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### Apple Phenology in Subtropical Climate Conditions

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#### 1. Introduction

There are two phases that characterize the annual cycle of apple (*Malus domestica* Borkh.): hibernal period (dormancy) and shoot growth. Temperature is the main factor related to dormancy, specifically low temperatures. The evolution of the apple phenological stages during the vegetative phase: induction and flower differentiation, budbreak, full bloom, fruit development and maturation, and yield and production quality, may be visually detected and are affected by climate conditions, mainly by temperature.

Plant-related factors such as cultivar and rootstock also influence this evolution dynamic. The type of the fruit structure and its foliar area affect the fruit formation, and are important parameters to the phenological evolution. Growing apples in the warm winter regions where the chilling requirement is not adequate, can cause the trees to develop a series of anomalies in the phenology referent to budbreak, flowering, growth and development of both fruit and tree (Skinner, 1964; Saure, 1985; Chariani & Stebbins, 1994; Bulon & Faust, 1995; Petri et al., 1996; Iuchi, 2002; Petri & Leite, 2004).

At the end of the dormancy, vegetative and reproductive apple buds evolve and pass through different external phases that are known as phenological stages. According to Saraiva (1973), the study of phenology is based on observations and measurements of a series of plant organs in a determined period. This constitutes an essential element to evaluate the adaptability of fruits species under particular environmental conditions.

The knowledge of the phenological stages is important under a practical viewpoint to manage some cultural practices such as fruit thinning and phytosanitary treatments. These phenological changes have been intensely studied and classified according their development from dormancy to fruitset. Flower differentiation, phenological stages, growth and fruit maturation could be correlated with the climate variables, making possible a forecast of each evolution by mathematical equations. The study of the variability indexes of the evolution of the flower phenological stages are also important, since may indicate the reproductive regularity of cultivars as well as to identify cultivars with coincidence of flowering to be able to recommend pollinators.

The methodology of correlations between chilling units and date of flowering allows to predict in advance the flowering date and consequently to estimate the date of maturation. Estimation of the growth and development of fruits is fundamental in warm winter regions because it is not only related to fruit caliber, but also to alterations that environmental conditions could cause regarding visual aspects mainly fruit form.

#### 2. Flower induction and flower bud development in apple trees

Apple trees have two distinct phases regarding their metabolism: the period of dormancy or vegetative phase. Both are affected by climate conditions, mainly by temperature which leads to an alteration in the phenological phases, including induction and flower differentiation.

Induction and differentiation of the apple flower buds take place just after the last year's flowering. Although it is possible to distinguish the different fruiting organs for the external appearance sometimes buds might not differentiate into flower bud. The flower formation coincides temporally with the shoot and fruit formation. So, that means fruitset affects the metabolism of young shoots and furthermore can inhibit the flowering the following year.

Analysis of bud permits to define the flower percentage and quality. The predicted knowledge of the flower bud formation will give important information about pruning intensity, fruit thinning, fertilization and pollination. In addition, the retrospective of last year's production also aids in the cultural practices to be adopted.

Flowering intensity is one of the parameters to set pruning. Consequently, to minimize the possible faults of this practical, however it does not full assure the fruitset since the latter is year dependent. The quantification of the flower buds will permit to make an equilibrated pruning, avoiding a winter heavy pruning what would promote a large vegetative growth and therefore nutrients competition with the new fruits. Excessive tree vigor reduces fruit set and flower induction.

The process of induction and flower differentiation is affected by several factors such as climatic, nutritional, cultural, physiological and genetic. Flower induction is privileged by the presence of leaf area and negative influenced by excessive fruits on tree. Tree defoliation before induction, excessive fruits and later fruit thinning prejudice the flower induction process.

The period of the apple flower induction occurs in the beginning of shoot growth which means around 45 to 60 days after full bloom. However, there are evidences that induction may occur later, even after fruit harvest in warm regions. As reported by Petri (2002) the period of induction can vary according to cultivar, bud position on tree, climate conditions and nutritional factors. Most flower induction occurs in the beginning of summer but may be extended up to begin of fall under subtropical conditions.

After flower induction flower differentiation takes place. It proceeds throughout vegetative cycle until the next year flowering. The sequence of the differentiation process begins with the appearance of sepals, then stamens, ovary, anthers, pollen and ovule. Since ovary and anthers are formed it is possible to distinguish the flower buds using a stereoscopic microscope.

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Though the external appearance of most apple fruit organs is an indicative of flower bud, precise information about the real percentage of flower bud and on which organ or branch it is located is important to the pruning purposes. Old spurs tend to produce small fruits of low quality in relation to new spurs developed from three or younger year old branches

The knowledge of the fruiting habit is important to the definition of cultural practices such as pruning, thinning and the use of growth regulators, aiming higher productions and fruit quality. Several studies have been developed on which type of flower structure is responsible for the formation of the largest fruit with commercial quality (Madail et al., 2010). Apple fruit buds are mixed buds, which means presence of flowers and leaves in the same bud. They are borne terminally or laterally on fruiting structure classified as brindle, and terminally on spurs. Brindles (10-40cm) are formed in the previous year and spurs are two or more year old buds. Axillar buds are located in the insertion of leaves along the brindles (Petri & Leite, 2006).



Fig. 1. Flower bud (left) and vegetative bud (right) of apple trees.

