# Benefits of Fundamental Modelling - the Case of Physiological Disorders

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**Abstract**

**A model was developed assuming that the occurrence and the development of physiological disorders are effects of a balance between processes of disorder formation and scavenging of initiating compounds. Based on this (simplified) mechanism and applying the fundamental rules of chemical kinetics, the differential equations can be derived. The observed effects depend on initial levels of some nutrients (N), free radicals (Ra) and scavenging activity (SS). All three types of compounds do depend on the growing conditions prior to harvest, and hence on season, weather and climate. Postharvest development of disorders (PD) depends also on conditions during storage, e.g. temperature and $CO_2$ level. Thus their influence was incorporated into a model based on fundamental knowledge in that sphere.**

**The benefit of the developed model is that it allows simulating very different ranges in type of development of disorder. From very fast and reaching high level of occurrence, very fast but stabilised at low level of occurrence, to slow but reaching high level of occurrence, and slow and stabilised at low level of occurrence. And of course simulating of cases where the disorder occurrence is not observed at all.**

**The model was checked against measured data on pears, indicating that the approach seems to be realistic and powerful enough to pursue. Existing data are however gathered in a manner not suitable for this approach. Therefore further studies on dynamics of physiological disorders require improvement of experimental methods.**

## INTRODUCTION

Traditionally, models in agriculture do not take the variance between batches or individuals into account. These models are useful for numerous practical applications, yet sometimes they lead to significant errors in predicted values.

Fundamental modelling is based on two major pillars: the basic processes responsible for changes of observed attributes and theoretical information on these processes and on biological variance within simulated batch of fruits. Fundamental models of this type describe the changes of attributes within individuals depending on the actual state these individuals have at the moment the experiments start. With this approach various observed cases can be explained and described with the same model. Incorporating the effects of variance between individuals in fundamental process oriented models could possibly decrease the number of erroneous simulation.

## FORMULATION OF THE MODEL

The observations of disorder occurrence showed that if disorders are found in a batch of fruits, only part of that batch is affected. The remainder of the fruits is not affected at all. This observed pattern is independent of fruit size, position in the canopy and many other investigated factors. Even in the batches of large fruits picked exclusively from upper part of canopy (which is a factor increasing the probability of disorder occurrence) one can find fruits without disorder. This leads to conclusion that there is probably some kind of "disorder seed" present in some fruits and absent in the others.

This "seed" is a chemical element, compound or a concentration of specific atoms or molecules, come into being in favourable conditions and creating sensitivity to formation of a disorder.

The general knowledge of chemistry and physiology (Tomala, 1999; Kruger et al., 1999; Lourens and Malherbe, 1999; Larrigaudiere et al., 2001) suggests that some physiological disorders are caused by free radicals. The analogy with development of chilling injury caused by free radicals (Tijskens et al., 1994) is evident. It is also widely accepted that occurrence of physiological disorders in fruits depends on their growing conditions (Tomala, 1999; Toivonen et al., 2003)

Binding this information together one can say that the physiological disorder (PD) affects the tissue of substrate (S) when free radicals (Ra) are present. And that this process depends also on other factors exemplified by richness in nutrients (N), which (according to experts' observations) enhance the occurrence of disorder. This process of disorder formation can be represented as:

$$S + N + Ra \xrightarrow{k_f} PD + 2\ Ra \qquad \text{eq. 1}$$

Veltman et al. (2003) and Larrigaudière et al. (2004) suggest that lack of antioxidants is responsible for occurrence of some physiological disorders in pears, while Franck et al. (2003) reported coincidence of sound tissue spots occurrence in the core breakdown zone and the higher ascorbic acid concentration. Therefore one can say that free radicals (Ra) are the subject of some scavenging system (SS), which in effect inhibits the process of disorder formation:

$$Ra + SS \xrightarrow{k_s} SS \qquad \text{eq. 2}$$

The numerous experiments concerning the storage of fruit in different conditions show that storage conditions also have a significant impact on the development of disorder. For example, the storage of pears with different levels of $CO_2$ showed that an increase of $CO_2$ in the storage atmosphere could cause a significant increase in number of fruits with disorder observed. Considering these observations, the explanation that the presence of $CO_2$ (or other gases in storage atmosphere) directly affects the formation of disorder has to be excluded, because when there is no $CO_2$ in storage chamber the disorder still can be observed. That means that the presence of $CO_2$ probably influences the scavenging system. This theoretical conclusion is consistent with observation that ascorbic acid concentration in many fruits decreases in high $CO_2$ conditions (Agar et al., 1997). Also Veltman et al. (2003) have found that adding $CO_2$ to the storage atmosphere not only decreased ascorbic acid levels but also increased the severity of brown core. So, the additional process, which causes the increased development of disorder, can be actually described as:

