula, P., Krska, B., Horna, A., Adam, V., Provaznik, I., Kizek, R. 2010. Content of phenolic compounds

and antioxidant capacity in fruits of apricot genotypes. Molecules. 15, 6285-6305.

Sokal, R., Rohlf, J. 1995. Biometry. Third edition. W.H. Freeman and Co., New York, U.S.A. 887p.

Sun, Y-F., Liang, Z-S., Shan, C-J., Viernstein, H., Unger, F. 2011. Comprehensive evaluation of natural antioxidants and antioxidant potentials in *Ziziphus jujuba Mill*. var. spinosa (Bunge) Hu ex H. F. Chou fruits based on geographical origin by TOPSIS method. Food Chemistry. 124, 1612-1619.

Taboada, E., Fisher, P., Jara, R., Zuñiga, E., Gidekel, M., Cabrera, J.C., Pereira, E., Gutierrez-Moraga, A., Villalonga, R., Cabrera, G. 2010. Isolation and characterization of pectic substances from murta (*Ugni molinae Turcz*) fruits. Food Chemistry. 123, 669-678.

Temime, S.B., Wael, T., Bechir, B., Leila, A., Douja, D., Mokhtar, Z. 2006. Changes in olive oil quality of Chétoui variety according to origin of plantation. Journal of Food Lipids. 13, 88-99.

van der Sluis, A.A., Dekker, M., De Jager, A., Jongen, W.M.F. 2001. Activity and concentration of polyphenolic antioxidants in Apple: Effect of cultivar, harvest year, and storage conditions. Journal of Agricultural and Food Chemistry. 49, 3606-3613.

von Gadow, A., Jourbert, E., Hansmann, C.F. 1997. Comparison of the antioxidant activity of aspalathin with that of other plant phenols of rooibos tea (*Aspalathus linearis*). α-Tocopherol. BHT. and BHA. Journal of Agricultural and Food Chemistry. 45, 632-638.

Wang, S.Y., Lin, H.S. 2000. Antioxidant activity in fruits and leaves of blackberry. raspberry. and strawberry varies with cultivar and developmental stage. Journal of Agricultural and Food Chemistry. 48, 140-146.

Wolfe, K., Wu, X., Liu, R.H. 2003. Antioxidant activity of apples peels. Journal of Agricultural and Food Chemistry. 51, 609-614.

Wojdylo, A., Oszmainski, J., Laskowski, P. 2008. Polyphenolic compounds and antioxidant activity of new and old apple varieties. Journal of Agricultural and Food Chemistry. 56, 6520-6530.

Wong, S.P., Leong, L.P., William, J.H. 2006. Antioxidant activities of aqueous extracts of selected plants. Food Chemistry. 99, 775-783.

Xin, Z., Browse, J. 2000. Cold Comfort farm: the acclimation of plants to freezing temperatures. Plant, Cell and Environment. 23, 893-902.

Zar, J. 1999. Biostatistical Analysis. Fourth edition. Prentice Hall, U.S.A. 663p.