Leaf area is a factor of great importance to the fruit development, to the cell division process and to the fruitset. Fruiting structure with larger leaf area shows higher photosynthetic capacity, higher production of assimilates and lesser competition among the different tree organs (Costes, 2003). Nevertheless, other authors affirm that the number of leaves is not sufficient to determine the bud performance (Lauri & Trottier, 2004; Lauri et al., 2006). The bud position in the canopy influences the fruit form. Webster (1976) observed that terminal buds of brindles of 'McIntosh' apples provided more elongated fruits than fruits from axillary buds. Rodrigues & Rodrigues (1977) verified better fruits formed in the basal portion of the brindle, since at this position the size of the fruiting structure and leaf area are larger than in the brindle tip.

From 15<sup>th</sup> January under the climate conditions of Southern Brazil, it was possible to identify flower and vegetative buds using a stereoscopic microscope (Fig. 1). In this case, the cultivar Gala already had showed well developed flowers, whereas in the cultivar 'Fuji' there was a

delay in flower development. There is little difference among fruiting structures regarding percentage of flower buds (Table 1). It is observed that brindle and spurs showed little difference for both cultivars, being numerically superior in spurs. This could cause a delay in the stopping of shoots growth.

Cultivor	Fruiting structure	s
Cultivar	Brindle	Spur
Gala	71.6	88.3
Fuji	75.0	78.3

Table 1. Flower buds percentage in Gala and Fuji apples under subtropical climate conditions. Caçador, Santa Catarina, Brazil, 2009.

	,	'Gala'	'Daiane'		
Fruiting structures	Leaf area	Fruit diameter	Leaf area	Fruit diameter	
	(cm <sup>2</sup> )*	(mm)	(cm <sup>2</sup> )	(mm)	
1. Weak spur	34.3 c	56.4 c	43.6 c	51.0 c	
2. Intermediate spur	60.4 c	62.1 b	58.4 c	57.3 b	
3. Strong spur	107.2 b	66.2 a	103.1 b	65.9 a	
4. Brindle	323.9 a	66.5 a	346.9 a	67.8 a	

\*Means followed by same letter do not differ by Duncan test (p<0.05)

Table 2. Leaf area and fruit diameter of 'Gala' and 'Daiane' apples in different fruiting structures. Caçador, Santa Catarina, Brazil, 2009.



Fig. 2. Percentage of the return of bloom at brindles in different defoliation dates in the apple cvs. Gala, Fuji and Daiane. Caçador, SC, 2009.

Tree precocious defoliation affects negatively the following year's crop. At Fig. 2, it is possible to observe a reduction in the return bloom when defoliation was done up to 16<sup>th</sup> February in the cvs. Gala, Fuji and Daiane. The reduction was higher in fruiting spurs than in brindle (Table 2). Up to 16<sup>th</sup> February the reduction of the return bloom was higher in 'Gala' and 'Fuji'. From 16<sup>th</sup> February reduction was lesser accentuated in the brindles of Daiane (Table 2). This difference among cultivars could be attributed to the longer differentiation process seen in the cvs. Gala and Fuji. However, whether defoliation was done on 1<sup>st</sup> May the return of bloom was higher, pointing out that at this time the flower differentiation had already occurred. The results show that the apple flower induction process under warm climate conditions is prolonged for a period superior to 60 days after full bloom. Also, it was observed that the flower induction of axillary buds is associated to the growth period, mainly in the cultivar Gala what shows higher flowering intensity in the axillary buds whether compared to 'Fuji' and 'Daiane'.



Fig. 3. Mean fruit weight (g fruit-1) and seed per fruit in different bud types of 'Fuji' apples. Caçador, Santa Catarina, Brazil, 2009. Means followed by same letter do not differ by Tukey test (p<0.05).

Treatmont	Fruit diameter									
Treatment	23/11*	05/12	20/12	03/01	20/01	11/04	C.V. (%)			
1 fruit/cluster	21.4 abF	27.8 abE	34.3 abD	39.9 abC	45.4 aB	59.6 aA	14.9			
2 fruit/cluster	21.3 abF	<b>2</b> 8.1 aE	35.4 aD	41.5 aC	46.5 aB	59.1 aA	13.0			
3 fruit/cluster	22.6 aF	28.9 aE	35.4 aD	41.0 aC	45.5 aB	61.0 aA	8.64			
4 fruit/cluster	19.9 bE	27.2 abD	34.3 abC	39.4 abB	43.6 abA		13.0			
5 fruit/cluster	19.3 bE	25.7 bD	32.0 bC	37.3 bB	41.1 bA		13.4			
C.V. (%)	17.8	12.8	-12.1	11.4	11.0	14.1				

\*Means followed by lowercase letters in the column, and means followed by uppercase letter in the line, do not differ by Duncan test (p<0.05)

Table 4. Fruit diameter according the number of fruits per cluster in Fuji apple trees. Caçador, Santa Catarina, Brazil, 2008.

Leaf area is variable, depending on fruiting structure. It was observed reduced leaf area in fruiting spurs, principally in weak spurs (Table 2). The largest leaf area is found in brindles where the largest fruits are produced. This increase in leaf area is not only determined by the size of leave, but also by the number of leaves, being smaller in weak spurs e higher in brindles (Table 2). The type and quality of fruiting organs also affect fruitset. Madail et al. (2010)

observed a significant increase of fruitset in longer brindles when compared to spurs. Larger leaf area structures also showed larger-diameter fruits, showing a direct relation between these two variables (Table 2). The lowest fruit fresh mass was observed in axillary buds, followed by spurs and brindles. The difference could be related to the number of seed per fruit which was superior in brindles whether compared to spurs and axillary buds (Fig. 3).

