First Attempts of Linking Modelling, Postharvest Behaviour and Melon Genetics

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Abstract

The onset of climacteric is associated with the end of melon fruit shelf-life. The aim of this research was to develop practical and applicable models of fruit ripening changes (hardness, moisture loss) also able to discriminate between climacteric and non-climacteric behaviour. The decrease in firmness was measured non-destructively by flat-plate compression; moisture loss was measured by weight loss. A set of 13-15 near-isogenic lines (NILs) derived from the climacteric line SC3-5 was used to verify the relationship among the climacteric behaviour and ripening related changes (weight loss, softening and color) during two consecutive seasons. The biological variance models for moisture loss and firmness followed a simple exponential behaviour that explained more than 90% of the total variance. Results of the analyses using these models could not be linked to properties of near-isogenic lines like climacteric behaviour, ethylene production or skin thickness. The results suggest that the phenotype is more important than genotype, when considering mean values. These results seem to suggest that relations may exist between the different processes and properties of NILs on an individual basis, not on mean values.

INTRODUCTION

Eduardo et al. (2005) developed a collection of near-isogenic lines (NILs) (Eduardo et al. 2005). The parental lines used to generate the NILs produced melons with non-climacteric behaviour. In contrast, SC3-5 and other NILs with shorter introgression in LG III showed climacteric behaviour during melon ripening (Moreno et al., in press). In this paper this variability is used to model the behaviour of melon firmness and weight loss during postharvest ripening, to understand the variation in firmness behaviour and to gain insights for the evaluation of genotypes with regard to their postharvest behaviour.

MATERIAL AND METHODS

Near-isogenic lines (NILs) of melons (*Cucumis melo L.*) were produced in two seasons (2005 and 2006) containing introgressions of different extent from the Korean accession ‘Shongwan Charmi’ PI161375 (SC) on the linkage group III into the ‘Piel de Sapo’ (PS) (Eduardo et al., 2005). Some lines (Eduardo et al., 2005) contained introgressions of SC into PS in linkage groups X and VII, respectively. A large range of weight, colour, shape and firmness was present in the NILs in both seasons. Lines were coded as 5Mx or 6Mx in 2005 and 2006 seasons, but the x codes were only common in some NILs (Table 2).

5-6 fruit per line chosen at random from the replicates (about 20 plants), were stored covered by plastic liners at 21 °C at a relative humidity of 66 % (2005) or 73 %

(2006). Fruit firmness was measured as the maximum force in a flat plate compression at the equator of the fruits (indicated spot) until a predefined compression distance of 2 mm (2.2 mm in 2006). Water loss was determined by weighing the same fruit during storage.

Statistical analysis of the data was conducted using non linear mixed effects regression using the procedure nlme of the free package R (www.R-project.org).

MODELLING

Firmness

Often the pattern of firmness loss is exponential towards an asymptotic end value (Tijskens et al. 2004, 2006, Schouten et al. 2007, Ergun et al. 2005, Lana et al. 2005). For melon fruit firmness the behaviour seems also to be the case. The behaviour can therefore be represented mathematically as Eq.1 (first order kinetics with an invariable part).

$$F = (F_0 - F_{min}) \cdot e^{-k_f \cdot t} + F_{min} \qquad \text{Eq. 1}$$

where F represents the textural property firmness (in $N \cdot mm^{-1}$), k_f the rate constant of softening (in day^{-1}), t the time (in days). Index *0* refers to initial (at harvest) and *min* to the asymptotic value (at infinite time). The asymptotic value F_{min} expresses the firmness generated from the structural aspects of the tissues involved. It is assumed that within the normal time frame of storage, these structural issues do not change upon ripening. The different individuals and the NILs showed a large variation in this end value and in the initial firmness F_0. These parameters have to be estimated per individual and will contain all the information on the biological variance of a specific NIL. The rate constant k_f is supposed to be connected to the chemical process of softening, and will be the same for all lines and individuals.

