# BLUEBERRY POLLINATION IN SOUTHERN BRAZIL AND THEIR INFLUENCE ON FRUIT QUALITY<sup>1</sup>

TIAGO MADRUGA TELESCA DA SILVEIRA<sup>2</sup>, MARIA DO CARMO BASSOLS RASEIRA<sup>3</sup>, DORI EDSON NAVA<sup>4</sup>, MARCELO COUTO<sup>5</sup>

**ABSTRACT** – Blueberry (*Vaccinium ashei*) is a relatively new crop in cultivation under Southern Brazil conditions. The first collection introduced in the area was formed by rabbiteye cultivars which need insect pollinators and also pollinizers. The aim of this work was to observe if there were differences between pollinizers on fruit quality of the commercial cultivar and also to observe the most effective and frequent insect pollinators, under natural conditions. It was concluded that pollen source has an effect on quality of blueberry fruits. Bumblebees are the most efficient pollinators; however the species found in southern Brazil are different from the ones mentioned in the U.S. literature.

**Index terms:** pollinators, metaxenia, rabbiteye blueberry.

## POLINIZAÇÃO EM MIRTILO NO SUL DO BRASIL E SUA INFLUÊNCIA NA QUALIDADE DO FRUTO

**RESUMO** - O mirtilo (*Vaccinium ashei*) é uma espécie relativamente nova em cultivo, nas condições do Sul do Brasil, Pelotas. A primeira coleção introduzida na área era formada por cultivares rabbiteye, que necessitam de insetos polinizadores e também plantas polinizadoras. O objetivo deste trabalho foi observar se havia diferenças entre as polinizadoras na qualidade dos frutos da cultivar comercial e também observar os mais eficazes e frequentes insetos polinizadores, em condições naturais. Concluiu-se que a fonte de pólen tem efeito na qualidade de frutos de mirtilo. As mamangavas são os polinizadores mais eficientes, contudo a espécies encontradas no Sul do Brasil são diferentes dos mencionados na literatura dos Estados Unidos. **Termos para indexação:** polinizadores, metaxenia, mirtilo rabbiteye.

#### INTRODUCTION

Blueberry cultivation is relatively new in Brazil. The first collection introduced in Southern Brazil was exclusively composed by rabbiteye cultivars. On these cultivars it is considered that 96% of the flowers would not form fruits if they are not pollinated by insects (SAMPSON; CANE, 2000). Pollinators are essential for blueberries because of their flower morphology (GALLETTA; BALLINGTON, 1996). The corolla has a shape of a tube or a bell. The flower has 8 to 10 stamens inserted at the base of the corolla, around a long style. The anthers have a poriferous dehiscence and the pollen is released at one end of them (MCGREGOR, 1976). This way, the pollen would fall out of the flowers and not on the stigmatic surface. Insects of the genus *Bombus* are considered better pollinators for Ericaceae family than honeybees due to their ability to buzzpollination (WILLIAMS, 2006). The literature refers to Bombus impatiens Cresson, 1865 (Hymenoptera: Apidae) as the Bombus species found in plantings of the United States. However, since Southern Brazil has a rather different clime from the main American blueberry production areas, it was not known if the insect species visiting blueberry flowers here were the same as the ones occurring there. It is considered that a good crop is obtained when 80% of the blueberry flowers are fertilized and develop fruits (GALLETTA; BALLINGTON, 1996). Rabbiteye cultivars are usually not self-compatible. Thus, besides pollinators they need a good pollinizer cultivar, which means a cultivar compatible with the commercial cultivar, with coincident blooming period, and that produces pollen in abundance, bloom regularly and preferably produce fruits of commercial value (CARVALHO; RASEIRA, 1989).

Another aspect to be considered is that in some species it is possible the occurrence of a phenomenon known as metaxenia. Xenia, including metaxenia, is described as the direct or immediate pollen effect on size, shape, color, development

<sup>&</sup>lt;sup>1</sup>(Trabalho 061-10). Recebido em: 01-03-2010. Aceito para publicação em: 04-01-2011.