$$SS + CO_2 \xrightarrow{k_{CO2}} CO_2 \qquad \text{eq. 3}$$

$k_f$, $k_s$ and $k_{CO2}$ are the rate constants of above processes and depend on temperature according to Arrhenius equation:

$$k_i = k_{iref}\ e^{\frac{E_i}{R}\left(\frac{1}{T_{ref}} - \frac{1}{T}\right)} \qquad \text{eq. 4}$$

where $k_i$ is rate constant of each of processes described above, $T_{ref}$ (K) is the reference temperature (chosen to be e.g. 293.15 K, i.e. 20 ºC), and R is universal gas constant (8.3143 J/K/mol).

The above mechanism can be converted into set of differential equations, using the rules of chemical kinetics:

$$\frac{\partial\ PD}{\partial\ t} = k_f\ S\ N\ Ra \qquad \text{eq. 5}$$

$$\frac{\partial S}{\partial t} = -k_f S N Ra \qquad \text{eq. 6}$$

$$\frac{\partial Ra}{\partial t} = k_f S N Ra - k_S SS Ra \qquad \text{eq. 7}$$

$$\frac{\partial N}{\partial t} = -k_f S N Ra \qquad \text{eq. 8}$$

$$\frac{\partial SS}{\partial t} = - k_{CO2} CO_2 SS \qquad \text{eq. 9}$$

**MODEL PERFORMANCE**

Assuming for the time being, a constant level of nutrients (N(t)=N0), and of scavenging activity (SS(t)=SS0), the simulation of disorder, substrate and free radicals might be as shown at Fig. 1.

The "Ra(t)" line at Fig. 1, which represents the simulation of radicals level changes, is the key for understanding of the development of disorder. At the harvest time (t=0), each individual fruit has some specific level of free radicals, built up during the growth of the product. It could be called "the seed of disorder". According to eq. 7 the radicals are the subject of two processes: formation and scavenging. However, at the beginning, the formation process is much more efficient due to high initial level of available substrate (S). Later on the substrate level decreases so much that the formation of new radicals becomes slower than the process of scavenging, and the level of free radicals begins to decrease. This is also the moment when formation of disorder slows down.

This model follows the mass preservation law that substrate turns totally into disorder: S(t)=S(0)+PD(0)-PD(t). However, the maximal level of disorder in individual fruit does not have to be the same in all cases and equal to initial substrate level. It depends on the balance between processes of formation and scavenging of free radicals (see eq. 7), and this balance changes within the time. The effect of the initial conditions as level of scavenging system (SS0=SS(0)) and nutrients (N0=N(0)), with fixed reaction rate constants ($k_f$ and $k_s$ at constant temperature) (see Fig. 2 and 3) is clear now. When the SS0 level is low, or the N0 level is high, then the formation of radicals overwhelms the scavenging process (eq. 7) and the development of disorder is very fast. All substrate turns into disorder. The opposite situation (high SS0 or low N0) causes a slow development of disorder and the final level is below the initial level of substrate: not the whole substrate is turned into disorder due to the effective scavenging of free radicals. Of course, combining the effect of these conditions will result in a rise or a reduction as compared to the situation described above. So, all types of observation found in practice can be described and simulated by the model.

The initial level of free radicals (Ra0=Ra(0)) plays a different role. Although it also affects the final level of disorder, that effect is relatively small (Fig. 4). The major importance of this condition is its effect on the delay of visible occurrence of disorder. The lack of, and precisely a very low level of, free radicals at the beginning of process (at harvest) requires a relative long time before the increase of presence of radicals enables the faster formation of disorder. On the other hand a very high level of Ra0 will cause a very fast (almost instantaneous) development of disorder. The brief analysis of equation 7 helps to explain the reason of this unique effect. The level of radicals is the only condition present in both processes (formation and scavenging) that governs the changes of the level (due to formation and scavenging) of free radicals. That is why, provided a certain balance between other parameters exists, the initial level of radicals has such a unique effect on disorder development.

