Fluorescence Excitation Spectroscopy for the Evaluation of Seeds

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Summary

The measurement of the emission of light from entire samples [Popp 1988, Popp 1993, Köhler et al. 1991] is a non-destructive system that is already in use for inspecting foods. The biophoton fluorescence excitation spectroscopy presented here differs from the classical biophoton evaluations in that its evaluation follows excitation by different wavelengths of light. Selected groups of foods generate typical spectral distributions[Strube 1997, Strube/Stolz 1999]. These are suitable as the basis for evaluation when inspecting foods, especially seeds such as grains, peas and beans, oilseeds and etc. from test cultures using a variety of cultivation methods. In 1997 and 1998, Markus Buchmann and Christian Hiss performed comparative plantings of beans both without earth (hydroponics) and with earth (biodynamic) [Buchmann 1999]. Coded samples from these test plantings were examined using induced luminescence. The individual growing procedures were able to be markedly differentiated statistically. There were demonstrable differences in the seeds after 2 generations.

Material and Methods

In biophoton testing one has to differentiate between the induced emission [Popp 1988, Popp 1993, Köhler et al. 1991] and the continuous emission of a sample following dark adaptation as originally measured by Popp and Ruth [Ruth 1977]. In the case of induced emission according to Popp, there was a defined illumination of the sample with white or colored light with the time-resolved measuring of the emission at the end of the illumination. This procedure is also known as time-resolved fluorescence excitation spectroscopy.

The relative medium-term luminescence emitted after 6 to 10 seconds (R40 %)) proved itself best suited for the purpose of differentiation in our studies [Strube 1997, Strube/Stolz 1999]. We term the emission as being wide-band when the red, yellow and green excitation increases and the blue decreases somewhat. Conversely, we speak of the tendency towards being narrow-band when the red, yellow and green excitability declines slightly with blue increasing somewhat.

These whole (non-comminuted) seed samples originated from controlled test cultures and were capable of germination. In this work 8 samples were measured 7 times each. The samples were illuminated with a halogen lamp through different colour filters. The lightemission of the sample was evaluated with a cooled photomultiplier (EMI 9558 QA) with single photon counting according to Strube 1997.

From previous work with a variety of different plant materials and their isolated ingredients, it is known [Strube 1997, Strube/Stolz 1999] that leaves, along with other vegetative parts of plants, demonstrate a wide-band spectral distribution (Figure 1, stinging nettle leaf). With induced luminescence, chemical substances (Figure 1, citric acid) on the other hand, show an extremely narrow band (virtually exclusive excitability by blue light) both during short-term and medium-term luminescence. Germinable seeds (Figure 1, wheat) show a position in between. Whereas the narrow-bandedness (blue) is especially pronounced during short-term luminescence (Figure 2, left), the medium-term luminescence shows the additional involvement of other colors with an enduring preference for excitation by blue light (Figure 1).

Seeds are a depiction of plants in their dormant state. They appear practically unchangeable, as if lifeless. They only enter the vegetative stage and begin to germinate with the introduction of water. All the previously studied seeds show a similar spectrum of excitation, so that wheat can be used to provide a more fundamental depiction in representing these seeds. If you compare the relative excitation spectrum of wheat (as a representative seed) with that of the stinging nettle leaf (representative of green plants) and citric acid (representing a chemical substance), the seed will fall in between the green plant and the lifeless chemical substance (Figure 1).

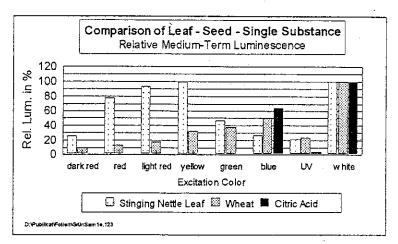
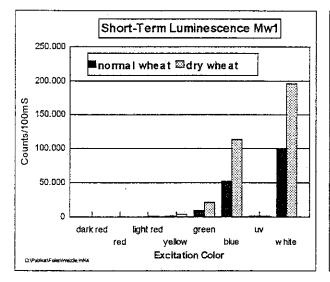


Figure 1: Seeds (represented here by wheat) lie between green plant material and chemical substances as determined by their excitability from different colors of light.

The blue of the excitation spectrum points towards lifelessness, while a wide-band indicates a plant origin. Corresponding to this is the increasing wide-bandedness of excitation during germination, ultimately becoming comparable to the level of green plant material in the case of seedlings as they begin leafing out.