<sup>&</sup>lt;sup>2</sup>MSc. in Temperate Fruit Crops, FAEM, UFPel, Pelotas-RS, Rodovia BR 392, km 78, P.O. Base 403, 96001-970. E-mail: telesca.tiagro@gmail.com

<sup>&</sup>lt;sup>3</sup>PhD. Fruit Breeder, Embrapa Clima Temperado, C. P. 403, 96001-970, Pelotas-RS. Bolsista CNPq, E-mail: bassols@cpact.embrapa.br <sup>4</sup>Dr. Researcher, Laboratório de Entomologia da Embrapa Clima Temperado, C. P. 403, 96001-970, Pelotas-RS. nava@cpact.embrapa.br <sup>5</sup>Dr. Researcher EPAGRI, Rua Abílio Franco, Bairro Bom Sucesso, C. P. 591 CEP 89500-000. E-mail: marcelocouto@epagri.sc.gov.br

period of fruits and/or seeds. In other words, it is the direct effect of the pollen on the female parent (DENNEY, 1992). Xenia is widespread in plants and has applications not only in plant genetics and physiology but also in plant breeding and crop production (LIU, 2008).

The present study had two main objectives. The first was to study the effect of different pollinizers on fruit quality and the second one was to identify species of insect pollinators for blueberry, under Southern Brazil conditions.

#### **MATERIAL AND METHODS**

The work was carried out at Embrapa Temperate Climate, located at 31°40 47"S, 52°26,24"W and 60m de altitude, with an average temperature of 17.2 °C and precipitation of 121.8 mm, in Pelotas, state of Rio Grande do Sul, Brazil.

Pollen effect on fruit quality: Two Embrapa's rabbiteye selections 103, and 110, and the cv. Climax were used as mother plants. The plants for this experiment were planted in large vases, and kept during blooming and fruit development period under greenhouse conditions. The pollen used on flowers of Selection 103 were from 'Delite' and 'Briteblue', the ones used on Selection 110 were from 'Bluegen' and 'Powderblue' and on cv. Climax were 'Aliceblue' and selection 110. Fifty flowers per plant were emasculated and pollinated with pollen. The obtained fruits were harvested at ripen stage and were taken to the laboratory where their diameter and soluble solids were measured and the number of seeds per fruit was also counted. Each fruit was individually analysed.

**Observations on pollinators:** Observations were made at the time of full bloom, period from September 19 to September 27 on six plants of rabbiteye blueberry, under field conditions. Each one of them was observed for 5 minutes at each half hour, during the period from 9 am until noon and from 13:30 pm until 14:30 pm. The observations were made on six different days.

Pollen load carried by the observed insect species: Ten insects of each observed group were captured, sacrificed and placed in vials with 4mL of lactic acid 85%, in order to estimate the number of pollen grains carried on their bodies. Once the suspension was homogenized, a drop was placed on a Newbauer slide followed by the counting of blueberry pollen grains and pollen grains of any species. The results were adjusted to 4mL. The percentage of blueberry pollen in relation to the total pollen carried by each insect was also calculated.

**Statistics analysis:** The data was submitted to variance analysis and means were compared by Tukey test at 5% probability. The number of pollen grains and number of seeds were transformed as  $(x+1)^{1/2}$  and the data on percentage were transformed as arcsine  $(x/100)^{1/2}$ 

#### RESULTS AND DISCUSSION

Pollen effect on fruit quality: Significant differences were obtained in fruit diameter in the selection 103, when pollinated by 'Powderblue' or 'Bluegen', being the fruits obtained with the first pollen, larger than the others. The same was true when pollen of 'Britteblue' was used on Selection 110, as compared to 'Delite' pollen. There was no difference on fruit diameter of cv. Climax resulting from pollination with 'Aliceblue' or 'Sel. 110'. However, in reference to total soluble solids (TSS) Selection 110 gave fruits of higher TSS than did with pollen of cv. Aliceblue. 'Powderblue' pollen also resulted in higher total soluble solids in the fruits than did 'Bluegen' pollen on selection 103. There was no difference in sugar content for Selection 110 (Figs. 1 and 2). Rodrigues et al. (2007), analyzed fruit of six cultivars of blueberry (Woodard, Powderblue, Bluegen, Briteblue, Bluebelle and Delite) found that they differed on pH, total solids and total acidity, and Bluegen cultivar showed pH 2.90, total soluble solids 12.2 ° Brix, TA 1.28. Machado et al. (2004) found total soluble solids content of 14.46 ° Brix, pH 1.07 and 2.97 in the cultivar Bluegen.