Different combinations of initial values of model parameters values allow the simulation of a very different range in type of development of disorder. From very fast

and reaching high level of occurrence (Fig. 5 a), very fast but stabilised at low level of occurrence (Fig. 5 b), to slow but reaching high final level of occurrence (Fig. 5 c), and slow and stabilised at low level of occurrence (Fig. 5 d). This suggests that different patterns of development of physiological disorders found in practical research over the years (Lourens and Malherbe 1999, Kruger et al. 1999) could be simulated with the same common values of kinetic rate constants. The variation of observed development of individuals within a batch of fruits can be now explained as the variation of initial levels of model batch parameters, i.e. S0, N0, Ra0, SS0. This in turn leads to conclusion that information on biological variation of batch parameters is essential for prediction of further changes of batch as a whole (Tijskens et al., 2003).

Finally, the simulated effect of different $CO_2$ levels on disorder development is shown in Fig. 6. The increase of $CO_2$ concentration causes decrease of scavenging system activity, and thus faster development of disorder.

It has to be stressed, however, that all values of all parameters used are chosen arbitrarily and are absolutely not calibrated against measured observations.

## COMPARISON WITH EXPERIMENTAL DATA

The experimental data were supplied by the members of European Research Project FAIR1-CT96-1803, coordinated by Dr. H. Peppelenbos of ATO-DLO (Wageningen, The Netherlands). Two of project partners harvested pears (cv. 'Conference') at several different stages of maturity and examined the occurrence of disorders three times during 6 or 6.5 months of storage periods. These experiments included two levels of $CO_2$ in the storage atmosphere. The infected fruit were sorted into three classes of internal browning severity and three classes of severity of cavities.

The data show (Fig. 7) that different locations, harvest dates and levels of $CO_2$ affected the number of fruit (within a batch) in which the disorders had developed. It is visible that at one location the harvest date is the factor which mostly affected the development of disorder (upper graph), while at an other location the most important factor is the level of $CO_2$ during storage (lower graph). The presented model can theoretically simulate both these observations. The data show also that even within each $CO_2$-harvest combination the development of disorder is not uniform. The different sets of data (especially for latest harvest date and high level of $CO_2$) present different individual development of disorder. This difference can also be simulated by the model, by simulating batches of individually developed fruits and averaging results within a batch. However, the disorder occurrence in some sets of fruits declined at the end of storage period. This decline demonstrates the weakness of used destructive method of assessment of disorder development. Since process of disorder cannot reverse, the observed decline of affected pears can be explained only as the lack of representativeness of fruits left after first two terms of disorder assessment.

Skipping this pitfall and taking into consideration only the data with increasing (or stable) development of disorder did not allow the statistical estimation (using non-linear regression analysis) of all parameters of the model. This attempt failed due to an insufficient number of data for each combination. Firstly the development of disorder was represented by only three terms of assessment, and there was no information on initial storage period when dynamics of disorder development could be various for the same final level of disorder occurrence (see Fig. 5). Secondly, there were only three sets of fruits investigated for each $CO_2$-harvest combination, which gave number of data not sufficient for statistical estimation of model parameters.

## CONCLUSIONS

The presented model shows that different initial levels of batch parameters (i.e. S0, N0, Ra0 and SS0) can have a tremendous effect on the predicted development of storage disorders. This includes the delay of occurrence of disorder and the final level of

disorder development in individual fruits. Therefore, the model is able to simulate actually every pattern of disorder development observed in practice.

Simulation of the effects of different storage conditions (e.g. $CO_2$ level, temperature) is also possible using the model.

Because the mechanisms of model are described as chemical processes and further developed following the fundamental rules of chemical kinetics, it can be used for simulation of any type of fruit tissue disorder (for which the processes are as described above). Each type of disorder will have the kinetic parameters (reference reaction rate constants and activation energies, described in eq. 4) for each of processes involved common for all batches from different locations or for all fruits grown at certain location. The differences between different locations or different batches of fruit from one location will be represented by variation of values of batch parameters (i.e. S0, N0, Ra0, and SS0).

Practical calibration of the model requires data with a good representation of disorder development. Several observations during first stages of disorder development are necessary. Since the disorder development is various for different individual fruits further studies on its mechanism require use of a non-destructive method of measurements.