The results of mean diameter of fruits, growth index and fresh mass of fruits in relation to the number of fruits per inflorescence showed little differences. The presence of one or five fruits per inflorescence showed similar results (Table 4, 5 and 6). So, at fruit thinning and pruning period it must be given preference to fruit production in fruiting structure such as brindles and vigorous spurs.

	Fruit growth rate (%)									
Treatment	23/11-	05/12-	20/12-	03/01-	20/01-					
mentiterit	05/12*	20/12	03/11	20/01	11/04	C.V. (%)				
	(12 days)	(15 days)	(13 days)	(17 days)	(81 days)					
1 fruit/cluster	131.2 aA	123.4 aB	116.0 aC	113.9 aC	134.8 aA	5.8				
2 fruit/cluster	136.4 aA	124.4 aB	116.3 aBC	112.5 abC	135.8 aA	8.0				
3 fruit/cluster	129.3 aA	123.0 aB	116.6 aC	110.3 bD	122.8 aB	3.4				
4 fruit/cluster	139.5 aA	126.2 aB	117.5 aC	112.4 abC	_	8.0				
5 fruit/cluster	135.6 aA	124.7 aB	116.9 aC	112.8 abC	_	7.7				
C.V. (%)	10.41	3.52	3.60	2.8	8.3					

\*Means followed by lowercase letters in the column, and means followed by uppercase letter in the line, do not differ by Duncan test (p<0.05)

Table 5. Fruit growth rate according the number of fruits per cluster in Fuji apple trees. Caçador, Santa Catarina, Brazil, 2008.

	Harvest 14/11/2006								
Treatment	Fruit diameter (mm)*	Fruit length (mm)	Mean fruit weight (g)						
1 fruit/cluster	59.6 a	47.6 a	104.3 a						
2 fruit/cluster	59.1 a	49.6 a	112.9 a						
3 fruit/cluster	61.0 a	54.4 a	147.0 a						
C.V. (%)	14.1	14.9	40.7						

\*Means followed by same letter do not differ by Duncan test (p<0.05)

Table 6. Diameter, length and mean fruit weight of Fuji apples according the number of fruits per cluster in Fuji apple trees. Caçador, Santa Catarina, Brazil, 2008.

#### 3. Apple phenological stages

The method used to determine the evolution of the phenological stages consists in the observation of the different stages of the buds, permitting to determine chronologically their development (Fig. 4). The result will provide information about the flower biology, will



Fig. 4. Apple phenological stages: A (dormant bud); B (silver tip); C-C3 (green tip); D-D2 (bud bursted); E (green leaf); E2 (pink); F-F2 (full bloom); G (beginning of petal fall); H (completely petal fall); I (fruit set); and J (fruit development).

Phenological		Anı	nual me (days)	ans	Du (1	Duration (days)		
Stages	1995	1996	1998	1999	2000	Mean	Accumu- lated	dates*
B e C	-	2	4	3	5	3.5	3.5	15/set.
C3	5	2	3	2	3	3.0	6.5	18/set.
D	3	1	2	2	2	2.0	8.5	21/set.
E	2	1	2	2	2	1.8	10.3	23/set.
E2	3	2	3	2	2	2.4	12.7	24/set.
F	2	3	4	5	3	3.4	16.1	26/set.
F2	2	2	3	3	3	2.6	18.7	29/set.
G	3	2	2	3	3	2.6	21.3	2/out.
Н	3	2	2	3	4	2.8	24.1	4/out.
Ι	5	6	4	7	4	5.2	29.3	7/out.
J	3	6	5	7	-	5.2	34.5	12/out.
Σ	31	29	34	39	31			

\*Referent to the mean of five years

Table 7. Duration of the different phenological stages of the cultivar Fuji during five years in Caçador, SC, Brazil.

make possible to compare the capacity of adaptation of the different cultivars in a same region and to verify the buds development and the influence of environmental factors, and moreover will contribute to some cultural practices such as fruit thinning, pollination and pest and diseases control.

Phenological		Annual (day	means ·s)		Du (d	Mean	
Stages	1995	1998	1999	2000	Mean	Accumu- lated	dates*
С			<u>2</u>	4	3.0	3.0	20/set.
C3	4	3	3	2	3.0	6.0	21/set.
D	3	2	2	2	2.3	8.3	24/set.
Е	2	2	2	2	2.0	10.3	26/set.
E2	2	4	3	4	3.3	13.6	28/set.
F	2	4	3	3	3.0	16.6	1/out.
F2	3	3	3	2	2.8	19.4	4/out.
G	2	2	3	3	2.5	21.9	6/out.
Н	2	3	4	2	2.8	24.7	9/out.
Ι	5	4	5	4	4.5	29.2	1/out.
J	4	4	7	-	5.0	34.2	17/out.
Σ	29	35	37	28			

\*Referent to the mean of four years

Table 8. Duration of the different phenological stages of the cultivar Golden Delicious during four years in Caçador, SC, Brazil.

Phenological		An	nual me (days)	ans	Du (d	Duration (days)		
Stages	1995	1996	1998	1999	2000	Mean	Mean Accumu- lated	
B e C	-	3	3	3	1	2.5	2.5	18/set.
C3	5	2	1	3	2	2.6	5.1	19/set.
D	4	2	2	3	2	2.6	7.7	21/set.
Е	2	2	2	2	2	2.0	9.7	24/set.
E2	3	3	3	3	2	2.8	12.5	26/set.
F	2	3	5	5	3	3.6	16.1	29/set.
F2	2	2	4	3	3	2.8	18.9	2/out.
G	3	2	2	3	2	2.4	21.3	5/out.
Н	3	4	3	3	3	3.2	24.5	8/out.
Ι	4	4	4	7	7	5.2	29.7	11/out.
J	3	6	5	9	-	5.8	35.5	16/out.
Σ	31	33	34	44	27			

\*Referent to the mean of five years

Table 9. Duration of the different phenological stages of the cultivar Gala during five years in Caçador, SC, Brazil.