Larger numbers of seeds were found in fruits originated from the crossing between 'Climax' and selection 110, followed by the crossing between cv. Climax and 'Aliceblue', which differed from the first. Both selections, independent of the utilized pollen, had less seeds that the cv. Climax, and similar fruit size (Fig. 3) which is an advantage of the selections over the cv. Climax. Fruits resulted from the cross between Selection 110 by 'Britteblue' had more seeds than the ones resulted from Selection 110 by 'Delite'. There were no differences between the two pollen types used on Selection 103 for number of seeds in the obtained fruits (Fig. 3). Selection 110 can be considered an interesting genotype for higher TSS. 'Powderblue' is a better pollinizer for Selection 103 than 'Bluegen' because the resultant fruits were larger and had higher TSS content.

The pollen source is considered important for maximum blueberry production. Even on selfertile cultivars, the cross-pollination increases productivity as compared with selfpollination (FREE, 1993), produces more seeds (HARRISON et al., 1993),

higher fruit set (LANG; DANKA, 1991; HARRISON et al., 1993) heavier and larger berries (EL-AGAMY et al., 1981), and earlier ripening of the berries (LYRENE, 1989).

Generally, pollen with more distant parentage produced heavier berries (GUPTON, 1984), and also larger number of seeds (HELLMAN; MOORE, 1983, GUPTON; SPIERS, 1994) than very closely related parents, even though these effects depend of the pollen donator (VANDER KLOET; TOSH, 1984; RABAEY; LUBY, 1988). The direct effect of the pollen on maternal tissues is called metaxenia and, in blueberry, it has been reported for berry size (EHLENFELDT, 2003). The knowledge of metaxenia is very important for planning commercial plantings of blueberry. According to Gupton, 1997, the time of harvest and the fruit development period has even larger pollen influence than the fruit weight. The phenomenon of xenia as already said can be considered as the effect of pollen on the embryo and endosperm, altering the genetic characteristics and providing qualitative and quantitative changes (ANDRADE; PEREIRA, 2005), these features, manifested in the generation of the parent plant (MERCER et al., 2002).

Observations on pollinators: The following species were observed foraging on blueberry flowers: two *Bombus* species, *B. morio* Swederus, and *B. atratus* Franklin, (*Bombinae*); *Xylocopa hirsutissima* Maidl, and *Xylocopa subcyanea*, Peres (Anthophorinae), *Apis mellifera*, Linnaeus, honeybees (Apinae); *Trigona* spp. Fabricius, (Meliponinae) and wasps, *Polybia* sp. Olivier, and *Brachygastra lecheguana*, Latreille, (Vespidae). It is interesting to note that *B. impatiens* the most commonly observed in blueberry planting in the United States was not observed.

There was no interaction between the periods of the day (morning and afternoon) and the days of observations, for the frequency of bumblebees or honeybees visits. The bumblebees were foraging more on the second and third day of observation, it was observed their presence even when temperatures were around (12°C), when temperature was lower (Fig. 4). According to Willmer and Unwin (1981) larger insects can reach and maintain a body temperature higher than the environmental, temperate. Thus they can start flight activities even under low radiation.

During all the days observed, a larger number of *A. mellifera* was found on morning periods coinciding with a higher number of sunlight hours. Afternoons were cloudy on the observed days. The largest number of honeybees was counted on September 24 (Fig. 5 and 6). On that day the

radiation was larger and the temperature and relative humidity were in a range considered as ideal for honeybees foraging. The maximum limit for these insect activities is considered to be 38°C and the minimum 15°C. The optimum is within these limits (GALLO et al., 2002). The number of bee visits, on cherry flowers, was four times higher with the increased of 4°C whereas the reduction of insolation and the increase in wind speed reduced the visits (BARBATTINI et al., 1997).

