The Biological Shift Factor: Biological Age as a Tool for Modelling in Pre- and Postharvest Horticulture

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Abstract
Individuals differ in development stage or biological age. This difference can be taken into account when modelling the quality behaviour of various fruits and vegetables. Even on a batch level, the same principle can be applied, provided the variation within a batch is not too large. By applying the biological shift factor, i.e. a shift in calendar time, the effects of different growing and harvesting condition can be included in modelling quality behaviour, which widely opens alleys for producing models applicable in the entire globalised food chain. The variation in biological shift factor over individuals in a batch and over several batches seems to exhibit a normal distribution pattern.

INTRODUCTION
The concept of biological age has been subject of many discussions and developments over the past decades. Especially in the pre-harvest period, this concept was developed as a normalising approach for products grown under different conditions and scenarios of temperature, light, fertilisation and other management issues (e.g. Heuvelink and Marcelis, 1989). Application of these viewpoints in research and practice was however always hindered not only by the lack of a suitable definition, but especially because biological age could not be measured or determined in a quantitative way.

The advantage of thinking in biological age is that it does not matter how long it takes in calendar time to reach a certain stage. Once the produce reaches that stage, it will be identical (at least highly comparable) to all other individuals at that stage, no matter how that stage is reached. For purposes of modelling and understanding product behaviour, this is a very powerful and useful concept. Temperature (during growth or storage), light, fertilisation etc. do no longer reflect on the product behaviour or state, only on the calendar time necessary to reach that state. This concept also allows to link pre- and postharvest phases, which is essential for understanding the sometimes erratic behaviour during postharvest stage and handling (Heuvelink et al., 2004). In Fig. 2 an example is given, based on a logistic behaviour as is frequently encountered in colour development.

The concept of biological age as a substitute for calendar time closely resembles the system of dimensionless representations of behaviour, frequently encountered in the field of engineering and physics. In this paper, some examples are provided based on measured data of several quality attributes for several products, and based on the modelling and analysis of these data. In this paper, the biological shift factor is introduced that can serve as a quantitative estimate for the biological age of individuals or batches of individuals. At the same time the biological shift factor of individuals in a batch do express the variation that exists in that batch.

DEFINITION OF BIOLOGICAL SHIFT FACTOR

Constant Conditions
Assuming that the physiological development of a product follows the same behaviour at constant environmental conditions (e.g. temperature), then differences between individuals of a same species will be due to differences in biological age. This difference can be expressed as a biological shift factor. That is the time that the development behaviour has to be shifted to obtain the same generic pattern as shown in
Fig. 2. The example is given for a case of logistic sigmoidal behaviour but the same line of reasoning is valid when others patterns are considered. So, the biological shift factor, represented by the arrows in Fig. 2., is the only parameter necessary to define the differences between the different individuals in a batch. The set of biological shift factors for all the individuals in a batch contains therefore all the information on the biological variance in that batch.

**Various Constant Conditions**

When the conditions like temperature are constant but different in different measuring series, matters do become more complicated. Still the physiological properties of a product develop along the same lines no matter how that development stage was reached, e.g. due to different conditions of temperature, fertiliser etc. One possibility to overcome the complexity by the various constant temperatures is to express time in a standardised manner. Standardised means here that all effects of these different conditions (again temperature, fertilisers etc.) are taken into account. This procedure closely resembles the techniques of dimensionless expression frequently used in the area of physics and engineering. A graphical representation of the behaviour of a simple exponential decay as shown in eq. 1, becomes quite cumbersome (see Fig. 3, top) when the rate constant k changes over a wide range by effects of temperature. Rate constants depend on temperature according to the Arrhenius relation (eq. 2).

\[
y = y_0 \cdot e^{-k \cdot t} \quad \text{eq. 1}
\]

\[
k = k_{ref} \cdot e^{\frac{Ea}{T-T_{ref}}} \quad \text{eq 2}
\]

Expressing the behaviour against \(k \cdot t\) (dimensionless) or \(k \cdot t / k_{stand}\) (dimension time) (see Fig. 3, middle) as standardised time reduces the complex graph to a set of lines, all with the same (standardised) rate constant. Combining this technique with the time shift system produces a single line (see Fig. 3, bottom) even when the initial conditions \(y_0\) are different for different series. The mathematical expression including the time shift technique is shown in the eq. 3:

\[
y = y_{0,stand} \cdot e^{-k \cdot (t+\Delta t)} \quad \text{eq. 3}
\]

Where \(y_{0,stand}\) represents a freely chosen initial condition and \(\Delta t\) the time shift factor for each series.