The duration of the stages varies according year and it is influenced by climatical factors. The mean duration of the phenological stages from silver tip (B) to fruit development (J) varied from 31 to 44 days in apples 'Gala'. The mean date of the occurrence of each phenological stage was estimated during five years of observation for the cvs. Fuji and Gala and four years for cv. Golden Delicious (Tables 7, 8 and 9). These variations were associated to the degree days (DD), the higher thermal amplitude the higher the sum of DD (see session 5).

In subtropical regions, the insufficient chilling accumulation required to the physiological necessities of the buds induces to a prolonged bloom period whether compared to temperate regions (Fig. 5). Erratic budbreak and flowering observed in warm regions is originated by the maximization of the heterogeneity of the buds regarding to chilling requirement and to the negative influence of the paradormancy promoted by the vigorous and advanced buds (Leite et al., 2004).



Fig. 5. Budbreak of the different buds along the brindle (t – terminal bud, m – median bud and b – basal bud) under natural conditions of chilling (left) and privation of chilling (right).

#### 4. Effect of rootstocks and interstocks on apple phenology

In most economically explored temperate fruit species the use of rootstocks is an usual practice. The benefits of rootstocks are related to tree vigor control, precocious fruiting and high quality fruits (Jackson, 2003). The use of different rootstocks influences differently the vegetative and reproductive development of the trees and probably it would exert influence on the physiological process of the canopy dormancy (Erez, 2000).

Exposing six apple rootstocks to chilling privation conditions, with the previous exposition of the scion to chilling, Young & Werner (1985) verified deficient development of budbreak of the scion cultivar. Contradictory, Erez (2000) did not observed the same response in peaches under similar experimental conditions tested by Young & Werner (1985). In order to Finetto (2004) rootstocks can have significant contribution in apple budbreak when scion was submitted to conditions of insufficient winter chilling accumulation. The same author evaluated 'Golden Delicious' apples grown on different rootstocks in Italy and observed the rootstocks affect the chilling requirement of the scion cultivar when conditions were not sufficient. Under Brazilian conditions, Couvillon et al. (1984) verified that apples of high chilling requirements such as Rome Beauty can grow and develop according to the rootstock chilling requirement. According Hauagge & Cummins (2000), the chilling

requirement of the apples rootstocks is an important factor that affects budbreak of 'Gala'. The use of the rootstocks 'MM111', 'M9' and 'M26' caused a delay on budbreak compared to 'MM106' and 'M7'; whereas the use of seedlings cultivar Anna tends to anticipate the budbreak.

The effect of rootstocks in the scion dormancy can result differences in the phenology of apples cultivars. The combination rootstock/scion alters the date of the beginning of flowering as well as the period of flowering (Fig. 6). 'Gala' raised on the rootstock 'Marubakaido' showed a delay in the beginning of flowering in comparison to 'Fuji' on the same rootstock. The duration of scion flowering on 'Marubakaido' was longer for 'Gala' than for 'Fuji', evidencing the scion cultivar dependence.

The use of 'M9' as interstock on the rootstocks 'MI.793', 'M.103' and 'Marubakaido' brings forward the beginning of flowering of the cultivar Fuji in comparison to the no use of interstock. Similar results were found by Scarpare Filho et al. (2000) who observed that the presence of plums interstock 'Januaria' anticipated flowering and budbreak of the peaches cultivars Tropical and Ouromel-2. Tomaz et al. (2010) also observed an influence of the length of the interstock on the duration of flowering; the longer the interstock the more precocious was flowering.



Fig. 6. Flowering period of 'Gala' and 'Fuji' apples grafted on different rootstocks and interstocks. Caçador, Santa Catarina state, Brazil, 2005.

#### 5. Duration of flowering and degree-days

The physiological process and functions of the plants occur under thermal limits in their developing environmental. In order to complete every physiological subperiod of the life cycle some crops require an accumulation of certain heat amount. It is commonly expressed by degree-days index and represents the thermal sum above the minimum base temperature

to the development. To complete each subperiod of the development plants need among other factors a thermal sum (Lozada & Angelocci, 1999).

The concept of degree-days admits that there is a base temperature which below plant growth and development are interrupted or extremely reduced. Besides, it is presupposed a linear relation between temperature and plant development since there is no limitation of other factors (Brunini et al., 1976). The sum of degree-days that plant requires to complete a subperiod or the whole cycle has been used to characterize the life cycle of the plants instead of the number of days. Also it is assumed as constant and independent from local and sowing time.

For all phenological phase it is necessary to calculate the index of heat units differently during the period, because the heat unit sum is one of the basic parameters that characterize the duration of the period from a phenological stage to another (Yazdanpanah et al., 2010).

Several models or expression have been suggested to the degree-days calculation or thermal units required to the plant to reach a certain stage of its development cycle (Brown, 1970; Arnold, 1959).

The heat unit index to each stage can be calculated using the following equation:

$$GD = \sum_{i=1}^{n} \left( \frac{T \max + T \min}{2} - Tb \right)$$

where: GD (DD) is the total accumulated degree-days; Tmax is the daily maximum air temperature (°C); Tmin is the daily minimum air temperature (°C); Tb is the base temperature (°C), normally used as 4.5°C according Richardson et al. (1975); n is the number of days of the flowering period.