In the present work, there was significant interaction between the periods and observed days, for Trigona and vesps (Figs. 7 and 8). On September 24 and September 25, the number of visits by vesps was larger in the afternoon, probably because the temperatures were lower in the morning. On September 26 was the opposite, that is, a larger number of visits were observed in the morning period. However on that morning the temperature was mild and the number of insolation was higher. On the other three observed days there were no significant differences between the two periods (Table 1). For Trigona the only differences between periods were observed on September 26 and September 27. On the first, there were more visits in the morning than in the afternoon, probably because the number of insolation hours was higher. On September 27, the number of Trigona present was higher in the afternoon, when the temperature was increased higher and the humidity lower than in the morning period. However, as it can be noticed by the climatic data, the number of this insect species was high when the temperature was around 20°C. Low temperatures and high relative humidity limit the outside activities of small size species of Meliponinae (RAMALHO et al., 1991; AZEVEDO, 1997). The differences among bees species are directly related to the size of their body being small insects more active at higher insolation hours (WILLMER; UNWIN, 1981).

**Pollen load carried by the observed insect species:** There was a significant difference among insects, in relation to the number of pollen grains which they carried on their bodies. The largest number was observed on bumble bees.

The percentage of blueberry pollen grains in relation to the total number of pollen grains was also calculated. On the average, on bumble bees 55.5% of the total pollen on their bodies was blueberry pollen whereas *A. mellifera* blueberry pollen represented only 14.3% of the total. So, honeybees are not faithful as blueberry pollinators, under the conditions of Southern Brazil.

The bumblebees insects have long buccal

parts which allow them to reach relatively deep nectar glands as in blueberry flowers. They have also a hairy body and they tighten themselves very strongly to the flowers and with wing movement they cause vibration and consequently the pollen is released by the anthers (WILLIAMS, 2006).

For an insect to be considered a preferred pollinator of a crop it has to be attracted to their flowers, it has to have a body size and behavior suitable to promote pollen release and favors the pollen deposit on the stigmatic surface. The insect has to transport in its body a good amount of pollen and visit the flowers when they are receptive and before the beginning of ovules degeneration.

The insect has also to be faithful to that vegetable species (FREE, 1993; FREITAS; PAXTON, 1996). Considering all these aspects, honeybees are not the best pollinators for blueberry. Bumblebees had a percentage of blueberry pollen, in relation to the total load they carry, three to four times greater than that of honeybees proportion. It was expected that the pollen load carried by them would be larger due to their body size, however, the percentage of this pollen which is blueberry pollen was 55.5%, which in comparison with honeybees (only 14.3%) showed that the last ones are not a liable pollinizer for blueberries when other species are blooming at coincident periods.

**TABLE 1** – Climatic data on the days of observations of blueberry pollinizers (morning period 9 am to 12 am; afternoon 1:30 pm to 4:30 pm). Embrapa Clima Temperado. Pelotas-RS.

	,	1	1 / 1	1	
Days	Period	Wind speed	Insolation	Temperature	humidity
		Km h-1	h	°C	%
18/09	M	3	_	15,5/16/16,5/18	65/60,5/55/50
	A	-	1.3	20/20/20/20	50/45/45/45
24/09	M	0.6	2.7	12/12/12/14	50/50/47/45
	A	-	2.5	14.5/14,5/15/16	45/45/45/45
25/09	M	0.9	3	18/18/19/21	60/55/54/50
	A	-	2.3	21/21/21/21	45/45/45/45
26/09	M	2	3	14/16/17/18	80/65/70/70
	A	-	1.5	19/20/20/20	70/60/60/60
27/09	M	1.4	2.1	15/16/17/19	75/65/55/55
	A	-	0.5	20/21/21/21	55/55/55/55
28/09	M	0.8	3	15/20/21/21	65/60/55/55
	A	-	3	21,5/22/22/21,5	50/50/49/45