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**Figures**

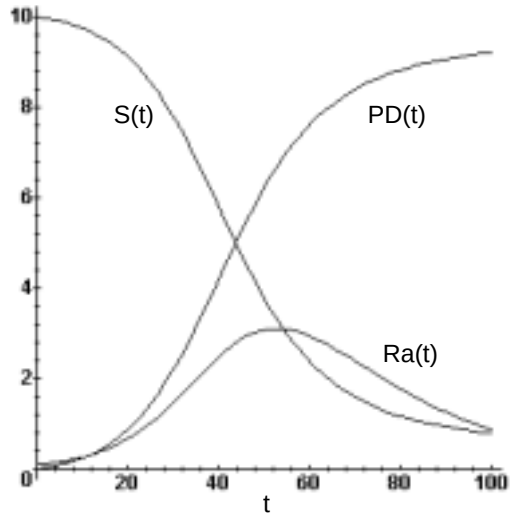


Fig. 1. Simulation of physiological disorder (PD), substrate (S), and free radicals (Ra) changes as a function of time (t). Along with rapid development of disorder, the amount of radicals first increases and then decreases when amount of available sound substrate becomes low.

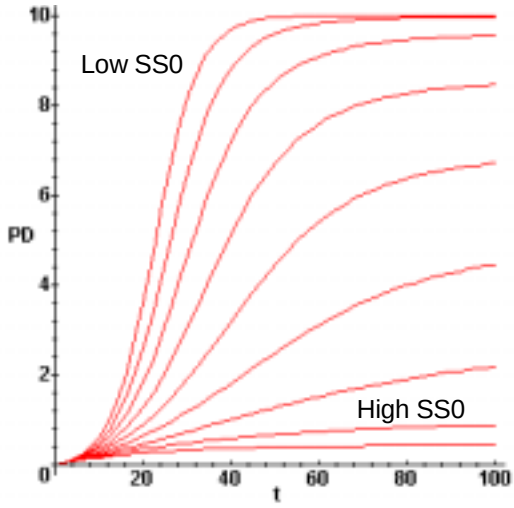


Fig. 2. Simulation of the effect of different scavenging activities on the development of physiological disorder (PD). High initial level of scavenging activity (SS0) keeps low amount of free radicals, and hence slow development of disorder.

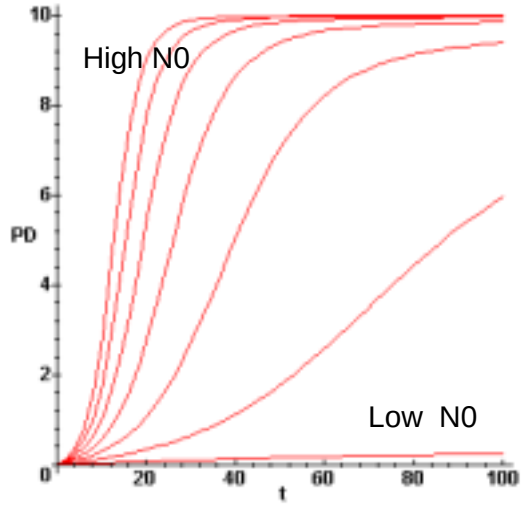


Fig. 3. Simulation of the effect of different nutrients levels on the development of disorder. High initial level of nutrients (N0) induces rapid development of disorder, while low N0 keeps formation of disorder very slow.

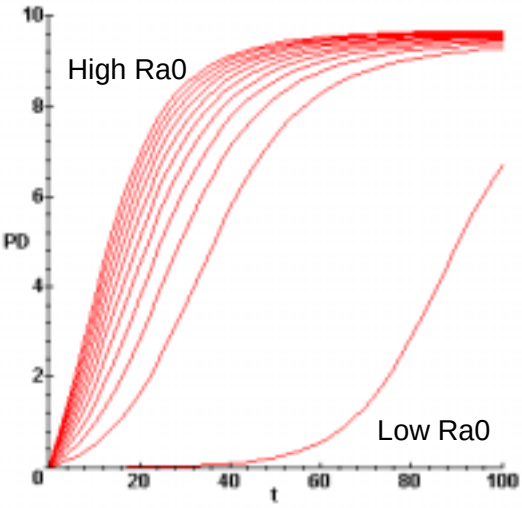


Fig. 4. Simulation of the effect of different initial free radicals levels on the development of disorders. High initial level of free radicals (Ra0) induces immediate development of disorder.