Fig. 7. Relation between duration in days and growing degree days of blooming period of 'Lisgala' and 'Condessa' apples during seven years of phenological observations. Caçador, Santa Catarina, Brazil.

Under subtropical conditions there is a high correlation between the duration of flowering period (in days) with the accumulated thermal sum. It can be observed in the Fig. 7 a high correlation between the duration of flowering and thermal sum in apples 'Lisgala' and 'Condessa' during seven years of phenological observation.

The relation between the duration of flowering and thermal sum is also observed among the subperiods of flowering. Considering the phenological observations of eight wild species (*Malus* spp.) in a period of eight years, it can be observed in the Fig. 8 a high correlation between the duration of full bloom to the end of flowering with the thermal sum, in the same magnitude of the period comprised between the begin and the end of flowering.

#### 6. Phenology of wild apple species in subtropical climate conditions

The study of phenology has had great importance in the new apple planting system using high density. Wild apples have been used as pollinators for commercial cultivars due to their flowering intensity.

Aiming to define some pollinators Petri el al. (2008) studied the phenological behavior of trees belonging to 13 apple wild species (*M. atrosanguinea, M. baccata, M. eleyi, M. floribunda, M. hopa, M. platycarpa, M. robusta,* 'John Downil', 'Prof. Spengler', 'Milalew imuni', 'Profusion', 'Winter gold' and 'Yellow Siberian') and two commercial cultivars Gala and Fuji. The measurements were done from 2001 to 2007 in four trees per cultivar. It was recorded the dates of beginning of flowering, full bloom and end of flowering of each species. The beginning of flowering was considered when trees showed 5% of open flowers; full bloom when 80% of the flowers were opened and end of flowering with the last flowers open.

The period of flowering and the percentage of coincidence of flowering among the wild species with the cultivars 'Gala' and 'Fuji and their differences regarding the dates of full bloom were estimated for each year. At the end of the evaluation the mean behavior and the standard deviation for the percentage of coincidence among flowering and for differences among flowering date were estimated and the index of variability was obtained for each species. The index of variability is used as an estimative of regularity of the species or cultivars assessed throughout years. Thus, it could be identified the species/cultivars that showed great stability in relation to the coincidence of flowering (Table 10).

The wild species showed a great variability in flowering and duration during years (Fig. 8), indicating larger difference regarding chilling requirements. In 2003 it was observed a decrease in the period of flowering and large coincidence of flowering of the wild species with the cultivars Gala and Fuji. This could be related to the accumulation of chilling observed in this year (Tables 10, 11 and 12). 2006 was characterized by low chilling accumulation and frequent alternation between high and low temperatures during hibernal period. This condition determined the prolonged period of flowering in most evaluated species (with medium/high chilling requirement). Species with low chilling requirement anticipated flowering in relation to 'Gala' and 'Fuji'. According Petri et al. (1996) in warm winter conditions where chilling requirements are not fully satisfied to overcome dormancy it can occur variability in the date of flowering among years. Cultivars of low chilling requirement tend to flower earlier.

Among the species studied, *M. eleyi*, *M. floribunda*, *M. hopa*, *M. robusta*, 'Milalew Imuni' and 'Yellow Siberian' showed variation concerning flowering in the different years; and in some

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years, except *M. eleyi*, they showed low density of flowering indicating their low potential of use as pollinators (Fig. 8). The occurrence of abundant flowering is an indispensable condition for any cultivar or species that would be used as pollinator. Cultivars or species that show low flowering density or have tendency to alternate bearing should not be used as pollinators.



Fig. 8. Period of flowering of apple wild species and cultivars 'Gala' and 'Fuji' in seven years of phenological observation (2001-2007). Caçador, SC, Brazil, 2007. (Petri et al, 2008).

The occurrence of simultaneous flowering period and full bloom between pollinators and commercial cultivars increases the probability of having high fruitset index (Soltész, 2003). Any of the studied species showed perfect coincidence in the date of full bloom with the cultivars Gala and Fuji. Considering the species that flowered in all years 'John Downil', *M. platycarpa* and *M. baccata* had the date of full bloom closer to the observed for 'Gala' (Table 11). *M. baccata* and 'John Downil' differed 5 and 8.1 days (mean of years) in the date of full bloom in relation to cv. Fuji, respectively. However, these species showed great variability along the years. 'Gala' and 'Fuji' showed the lowest difference in the dates of full bloom and the lowest variability of this behavior along the evaluated years in comparison to the wild species. The percentage of coincidence of flowering of the wild species with 'Gala' and 'Fuji' varied among years and species (Table 11 and 12). The mean percentage of coincidence of the flowering

period of the wild species with 'Gala' and 'Fuji' was 39.7 and 39.9%, respectively. In some years, some wild species had 100% of the flowering coinciding with the 'Gala' and 'Fuji', but in other years they did not coincide. This behavior highlights the high variability of some apple wild species regarding flowering under warm winter conditions.