**EXAMPLES FROM PRACTICE**

Recently some interesting developments in modelling and data analysis have been reported that rely on this biological age to pool the information over large numbers of individuals, circumstances and stages in maturity at harvest (Lana et al. 2004a,b, Schouten et al. 2003, 2004a). Provided a good and reliable model is available to describe a particular property or attribute, data from different studies and different experimental designs can be pooled to be analysed together, expressing the time relative to some arbitrary point, e.g. moment of anthesis. The applied technique connects the mechanism active in changing the property under study very closely to the biological variance, present in all samples of living material (Schouten 2004b, Tijskens et al. 2003, 2004).

**Tomatoes at Different Stages of Maturity at Harvest**

In a recent report, a model for exponential firmness decay in sliced tomatoes (Lana et al. 2004a) was represented as:

\[
F = (F_0 - F_{fix}) \cdot e^{-k_{g,s} \cdot (t+\Delta t)} + F_{fix} \quad \text{eq. 4}
\]

where \(t\) is the time, \(F\) is the firmness, \(k\) the rate constants, subscripts 0 for initial at some arbitrary point, fix indicates the asymptotic value, \(g\) for growth and \(s\) for storage. Using
statistical mixed effects non-linear regression analysis on mean values for firmness for each maturity at harvest, all values of the model parameters could be estimated in common for all individuals. The term $\Delta t$, which is actually the time shift factor for batches, could be estimated separately for each individual batch of tomatoes (Table 1), relative to the time of the first stage (green). The value of $\Delta t$ is the time that each individual has to be shifted over the experimental time to fall over the same curve. It represents the biological age, relative to some arbitrary point. Through the incorporation of the time shift factor, successive ripening stages could be analysed together as belonging to the same curve (Fig. 1).

The same approach, with very similar results, could be followed in an analysis of the colour of the same tomatoes (Lana et al, 2004b).

**Colour of Bell Peppers during Growth**

In an effort to predict the ripening of bell peppers during growth to optimise the allocation of harvest labour, the colour development could be described by a simple first order exponential behaviour (see eq. 3) based on about 130 individual fruits, for both a red and a yellow cultivar (Tijskens et al., 2004). The estimated biological shift factors for all these individual fruits, relative to the recorded harvest time, did exhibit an almost normal distribution (Fig. 5, Table ).

**Colour of Apples**

The development of the colour of Granny Smith apples was measured for more than 500 individual fruits harvested at three orchards, at two stages of maturity, and stored at three different temperatures. The colour development for all these individuals could be analysed (mixed effects non-linear regression) based on the same simple but now increasing exponential curve (eq. 3), which can be regarded as the approximation of the first part of the logistic curve (see Fig. 2, Right). Again the distribution of all the estimated values for the biological shift factor (see Fig. 5) can be used to characterise the different batches. An overview of the statistical analysis for all three orchards and ripeness stage at harvest is shown in Table . Converting the individual time shift factors into a shift in the colour attribute, using exactly the same model expression, information can also be extracted on the dynamics of the changes in these distributions.

**Keeping Quality of Cucumbers**

Schouten et al (2003, 2004a) investigated the colour development of cucumbers and applied a colour model, based on information from literature (Fig. 8, Fig. 6) to estimate precursor (protochlorophyll) concentrations at harvest. The same precursor concentration for two cucumbers ensures the same postharvest colour development. So, to differentiate between cucumbers or cucumber batches no biological shift factors were determined, but precursor concentrations at harvest (Fig. 7). Precursor concentrations and biological shift factors are, however, not that different as both are maturity indicators. The precursor concentration at harvest is independent of the postharvest storage temperature. Also, a shift in precursor concentration can be envisioned for each cucumber as to generate a generic colour pattern. This is analogous to the biological shift factor. Although biological shift concept is clear and applicable, the use of precursor concentrations may have advantages. For instance, the protochlorophyll distributions are moving between two borders and show a different shape as function of the position between those borders (Fig. 7). The upper border indicates a protochlorophyll concentration that is specific for each cucumber cultivar. In general, precursor concentrations may be easier to handle than biological shift factors when attempting to connect the influence of preharvest factors on postharvest behaviour.