Once the individual values of the end value F_{min} are estimated, Eq. 1 can be converted into a more appropriate fashion for a standardised firmness:

$$\begin{aligned} F_{stan} &= F - F_{min} = (F_0 - F_{min}) \cdot e^{-k_f \cdot t} \\ F_{stan} &= F_{ref} \cdot e^{-k_f \cdot (t + \Delta t)} \end{aligned} \qquad \text{Eq. 2}$$

where index *ref* stands for an arbitrary chosen value (here 20 N), *stan* stands for the standardised value, and Δt is the biological shift factor that expresses the difference in development stage of the individual fruits and can be calculated as shown in Eq. 3.

$$\Delta t = -\frac{\log\left(\dfrac{F_0 - F_{min}}{F_{ref}}\right)}{k_f} \qquad \text{Eq. 3}$$

In Eq. 2, $t{+}\Delta t$ represents the biological time (in days) relative to the point of reaching a firmness equal to F_{ref} (Tijskens et al., 2005). It is a stochastic variable that contains all the information of the biological variance for a batch of melons.

Weight Loss

Weight loss, primarily caused by water loss, is usually modelled based on Fick's first law (De Smet et al. 2002). Applying the central difference technique to approximate the second derivative, one arrives, at constant external conditions, at an exponential behaviour, similar as for softening (Eq. 1). The same line of reasoning is followed and the same models were applied as for firmness decay. The asymptotic end value W_{min} was found to be linearly related to the initial weight W_0. Expressing the data as weight loss (1-W/W_0) and replacing W_{min} with its linear relation ($W_{min}=\alpha \cdot W_0+\beta$) result it the following expression:

$$WL = (\alpha - 1 + \frac{\beta}{W_0}) \cdot (-1 + e^{-k_w \cdot t}) \qquad \text{Eq. 4}$$

Here W_0 is the (measured) initial weight of the fruit, strongly related to its size, while β represents the external conditions (Vapour Pressure Deficit based on RH and temperature) and α the fraction of fruit not available for evaporation (dry matter, bound water, cytoplasm).

RESULTS

In 2005, lines showed climacteric behaviour except PS, 5M2 and 5M7. In 2006, the NILs showed non-climacteric with four exceptions (5M1, 5M4, 5M6, 5M10; data not shown).

In Table 1, the results for the analysis of firmness using Eq. 1 are shown. The parameters estimated for the two seasons are quite similar, except for the variation over the individuals (σ) for F_0 and F_{min}. F_0 is strongly related to the maturity at harvest, and therefore prone to variation induced by the actual moment of harvest. F_{min} on the other hand should be related to the different properties of the NILs. In Table 2, the mean and standard deviation of these asymptotic values are shown per NIL. A huge variation exists, not only between (mean) but also within the NILs (σ). The first one is expected as it reflects the differences in firmness properties between the lines. The second one, however, is surprising. It would indicate that the phenotype is at least equally important as the genotype.

In Fig. 1, two examples are shown for the firmness behaviour measured (symbols) and simulated (lines) for two NILs. In Fig. 2, the standardised firmness (Eq. 2) is shown for all fruit of all lines for both seasons. The approach used seems to be valid, indicating that the rate constant of softening is the same for all fruit of all lines, irrespective of their differences in genotype: the parental lines used to generate this NIL collection produced melons with non-climacteric behaviour (Moreno et al., in press). In contrast, SC3-5 and NILs with shorter introgression in LG III showed climacteric behaviour during melon ripening (Obando-Ulloa et al., 2008). Whether the (individual) end value F_{min} is related to the genetic pattern (line) or the level of being climacteric (of individual fruit) is still under investigation.