Creation / Crafting	Difference of full bloom date in relation to 'Gala' apples						
Species/Cultivar	Mean ±standart error	Variability index					
	Days*	%					
Malus eleyi	29.3±15.6 ab	53.2					
'Yellow Siberian'	7.8±9.8 d	125.4					
'Milalew Imuni'	10.3±14.4 bcd	139.7					
Malus robusta	1.3±1.2 d	86.6					
Malus floribunda	3.5±2.1 d	60.6					
Malus platycarpa	8.3±2.9 cd	35.3					
Malus baccata	8.9±7.0 bcd	79.1					
Malus hopa	11.9±6.7 bcd	56.7					
Malus atrosanguinea	36.4±22.1 a	60.8					
'Winter gold'	10.9±5.7 bcd	52.2					
'John Downil'	8.0±7.5 cd	93.3					
'Prof. Spengler'	22.0±18.2 abcd	82.8					
'Profusion'	29.0±17.4 abc	60.2					
Fuji	6.1±4.5 d	72.5					
Second / Cultimor	Difference of full bloom date in relation to 'Fuji' apples						
Specie/Cultivar	Mean ±standart error	Variability index					
	days	%					
Malus eleyi	24.3±14.9 ab	61.0					
'Yellow Siberian'	10.0±11.2 bc	112.0					
'Milalew Imuni'	14.0±19.9 abc	142.3					
Malus robusta	6.3±6.0 bc	95.2					
Malus floribunda	6.0±5.7 bc	94.3					
Malus platycarpa	11.0±5.7 bc	52.0					
Malus baccata	5.0±5.4 c	107.7					
Malus hopa	12.0±4.9 bc	40.5					
Malus atrosanguinea	31.1±18.4 a	59.2					
'Winter gold'	11.0±8.8 bc	79.6					
'John Downil'	8.1±9.4 bc	115.5					
'Prof. Spengler'	17.0±14.5 bc	85.1					
'Profusion'	23.7±14.2 ab	59.9					
Gala	6.1±4.5 bc	72.5					

\*Means followed by lowercase in the column do not differ by Tukey's test (p<0.05).

Table 10. Means and index of variability of the difference of full bloom date of apple wild species in relation to 'Gala' and 'Fuji' (*Malus domestica* Borkh.). Caçador/SC, 2007. (Petri et al, 2008).

Apple Phenology in Subtropical Climate Conditions

				Year				Mean	Variability
Specie/cultivar	2001	2002	2003	2004	2005	2006	2007	±standart error	index (%)
Percer	ntage o	of coinc	idence	of the	bloomi	ng peri	iod wit	h 'Gala' apples (S	%)
Malus eleyi	0.0*	0.0	56.3	0.0	82.4	38.5	0.0	25.3±34.0 ab	134.5
'Yellow Siberian'	0.0*	0.0*	56.3	0.0	100.0	84.6	44.4	40.8±42.2 ab	103.5
'Milalew Imuni'	0.0*	0.0*	0.0*	0.0*	41.2	20.5	55.6	16.7±23.2 b	138.7
Malus robusta	0.0*	10.0	0.0*	0.0*	100.0	25.6	77.8	30.5±41.4 ab	135.9
Malus floribunda	0.0*	0.0*	0.0*	0.0*	100.0	0.0*	55.6	22.2±40.1 ab	180.3
Malus platycarpa	78.6	20.0	68.8	0.0	100.0	76.9	77.8	60.3±36.1 ab	59.9
Malus baccata	0.0	20.0	50.0	0.0	11.8	43.6	100.0	32.2±35.8 ab	111.2
Malus hopa	0.0	100.0	87.5	6.7	100.0	82.1	100.0	68.0±44.8 a	65.8
Malus atrosanguinea	0.0	0.0	50.0	0.0	0.0	38.5	0.0	12.6±21.8 b	172.8
'Winter gold'	35.7	20.0	100.0	0.0	100.0	46.2	100.0	57.4±42.3 ab	73.7
'John Downil'	0.0	40.0	50.0	33.3	100.0	89.7	100.0	59.0±38.5 ab	65.2
'Prof. Spengler'	0.0	0.0	43.8	0.0	100.0	38.5	66.7	35.6±38.7 ab	108.9
'Profusion'	0.0	0.0	75.0	0.0	100.0	76.9	66.7	45.5±43.8 ab	96.1
Fuji	35.7	70.0	43.8	53.3	29.4	12.8	100.0	49.3±28.7 ab	58.3
Mean	10.7	20.0	48.7	6.7	76.1	48.2	67.5	39.7	107.5

\*no blooming; ns no significant; Means followed by lowercase in the column do not differ by Tukey's test (p<0.05)

Table 11. Coincidence of blooming period of wild species in relation to 'Gala' apples in seven years of phenological observations. Caçador, Santa Catarina, Brazil, 2007. (Petri et al., 2008).

Petri (2006) reported that in areas with inconstant climate during winter apples have showed irregularity in flowering. These species not often coincide the flowering period with the main commercial cultivars grown in Southern Brazil, showing anticipation in the period more than 30 days.

*M. atrosanguinea,* 'Milalew imuni', *M. floribunda, M. eleyi, M. robusta, M. baccata,* 'Prof. Spengler' and 'Yellow Siberian' exhibit coincidence of flowering inferior to 41% in the mean of the years for 'Gala' (Table 11). 'Profusion', Fuji, 'Winter gold', 'John Downil', *M. platycarpa* and *M. hopa* were the species that showed the greatest coincidence of flowering with 'Gala', highlighting 'Profusion' and *M. hopa* by the flowering regularity and intensity in the several years. Some species showed no coincidence at some years observed.

'Milalew imuni', *M. floribunda, M. atrosanguinea, M. robusta,* 'Yellow Siberian', *M. platycarpa* and *M. eleyi* showed the lowest performance regarding to the coincidence of flowering with the cultivar Fuji (Table 12). Although *M. eleyi* does not show great coincidence of flowering with 'Gala' and 'Fuji' it has the advantage of having great density of flowering for a long period. 'Winter gold', 'John Downil', *M. baccata,* 'Prof. Spengler', 'Profusion', Gala and *M. hopa* were the species that better coincided the flowering with 'Fuji'. From these, 'Prof. Spengler', 'Profusion' and *M. hopa* showed high flowering intensity. Williams (1977)

described the wild species as good producers of pollen, and were superior to commercial cultivars. He recommends two to four pollinators to make up for the difference of coincidence of flowering.

