



Field phenotyping and an example of proximal sensing of photosynthesis.

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**A paper from the Proceedings of the
14th International Conference on Precision Agriculture
June 24 – June 27, 2018
Montreal, Quebec, Canada**

Abstract. *Field phenotyping conceptually can be divided in five pillars 1) traits of interest 2) sensors to measure these traits 3) positioning systems to allow high throughput measurements by the sensors 4) experimental sites and 5) environmental monitoring. In this paper we will focus on photosynthesis as trait of interest, measured by remote active fluorescence. The sensor presented is the Light Induced Fluorescence Transient (LIFT) instrument. The LIFT instrument is integrated in three positioning systems. First in an automatized rail based positioning system moving in x, y and z direction above 120 miniplots. Miniplots are large (0.8m²) soil-filled containers placed inside and outside the greenhouse. Second the sensor is mounted on a manual operated Field4cycle and third on a fully autonomously moving, engine driven, GPS steered cart, called FieldCop. Photosynthetic traits were quantified for major crop species across seasonal changes in environment, grown at elevated CO₂, or at different irrigation regimes. The quantum efficiency of photosystem II (F_q'/F_m') was light dependent whereas the electron transport rate efficiency (Fr₂'/F_m') was temperature dependent. Mean values of these photosynthetic traits or their interaction with environment allowed for characterization of different phenotypes. The LIFT instrument combined with selected positioning system provides high throughput proximal sensing of novel photosynthetic traits.*

Keywords. *Field phenotyping, LIFT, active fluorescence, high throughput, photosynthesis.*

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Introduction

In plant phenotyping the goal is to deliver quantitative data on the dynamic responses of plants to the environment (Fiorani and Schurr 2013). Non-invasive sensors provide a tool to enable, most preferably at affordable costs, field phenotyping (Araus and Cairns 2014). To face the challenges the future climate imposes a field phenotyping framework was described in (Cendrero-Mateo, Muller et al. 2017) where sensors and positioning systems for proximal sensing are two of the five main pillars (fig. 1). To understand how the expression of crop genetic information is modified by the environment, the first pillar is to identify the relevant traits that need to be monitored, to define what sensors are needed. The third pillar are positioning systems to enable accurate and high-throughput field phenotyping matching the traits to be measured. The next pillar consist of environmental sensors, obvious in determining the result of gene x environment interaction, but often underrepresented in field trials. The remaining pillar are the experiments, field trials or breeding plots the field phenotyping framework is built for and/or defined by. In this paper we will describe the Light Induced Fluorescence Transient (LIFT) instrument, a sensor for proximal sensing of photosynthetic traits, integrated in 3 kinds of position systems; a gantry system above miniplots, manual operated field4cycle and the fully automatized fieldcop. In this paper we describe the sensor and positioning systems in detail, whereas the photosynthetic traits measured will be shown in the presentation and are currently being submitted for publication.

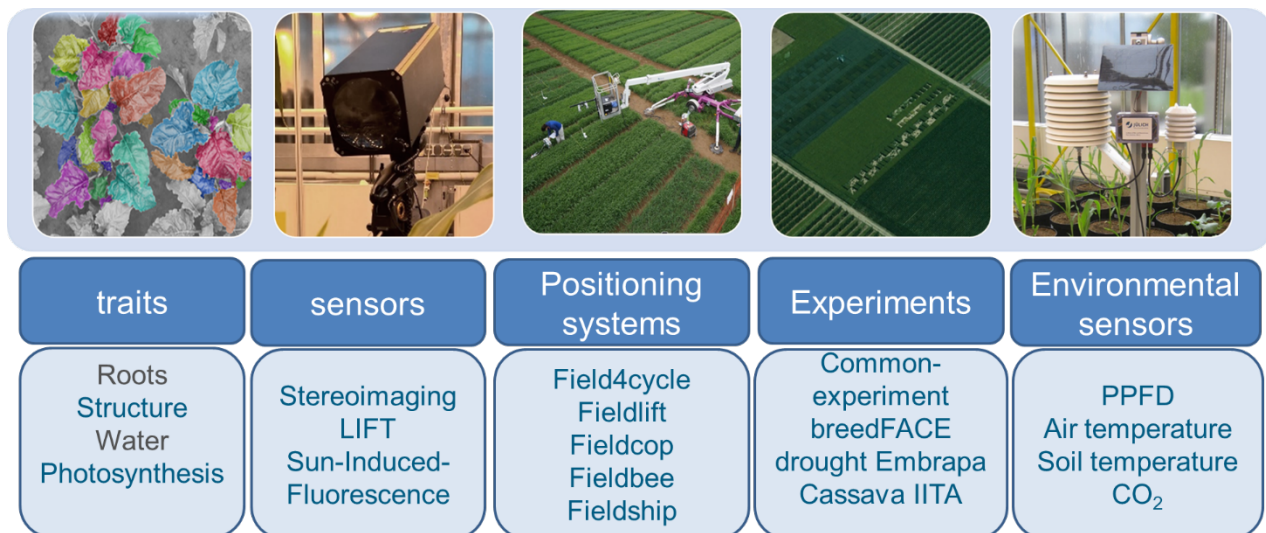


Fig 1: The pillars of field phenotyping, modified from [3] showing examples of the field phenotyping infrastructure developed for shoot phenotyping in Forschungszentrum Jülich, Germany