M= morning A= afternoon

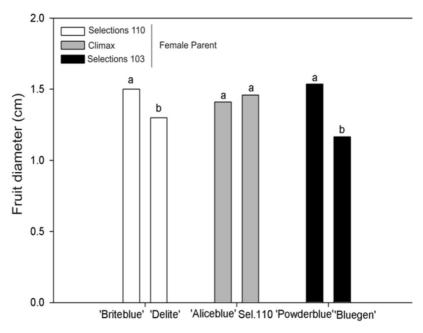
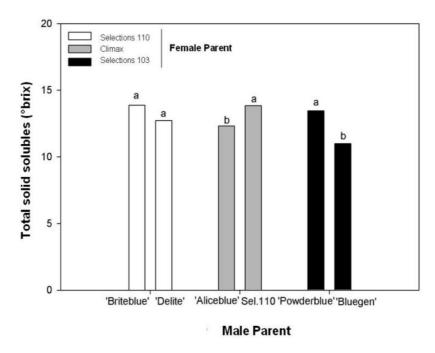
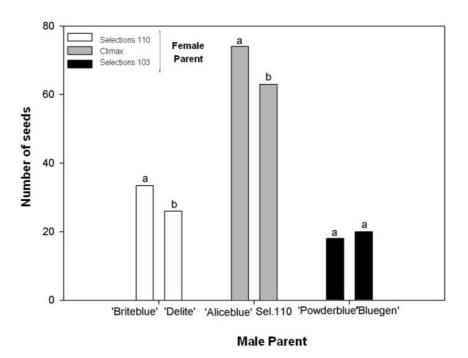


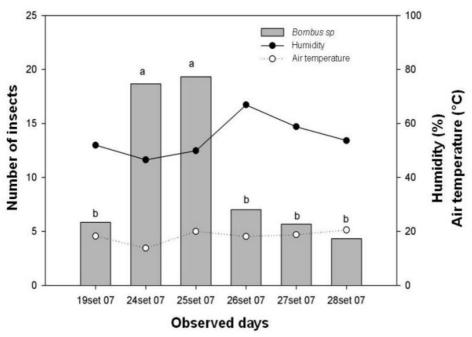
FIGURE 1 – Average diameter of fruit from different crosses. Embrapa Clima Temperado.



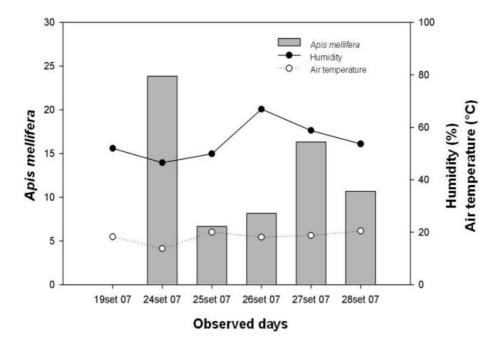
**FIGURE 2** – Average soluble solids content in fruits of different blueberry crosses. Embrapa Clima Temperado.



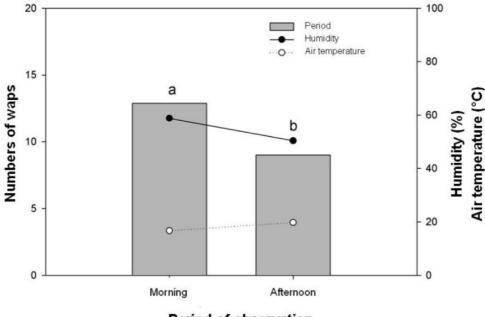
**FIGURE 3** – Average number of seeds of fruits from different blueberry crosses. Embrapa Clima Temperado.



**FIGURE 4** – Average number of Bombus sp. Observed on blueberry plants in the day periods morning and afternoon, days of evaluation and average daily temperature and relative humidity. Embrapa Clima Temperado.



**FIGURE 5** – Average number of Apis mellifera. Observed on blueberry plants in the day periods morning and afternoon, days of evaluation and average daily temperature and relative humidity. Embrapa Clima Temperado.



### Period of observation

**FIGURE 6** –Average number of Apis mellifera. Observed on blueberry plants in the day periods morning and afternoon, days of evaluation and average daily temperature and relative humidity. Embrapa Clima Temperado.