Crancia / aultionar				Year				Mean	Variability
Specie/cultivar	2001	2002	2003	2004	2005	2006	2007	±standart error	index (%)
Percen	tage of	coincic	lence c	of the b	loomin	g perio	od witl	n 'Fuji' apples (%	
Malus eleyi	0.0*	0.0	100.0	0.0	100.0	100.0	0.0	42.9±53.5 ns	124.7
'Yellow Siberian'	0.0*	0.0*	33.3	23.1	55.6	25.0	44.4	25.9±20.9	80.8
'Milalew Imuni'	0.0*	0.0*	0.0*	0.0*	55.6	0.0	55.6	15.9±27.1	170.8
Malus robusta	0.0*	0.0	0.0*	0.0*	77.8	8.3	77.8	23.4±37.3	159.2
Malus floribunda	0.0*	0.0*	0.0*	0.0*	100.0	0.0*	55.6	22.2±40.1	180.3
Malus platycarpa	64.7	0.0	16.7	0.0	88.9	0.0	77.8	$35.4 \pm 40.1$	113.0
Malus baccata	41.2	0.0	33.3	0.0	66.7	100.0	100.0	48.7±42.1	86.3
Malus hopa	52.9	88.9	41.7	46.2	100.0	8.3	100.0	62.6±34.7	55.5
Malus atrosanguinea	5.9	0.0	50.0	0.0	0.0	100.0	0.0	22.3±38.9	174.5
'Winter gold'	82.4	0.0	16.7	38.5	88.9	0.0	100.0	46.6±43.3	92.8
'John Downil'	47.1	11.1	33.3	76.9	55.6	0.0	100.0	46.3±35.3	76.2
'Prof. Spengler'	29.4	0.0	41.7	7.7	100.0	100.0	66.7	49.3±40.9	83.0
'Profusion'	41.2	0.0	75.0	7.7	100.0	100.0	66.7	$55.8 \pm 40.9$	73.3
Gala	29.4	77.8	58.3	61.5	55.6	41.7	100.0	60.6±23.1	38.2
Mean	28.2	12.7	35.7	18.7	74.6	41.7	67.5	39.9	107.7

\*no blooming; ns no significant; Means followed by lowercase in the column do not differ by Tukey's test (p<0.05)

Table 12. Coincidence of blooming period of wild species in relation to 'Fuji' apples in seven years of phenological observations. Caçador, Santa Catarina, Brazil, 2007. (Petri et al., 2008).

As exposed in the Table 13 the chilling accumulation during the hibernal period differed among years, having repercussion on the coincidence of flowering among cultivars (Table 11 and 12). Species that show lower variability along the years as for the coincidence of flowering with the cultivars Gala and Fuji, even when there was different chilling accumulation, show good ability as pollinators due their stable behavior at differentiated chilling regime during winter. The occurrence of low variability index in a determined pollinator may indicate that the pollinator responds similarly to the commercial cultivars under changeable chilling accumulation from one year to other. Considering the occurrence of winter with differenced chilling accumulation the use of pollinators that present similar behavior to their respective commercial cultivars is desirable in order to minimize the problems related to the non coincidence of pollination. Therefore, species showing the highest means of coincidence of flowering as well as the lowest index of variability are those that showed the best regularity regarding flowering period and are indicated to be used as pollinators whether considering the flowering period. It was observed that most cultivars showed index of variability superior to 100% which indicates that they have inconstant behavior along years and are greatly influenced by environmental conditions.

In general, it could be observed that no apple wild species had similar flowering phenological behavior to 'Gala' and 'Fuji'. Consequently, the use of a single specie as pollinator does not permit to embrace all the period of flowering of the commercial cultivars.

Soltész (2003) pointed out that it is necessary at least two cultivars to comprise the period of flowering of a particular cultivar of interest. Among those species 'Prof. Spengler', 'Profusion', 'Winter gold' and 'John Downil' are the apple wild species showing good potential of use because of the coincidence of flowering with 'Gala'. 'Profusion' and *M. hopa* were greater by the flowering regularity in the several years observed and flowering intensity in the year's average. Some species showed no coincidence at some years observed.

Although *M. baccata* and *M. platycarpa* have showed good coincidence of flowering with 'Gala' and 'Fuji' they provided larger fruits which required to be eliminated after flowering to avoid alternating flowering. This condition makes these species inadequate.

Voor	Chilling hours	Chilling units
Tear	below 7.2°C	(Modified North Carolina model)
2001	418	782
2002	269	549
2003	500	824
2004	400	1056
2005	309	571
2006	363	939
2007	535	918

Table 13. Accumulation of chilling hours below 7.2°C and chilling units according the model of Modified North Carolina from May to August in the years 2001 to 2007. Caçador. SC. Brazil. 2007.

#### 7. Effect of budbreak promoters on apple phenology

The occurrence of simultaneous flowering between cultivars of interest pollinators is an essential condition to assure adequate fruit production for the apple crop. Problems related to pollination and fecundation can reduce apple production and fruit quality due to the low fruitset and number of seed formed per fruit (Brault & Oliveira, 1995; Keulemans et al., 1996).