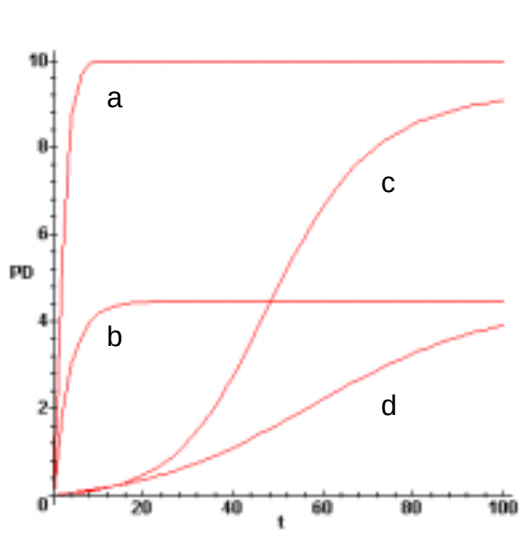


Fig. 5. Simulation of different patterns of development of disorders. Lines "a" and "b" represent rapid development of disorder which continues until whole substrate became affected by disorder ("a") or until there is no more free radical or nutrients available ("b"). Lines "c" and "d" represent similar situations but with lower initial level of free radicals.

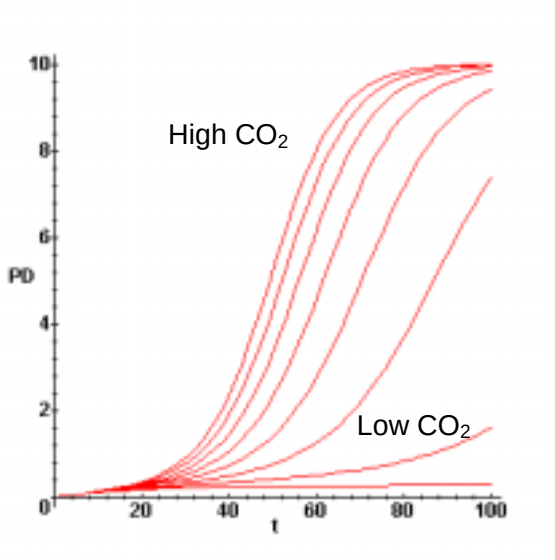


Fig. 6. Simulation of the effect of $CO_2$ concentration on the development of disorder. High $CO_2$ level reduces activity of scavenging system, and hence induces faster development of disorder. Low $CO_2$ level does not affect so much the scavenging system, which slows down the development of disorder.

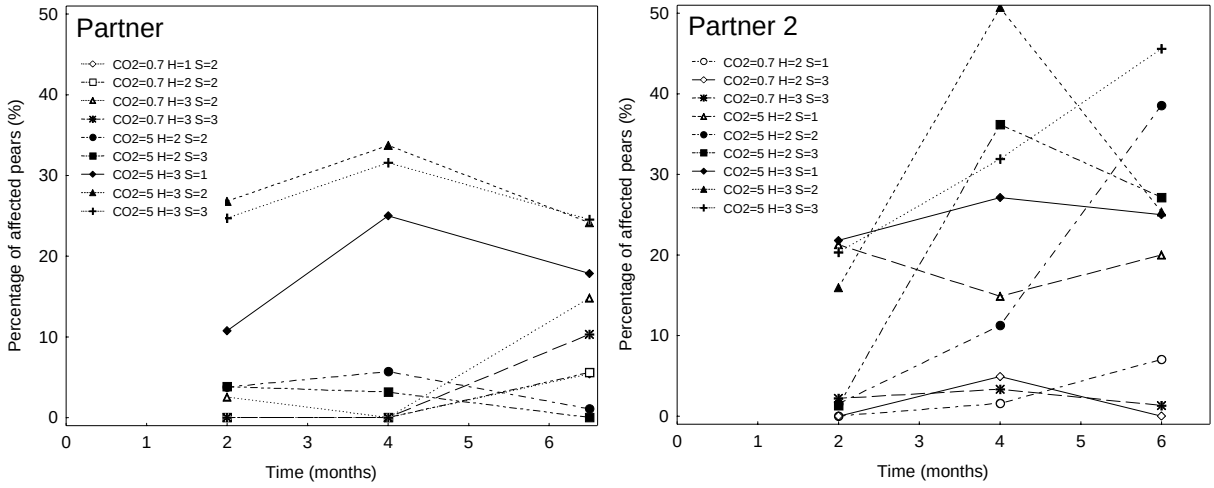


Fig. 7. Differences in disorder development between sets of fruits (S) examined as repetitions at selected combinations of $CO_2$ level and harvest number (H) for two different locations. Sets not present at the graphs showed no disorder development at any of the examinations.