For weight loss similar results are obtained. The data of both seasons were analysed together, using Eq. 4 estimating α and β in common while estimating a separate rate constant (k_w) for each individual. The explained part is high and the standard error of estimates (s.e.e) are low (see Table 3), except for the parameter β. Since β reflects the external conditions (the same for each fruit), and only one condition is applied in these data, nothing much can be said about its reliability. Although the variation over the estimated rate constant for individual fruit seems not too large (see Table 3, σ_{kw}), it is too large to neglect. The analysis revealed no relation whatsoever with the genetic lines. No relation was found with the thickness of the skin (k_w should reflect the skin permeance), measured on other fruit. So, again, the phenotype seems to be more important than the genotype in determining the properties of NILs.

In Fig. 3 some examples are shown for the general behaviour of weight loss as a function of time for different fruit in the same NIL. The asymptotic end value depends on the initial weight (or size) of the fruit.

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Tables

Table 1. Results of the integral non-linear mixed effect regression analysis on firmness using Eq. 1. Values for F_0 and F_{min} represent the mean of the whole melon fruit firmness values estimated. σ represents the standard deviation in F_0 and F_{min} over all the individual fruit in the data set. s.e. is the statistical standard error of estimates indicating the reliability of the parameter estimate.

		2005			2006		
Name	Units	Value	s.e.	σ	Value	s.e.	σ
F_{min}	N/mm	16.89	3.502	10.39	23.75	1.848	5.37
F_0	N/mm	46.29	3.557	8.58	44.00	1.947	6.15
k_f	1/day	0.1005	0.0082		0.1121	0.0073	
N_{obs}	-	533			829		
N_{groups}	-	63			76		
R^2_{adj}	-	0.972			0.905		

Table 2. Mean and standard variation of the asymptotic end value F_{min} of individual fruit per NIL (n= 5 to 6).

F_{min} of individual fruit (N/mm)					
Season 2005			Season 2006		
NIL	mean	σ	NIL	mean	σ
5M1 (=SC3-5-1)	12.78	14.10	6M1 (= 5M1)	15.81	6.91
5M2	18.80	12.41	6M2	27.67	3.07
5M3	1.52	4.44	6M3	15.88	3.93
5M4	1.25	9.04	6M4	21.70	5.27
5M5	15.32	6.48	6M5	33.54	4.53
5M6	21.36	10.71	6M6 (= 5M6)	20.94	8.12
5M7	30.50	7.76	6M7	20.99	6.10
5M8	5.27	11.97	6M8	22.21	7.76
5M9	15.91	7.25	6M9	20.72	4.21
5M10	22.11	19.43	SC3-5	17.54	7.52
PS	41.22	10.49	PS	31.78	3.72
			SC7-1	36.17	5.64
			SC10-2	24.24	4.71

Table 3. Results of nonlinear regression analysis for weight loss (Eq. 4).

	Value	S.e.e.
α	0.923	0.0018
β	8.326	2.3635
k_w	0.0464	0.00120
σ_{kw}	0.0118	
N_{obs}	1450	
N_{groups}	135	
R^2_{adj}	0.992	