LIFT

The light induced fluorescence transient (LIFT) method is a unique approach to probe photosystem II from a distance under natural conditions (Kolber, Prášil et al. 1998, Pieruschka, Klimov et al. 2010, Osmond, Chow et al. 2017). The short measuring time of the LIFT method, taking no more than ~0.2 s per measurement, is ideal for integration into automated systems for high throughput phenotyping. In the LIFT standard protocol, QA is reduced within 750 μs using fast repetition rate (FRR) subsaturating (actinic) measuring flashlets to induce FmQA. Subsequently, a decreasing repetition rate of flashlets allows reoxidation of QA which can be simultaneously monitored as fluorescence relaxation. For terrestrial plants, a previously developed LIFT system using a laser excitation source was able to measure fluorescence transients in leaves and canopy from a distance of 50 m (Pieruschka, Albrecht et al. 2014,

Raesch, Muller et al. 2014). The operating efficiency of photosystem II (F_q'/F_m') measured with this previous LIFT system correlated well with pulse amplitude modulation (PAM) measurements ($r^2=0.89$) and CO₂ assimilation rates ($r^2=0.94$) (Ananyev, Kolber et al. 2005, Pieruschka, Klimov et al. 2010, Pieruschka, Albrecht et al. 2014). But the LIFT also measures photosynthetic parameters thus far not available in proximal sensing such as the functional absorption cross-section (Osmond, Chow et al. 2017) and reoxidation efficiency from 0.8 ms to 5.9 ms (F_r2/F_m) (Keller, Vass et al. 201x).



Fig 2. Light Induced Fluorescence Transient (LIFT) instrument (black rectangular camera) integrated in different positioning platforms from left to right measuring various crops in the miniplots, in the field in soybean Embrapa, Brazil, in wheat BreedFACE, Germany and with the field cop measuring maize in the common experiment in Germany,

Miniplots

The miniplots are large planting containers where plants can grow in a canopy that resembles field conditions (fig 2.). The miniplots are 535 liter plastic containers (inside size 111 x 71 x 61 cm) filled with soil upon a gravel layer for drainage. For this experiment the miniplots were filled with a loamy-clay silt soil (luvisol) from the surrounding agricultural fields. Each miniplot has computer steered drip irrigation system. In total 120 miniplots are placed in a fenced-in area of which 90 are inside and 30 outside. The fenced in area is in accordance to safety regulations for continuous operation of the automated sensor positioning system. This system was co-developed by Forschungszentrum Jülich and consists of a stable and motorized x, y, z rail based traversing unit that moves a universal base plate, on which various sensors of up to 50 kg can be attached and positioning <2cm accuracy above the miniplots. The sensors and traversing unit are controlled by an in-house developed application based on LabVIEW (National Instruments, USA) allowing for a wide variety of measurement protocols upon demand of the scientist. Furthermore multiple environmental sensors are placed in the miniplot area to monitor the environmental conditions and potential gradients; monitoring includes irradiance, air temperature and humidity, as well as soil moisture and temperature.

Field4Cycle

The field4cycle is a manually pushed frame on wheels at the exact width of existing (seeding) tractor trails based upon the design of White and Conley, 2013 and can carry a wide range of sensors. This affordable positioning system allows to measure crops from above up to 1.5m and easy operation for stationary measurements or scanning at self-defined speed (fig. 2).

Fieldcop

The FieldCop is a high precision 3D-positioning system for field measurements with custom sensor equipment based on a hydrostatically driven small vehicle (fig. 2). The vehicle has a deadweight of 1.6 metric tons and chassis dimensions of 300 x 130 x 98 cm (L x W x H). In addition it offers standard agricultural connectors such as a 3-point hitch, two power take-offs and

hydraulics connections for accessory equipment. A maximum velocity of 8 km/h is possible. In manual mode the vehicle is controlled by using a remote control. A boom is installed on top of the vehicle (Pic. 2), which is able to pivot laterally 90 degrees above experiment plots. The vertical position of the boom is controlled by a hydraulic lift system. The booms platform can spatially position a variety of sensors by using two motor driven shafts. The platform can reach a height of 4 m above ground and up to 3.8 m apart from the vehicle. The sensor platform has a maximum payload of 12 kg (Pic. 4). Coarse positioning (+/- 15 cm) of the vehicle towards the experiment plots is implemented by using a built-in autopilot / steering support based on RTK-GPS (Real Time Kinematic) (Pic. 3). Spatial fine positioning (+/- 5 cm) is implemented using an additional RTK-GPS-sensor mounted directly to the sensor platform. Depending on the distance between measuring points or plots and the used sensors 10-50 measurements an hour are possible.

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