The flowering period of the cultivars is affected by environmental conditions (Soltész, 2003). Apple cultivars that normally flower in the same time in regions of good adaptation, when grown under warm winter conditions where the chilling exigencies are not totally satisfied trees might present great variability in the period of flowering causing pollination problems and consequently, affecting fruit production. Considering that most of the apple grower regions of the subtropical climate chilling are insufficient to deal with the chilling requirements of the main apple cultivars used, the adoption of management practices aiming coincidence of flowering between cultivars is of a great importance. According Erez (2000) the enhancement of the coincidence of flowering between cultivars can be reached with the use of budbreak promoters which anticipate flowering of the late flowering

cultivars. Budbreak promoter's substances can be used to reduce the chilling requirement of cultivars of low and medium exigencies and to modify the budbreak period, flowering and fruit maturation of temperate fruit species (George et al., 2002).



F (budbreak promoter x genotype) = 216,09\*\*

CV (%) = 4,96

\*\* F significant value at 1% of probability;

CaN = Calcium nitrate; MO = Mineral oil; HC = Hydrogen cyanamide; Means followed by same lowercase letter in the same cultivar do not differ by Tukey test (p<0.05), and means followed by same uppercase letter in the same budbreak promoter do not differ by Tukey test (p<0.05).

Fig. 9. Flowering period of 'Imperial Gala' and 'Fuji Suprema' apples treated with different budbreak promoters. Caçador, SC, Brazil, 2010. (Hawerroth et al., 2010).

At Fig. 9 it can be observed that the duration of apple flowering differs between cultivars and among budbreak promoters. Results obtained by Hawerroth et al. (2010) under subtropical conditions show that when no budbreak promoter is applied the cultivars Imperial Gala and Fuji Suprema tend to increase the duration of flowering period. According Petri & Leite (2004) the prolongation of the budbreak and flowering phases in apple is a typical symptom of insufficiency of hibernal chilling. The longer the flowering period the more difficult will be the carrying out of some cultural practices such as fruit thinning and diseases control due to the occurrence of different phenological stages in the same tree. Under warm winter conditions, where the chilling exigencies are not completely satisfied, cultivars with distinct chilling requirements showed immense variability in the flowering period from year to year (Petri et al. 2008). The lack of synchrony of flowering between cultivars is clearly observed in the control treatment (Fig. 9) where the full bloom of 'Imperial Gala' coincides with the end of flowering of 'Fuji Suprema'.

The use of substances to induce budbreak can improve the synchrony of flowering between cultivars, evidenced by the greater coincidence in the dates of full bloom between cultivars treated with budbreak promoters (Fig. 10). The occurrence of simultaneous flowering of commercial cultivars and pollinators enhances the possibility to occur high indexes of fruitset (Soltész, 2003).

In relation to flowering period the application of budbreak promoters can anticipate the apple flowering, reducing the time between the application of the treatments and full bloom. The application of budbreak promoters such as mineral oil and hydrogen cyanamid were efficient in the anticipation of flowering of 'Imperial Gala' and 'Fuji Suprema', as observed by Hawerroth et al. (2009) (Table 14). The differenced behavior among the level of the factor budbreak promoters regarding flowering time proves the importance of the use of budbreak promoters as strategies of management to maximize pollination. This is due to the greater synchronization of flowering between cultivars of commercial importance and their pollinators.

Treatmet	Interval between application of treatments and beginning of flowering	
	Imperial Gala	Fuji Suprema
	Days	
Control	30.0Aa	26.0Ba
MO 3.2%	28.0Ab	25.0Bb
MO 3.2% + HC 0.20%	25.0Ac	23.0Bc
MO 3.2% + HC 0.39%	25.0Ac	23.0Bc
MO 3.2% + HC 0.59%	25.0Ac	23.0Bc
Mean	26.6	24.0
CV (%) = 1.12	F (budbreak promoters x genotype) = 29.90**	

\*\* F value significant at 1% of probability. Means followed by uppercase letter in the line, and means followed by lowercase in the column do not differ by Tukey's test (p<0.05)

Table 14. Interval between application of treatments and beginning of bloom in 'Imperial Gala' and 'Fuji Suprema' cultivars treated with different concentrations of hydrogen cyanamide (CH) and mineral oil (MO). Caçador, SC, Brazil, 2008. (Hawerroth et al., 2009).

#### 8. Conclusions

Under warm climate, such as the Brazilian conditions, the apple flower induction and differentiation tend to extend along growth cycle as there is vegetative growth. That means that in the meantime as buds flowers are formed the process of induction is still occurring. This particular situation might lead to an alteration in the tree physiology making necessary a differentiated management on tree training. So, cultural and climatic conditions alters positively or negatively the flower development to the next year.

In general, in fruit tree orchards, when trees are grown under unstable climate regions where cultural practices are altered, the timing of flowering and the phenology could have important impacts on fruit production because of possible indirect influences of phenology on pollination and fruit-set efficiency, and consequentially on bud differentiation.

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#### Phenology and Climate Change

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Phenology, a study of animal and plant life cycle, is one of the most obvious and direct phenomena on our planet. The timing of phenological events provides vital information for climate change investigation, natural resource management, carbon sequence analysis, and crop and forest growth monitoring. This book summarizes recent progresses in the understanding of seasonal variation in animals and plants and its correlations to climate variables. With the contributions of phenological scientists worldwide, this book is subdivided into sixteen chapters and sorted in four parts: animal life cycle, plant seasonality, phenology in fruit plants, and remote sensing phenology. The chapters of this book offer a broad overview of phenology observations and climate impacts. Hopefully this book will stimulate further developments in relation to phenology monitoring, modeling and predicting.

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