Figures

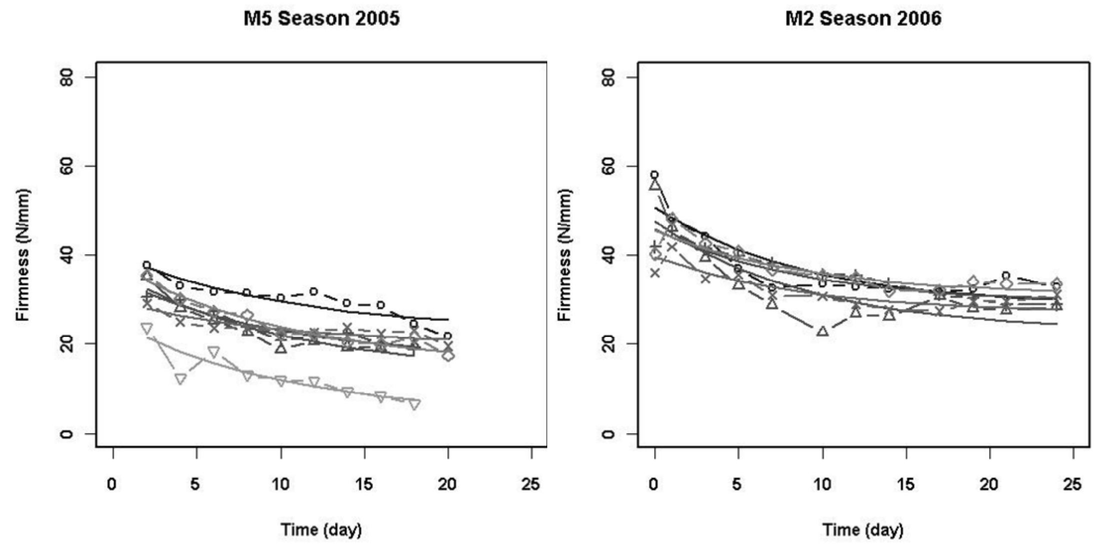

Fig. 1. Behaviour of firmness of two NILs (M5 in 2005 was a climacteric line while M2 in 2006 showed non-climacteric behaviour), Symbols=measured, lines= simulated. Notice the large differences in asymptotic end value within and between them.

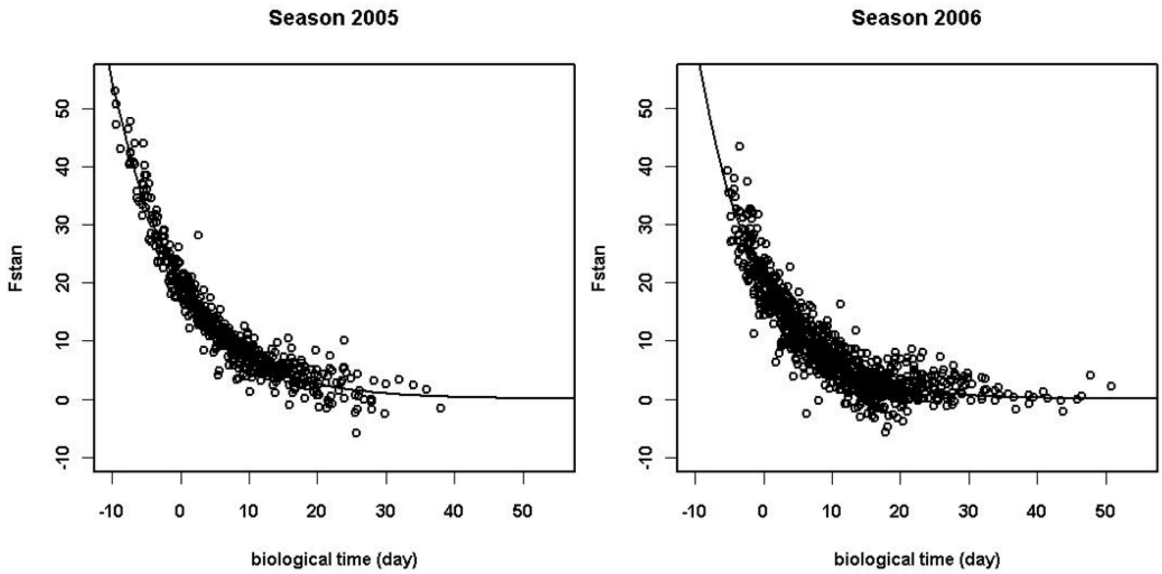

Fig. 2. Measured and simulated standardised melon whole fruit firmness (Eq.2) (all fruit of all lines) versus biological time ($t+\Delta t$).