Differences in the samples' humidity show how sensitive and consistent this method is. Wheat was stored at <10% and 60% relative humidity at constant temperature for 3 weeks. The seed's dormancy was more distinct in the dryer wheat, identifiable by its more narrow-banded luminescent behavior with a relative increase in excitation through blue light (Figure 2). Conversely, the moister seeds showed a lower, but more wide-band, emission.



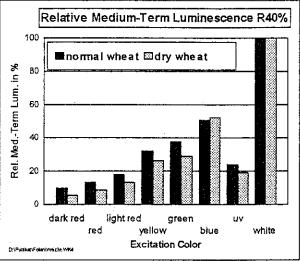
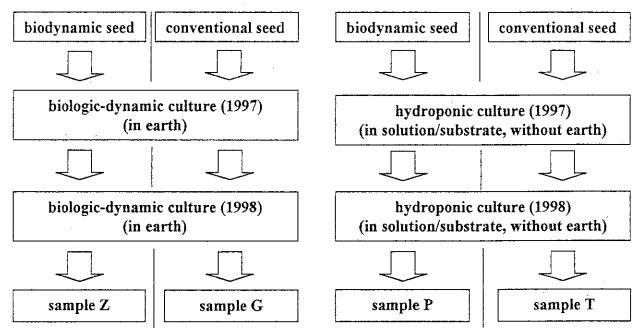


Figure 2: At lower humidity, seeds (here wheat) show a higher short-term luminescence and a narrow-band, and a more bluish excitation during relative medium-term luminescence.

This behavior already indicates that it is possible to make an evaluation of seeds: We see dormancy as characteristic for seeds. This state of quiescence appears in biophoton excitation spectroscopy as a higher Mwl value and as a relatively narrow-banded blue in the R40% value.

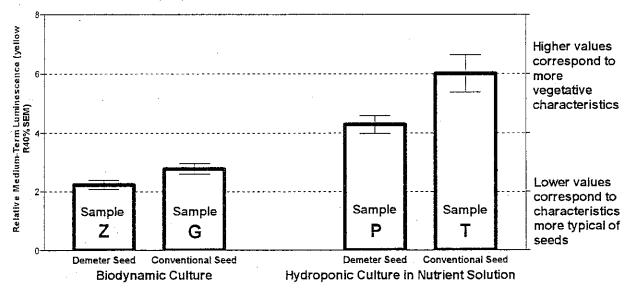
The beans were examined using the luminescence method (fluorescence excitation spectroscopy) [Strube 1997, Strube/Stolz 1999] after a cultivation period of 2 years. To provide a better overview, only that part of the entire excitation spectrum generated by yellow light is depicted (Figure 3).

Overview of Test Cultures of White Beans According to Buchmann et al. 1999



Luminescence-Characteristics of white Beans for Different Cultivations and Variations of Seed Origin

(Samples from the Buchmann/Hiss Experiment 1998)



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Figure 3: The bars in this figure show the relative medium term luminescence (R40%) for yellow light as example. The height of the bars show the luminescence intensity standardized to emission after white light. Even after 2 cultivation periods there was a clear difference between beans originating from biodynamic and conventional seeds. The beans originating from conventional seeds deviated after two biodynamic culture periods still in the same direction as all seeds under hydroponic cultivation. Under hydroponic culture beans originating from biodynamic seeds showed data with a tendancy to in earth culture.

Bahn98gelbNachbauE.pzi

Results

The encoded samples of white beans are classified correct to the different cultivation by fluorescence excitation spectroscopy. Lower values of luminscence after illumination with yellow light point to sample characteristics more typical of seeds. The bandwidth of excitation (see above) is the actual criterion. Narrow-bandedness corresponds to lower values for yellow, so that in this case only the values for yellow are depicted in order to provide better overview. Sample Z was the Demeter seed sample that was twice cultivated biodynamically. The conventional seed shows altered characteristics in as few as two biodynamic cultivations. Samples P and T, originating from two hydroponic cultures, show clear vegetative qualities. Even here the seed origin had an impact. The Demeter seed showed less vegetative qualities than conventional seed as a result of its hydroponic cultivation.

Discussion

These results show that different cultivation procedures lead to different products and that the origin of the seed will have an effect even after 2 generations. It still has to be examined as to whether or not the results attained from the beans will also appear in other cultures. However, it supports, that it is justifiable today to prescribe the use of ecological seed in ecological farming.

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