**CONCLUSIONS**

Based on a number of practical and theoretical examples, the technique of estimating the biological shift factor in pooled data sets is demonstrated and the power of
the biological age system is elucidated. The variation between individuals, expressed as the biological shift factor, does seem to be normally distributed. An additional advantage of the procedure is that the biological variance in batches of horticultural produce is made explicit. This biological variance is concealed in the distribution, (mean and standard deviation) of all these time shift values for the individuals in a batch.

This technique opens a wide range of applications, both practical and scientific for more dedicated approaches to improve the quality and managerial possibilities in the horticultural product chain.

Literature Cited

Tables

Table 1. Results on the time shift factor for tomatoes harvested at different stages of maturity, expressed in day at 20 °C.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value (days)</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\Delta t_{\text{green}}$</td>
<td>0</td>
</tr>
<tr>
<td>$\Delta t_{\text{pink}}$</td>
<td>2.31</td>
</tr>
<tr>
<td>$\Delta t_{\text{red}}$</td>
<td>6.21</td>
</tr>
<tr>
<td>$R^2_{\text{adj}}$</td>
<td>91.9</td>
</tr>
<tr>
<td>Nobs</td>
<td>85</td>
</tr>
</tbody>
</table>
Table 2. Statistical result for the biological shift factor $\Delta t$ (season 2002) for bell peppers.

<table>
<thead>
<tr>
<th></th>
<th>Red</th>
<th>Yellow</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean</td>
<td>-5.39969</td>
<td>-1.85395</td>
</tr>
<tr>
<td>StdDev</td>
<td>2.082247</td>
<td>2.569593</td>
</tr>
</tbody>
</table>

Table 3. Overview of the mixed effects non-linear regression analysis for the colour of Granny Smith apples from three orchards (AS, BL, KK) at two stages of maturity at harvest.

<table>
<thead>
<tr>
<th>colmin</th>
<th>k</th>
<th>$\Delta t$</th>
<th>stdev $\Delta t$</th>
</tr>
</thead>
<tbody>
<tr>
<td>AS1</td>
<td>-20.75</td>
<td>0.01637</td>
<td>3.72</td>
</tr>
<tr>
<td>AS2</td>
<td>-20.66</td>
<td>0.01739</td>
<td>4.11</td>
</tr>
<tr>
<td>BL1</td>
<td>-21.27</td>
<td>0.01862</td>
<td>3.63</td>
</tr>
<tr>
<td>BL2</td>
<td>-21.33</td>
<td>0.02055</td>
<td>3.69</td>
</tr>
<tr>
<td>KK1</td>
<td>-22.91</td>
<td>0.01433</td>
<td>2.45</td>
</tr>
<tr>
<td>KK2</td>
<td>-22.67</td>
<td>0.01503</td>
<td>2.72</td>
</tr>
</tbody>
</table>

Figures

Fig. 1. Left: Development of an arbitrary attribute normalised between 0 and 1 for a sigmoidal process in calendar time, revealing the differences between individuals, as shown by the arrows indicating the biological shift factor for each individual. Right: Same representation in biological time, avoiding the differences between individuals, but stressing the underlying generic behaviour (biological time = calendar time + biological shift factor).
Fig. 2. Example for the system of standardised time, combined with the biological shift factor.

Fig. 3. Example of tomato pericarp firmness at three different stage of development at harvest (green, pink and red), stored at different temperatures (standardised).
Fig. 4. Distribution of the biological shift factor ($\Delta t$) in bell peppers ripening at the vine in a greenhouse (top red cultivar, bottom yellow cultivar).

Fig. 5. Distribution of biological shift factor ($\Delta t$) for individual apples from orchard Krško (Slovenia) harvested at commercial ripeness (top) and one week earlier (bottom).
Fig. 6. Colour behaviour of cucumbers (expressed as RGB B/R) for different stages of maturity. Indicated is the acceptance colour limit.

\[ \text{Pchl} \leftrightarrow \text{chl} \leftrightarrow \text{CHL} \]
\[ \text{colourless} \]

Fig. 7. Extended mechanism of colour change in cucumbers.

Fig. 8. Example of two Pchl distributions at harvest that are indicative of different batch maturities. The dashed line indicates the cultivar dependant maximal precursor value.