#### CONCLUSIONS

- 1 The pollen source has an effect on the fruit quality of blueberries fruits.
- 2 Bumblebees (*Bombus* sp.) are the most efficient blueberry pollinizers under Southern Brazil conditions but the species found on this area differ from the ones observed in the USA.
- 3 Honeybees (*Apis mellifera*) have a complementary role to the bumblebees' activity in blueberry pollination.

#### REFERENCES

ANDRADE, J.A.C.; PEREIRA, F.C.D. Uso do efeito xênia em híbridos comerciais de milho (*Zea mays* L.). **Revista Brasileira de Milho e Sorgo**, Sete Lagoas, v.4, n.3, p.65-78, 2005.

AZEVEDO, G.G. Atividade de voo e determinação do número de instares larvais em Partamona helleri (Friese) (Hymenoptera, Apidae, Meliponinae). 1997. 64 f. Dissertação (Mestrado em Entomologia) - Universidade Federal de Viçosa, Viçosa, 1997.

BARBATTINI, R.M.; GREEATTI, M.; SABATINI, A.G.; MARCAZZAN, G.L.; COLOMBO, R. 1997. Apiarium interest of *Metcalfa pruinosa* (Say). Production and Physiochemical Characteristics of Honey Derived from its Honeydew. In: INTERNATIONAL CONGRESS OF APICULTURAL, 33., 1993, Bucarest. **Proceedings...** p.240-243.

CARVALHO, T.C.P.; RASEIRA, M.C.B. Período de floração e percentagem de frutificação efetiva em cultivares de Ameixeira-Japonesa (*P. salicina*, Lindl) no Rio Grande do Sul. Pelotas: Embrapa Clima Temperado, 1989. 14p. (Boletim de Pesquisa, 16).

DENNEY, J.O. Xenia includes metaxenia, **HortScience**, Alexandria, v. 27, p.722-727, 1992.

EHLENFELDT, M.K. Investigations of metaxenia in Northern Highbush Blueberry (*Vaccinium corymbosum* L.) Cultivares, **Journal of the American Pomological Society,** Urbana, v.57, n.1, p.26-31, 2003.

El-AGAMY, S.Z.A.; SHERMAN, W.B.; LYRENE P.M. Fruit set and seed number from self-and cross-pollinated highbush (4x) and rabbiteye (6x) blueberries. **Journal of American Society for Horticultural Science**, Mount Vernon, v.106, n.4, p.443-445, 1981.

FREE, J.B. **Insect pollination of crops**. London: Academic Press, 1993. 684p.

FREITAS, B.M.; PAXTON, R.J. The role of wind and insects in cashew (*Anacardium occidentale*) pollination in NE Brazil. **Journal of Agricultural Science**, Cambridge, v.126, n.3, p.319-326, 1996.

GALLO, D.; NAKANO, O.; SILVEIRA NETO, S.; CARVALHO, R. P. L.; BAPTISTA, G. C.; BERTI FILHO, E.; PARRA, J. R. P.; ZUCCHI, R.A.; ALVES, S. B.; VENDRAMIM, J. D.; MARCHINI, L. C.; LOPES, J.R. S.; OMOTO, C. Entomologia agrícola. Piracicaba: FEALQ, 2002. 920p.

GALLETTA, G.L.; BALLINGTON, J.R. Blueberries, Cranberries and Loganberries. In: JANICK, J.; MOORE, J.N. Ed. **Fruit breeding**: vine and small fruit crops. New York: John Wiley & Sons, 1996. v.2, p.1-107

GUPTON, C.L. Effect of pollen source on frit characteristics of low chiling highbush type blueberries. **HortScience**, Alexandria, v.19, n.4, p.531-532, 1984.

GUPTON, G.L. Evidence of xenia in blueberry. **Acta Horticulturae**, The Hague, v.446, p.119-123, 1997.

GUPTON, C.L.; SPIERS, J.M. Interspecific and intraspecific pollination effects in rabbiteye and southern highbush blueberry. **HortScience**, Alexandria, v.29, p.324-326, 1994.