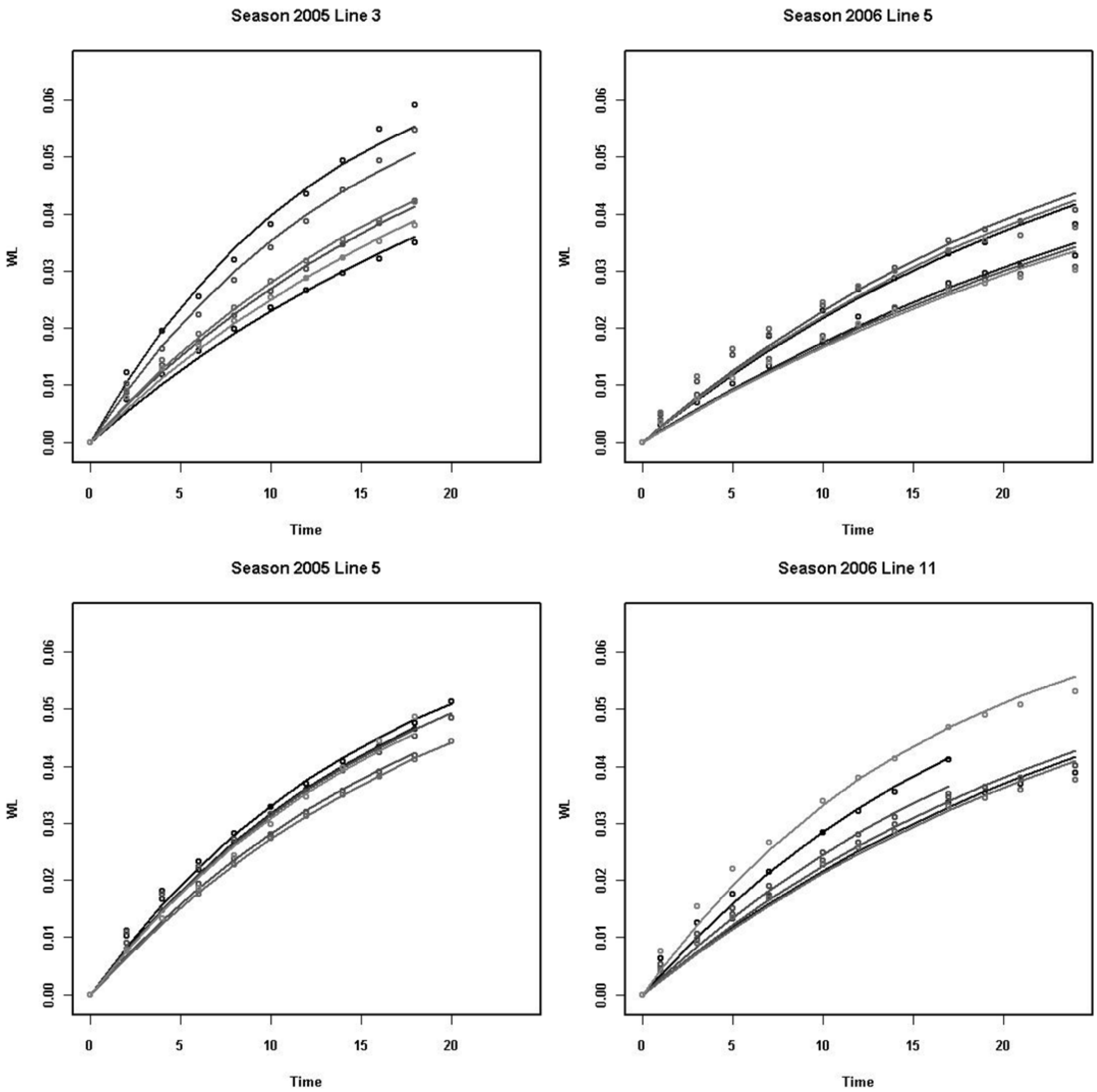

Fig. 3. Example for weight loss (*WL* in g) as a function of storage time (days) for both seasons combined. Line 11 was the parental line 'Piel de sapo' (PS) or control. The lines presented of 2005 showed climacteric behaviour and the lines of 2006 were non-climacteric.