HARRISON, R.E.; LUBY, J.; ASCHER, P.D. Pollen source affects yield components and reproductive fertility of four half-high blueberry cultivars. **Journal of the American Society for Horticultural Science,** Mount Vernon, v.119, n.1, p.84-89, 1993.

HELLMAN, E.W.; MOORE, J.N. Effect of genetic relationship to pollinizer on fruit, seed, and seedling parameters in highbush and rabiteye blueberry. **Journal of American Society for Horticultural Science**, Mount Vernon, v.108, n.3, p.401-405, 1983.

LANG, G.A.; DANKA, R.G. Honey-bee-mediated cross-versus self-pollination of "Sharpblue" blueberry increases fruit size and hastens ripening. **Journal of the American Society for Horticultural Science**, Mount Vernon, v.116, n.5, p.770-773, 1991.

LIU, Y. A Novel Mechanism for xenia? **HortScience**, Alexandria, v. 43, n. 3, 706, 2008.

LYRENE, P. M. Pollen source influences fruiting of "Sharpblue" blueberry. **Journal of the American Society for Horticultural Science**, Mount Vernon, v.114, n.6, p.995-999, 1989.

MACHADO, N.P.; FRANCHINI, E.R.; RISTOW, N.C.; COUTINHO, E.F.; CANTILLANO, F.R.F.; MALGARIN, M.B. Conservação pós-colheita de mirtilos Flórida, Woodard e Bluegen em atmosfera

com oxigênio ionizado. In: SIMPÓSIO NACIONAL DO MORANGO, 2., ENCONTRO DE PEQUENAS FRUTAS E FRUTAS NATIVAS DO MERCOSUL, 1., 2004. Pelotas. **Anais...** Pelotas: Embrapa Clima Temperado, 2004. p.300-304.

McGREGOR, S.E. Insect pollination of cultivated crop plants. Washington: USDA, 1976. p.411. (Agriculture Handbook, 496)

MERCER, J.R.; RAMALHO, M.A.P.; RAPOSO, F.V. Implicações do fenômeno de xênia nos programas de melhoramento de milho. **Ciência Agrotécnica**, Lavras, v.26, n.6, p.1338-1343, 2002.

RAMALHO, M.; IMPERATRIZ-FONSECA V.L.; KLEINERT-GIOVANNINI, A. Ecologia nutricional de abelhas sociais. In: PANIZZI, A.R.; PARRA, J.R.P. Ecologia nutricional de insetos e suas implicações no manejo de pragas. São Paulo: MANOLE, 1991. p.225-252.

RABAEY, A.; LUBY, J. Fruit set in half-high blueberry genotypes following self and cross pollination. **Fruit Varieties Journal**, Urbana, v.42, n.4, p.126-129, 1988.

RODRIGUES, S. A.; GULARTE, M.A.; PEREIRA, E.R.B.; BORGES, C.D.; VENDRUSCULO, C.T. Influência da cultivar nas características físicas, químicas e sensoriais de *topping* de mirtilo. **Revista Brasileira de Tecnologia Agroindustrial**, Campinas, v.1, n.1, p.9-29, 2007.

SAMPSON, J.B.; CANE, J.H. Pollination efficiencies of tree bee (Hymenoptera: Apoidea) species visiting Rabbiteye Blueberry. **Journal of Economic Entomology**, College Park, v.93, n.6, p.1726-1731, 2000.

VANDER KLOET, S.P.; TOSH, D. Effect of pollen donors on seed production, seed weight, germination and seedlings vigor in *Vaccinioum corymbosum* L. **American Midland Naturalist**, Notre Dame, v.112, n.2, p.392-396, 1984.

WILLIAMS, I.H. Insect pollination and crop production: a European perspective. In: KEVAN, P.G.; IMPERATRIZ-FONSECA, V.L. **Pollinating bees:** the conservation link between agriculture and nature. 2. ed. Brasília: Ministério do Meio Ambiente, 2006. p.59-65.

WILMER, P.G.; UNWIN, D.M. Field analyses of insect heat budgets: reflectance, size and heating rates. **Oecologia**, Berlin, v.50, n.2, p.250-255, 1981.