

# **Session V**

# Management of abiotic stresses for sustainable intensification of root and tuber crops

# Lead lecture

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# Drought stress tolerance traits of potato

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Demand for potato is steadily rising in developing countries, where present per hectare production levels reach merely fractions of the yields achieved in Europe or North America. Many potato production areas in developing countries are located in semi-arid areas, where drought spells account for large harvest losses. In these regions, where the yields actually range around 30% of the global mean, adapted drought tolerant potato varieties could confer considerable yield increments and thus contribute to satisfy the growing demand. Physiological, biochemical and molecular analysis of tolerant and susceptible potato cultivars under water stress pinpoints traits that could mitigate yield drops caused by drought. Principal tolerance traits that could diminish the vulnerability of potato yields to drought stress include improved transpiration efficiency, which is associated with optimized stomatal control under drought to reduce water loss but at the same time allow for continuous CO<sub>2</sub> access for photosynthesis, enhanced detoxification of reactive oxygen species produced during stress, and mechanisms to protect proteins and membranes from damage by water stress. Candidate genes underlying these traits as well as genotypes that express them are available and, after appropriate validation, could be used in breeding. Future efforts could address improving photosynthetic efficiency and reducing photorespiration under drought.

**Keywords:** Potato, drought tolerance, photosynthesis.

# Introduction

Potato is a drought-susceptible crop. Nevertheless potato water productivity on dry weight basis is twice as high as that of wheat or maize and three times higher than that of rice (Trebejo and Midmore, 1990; FAO, 2003). Drought decreases both potato yield and quality. Water stress affects potato development at virtually any stage. During early developmental stages, before tuberization, drought may reduce stolon number (Haverkort et al., 1990), while drought during tuberization decreases tuber number and during bulking tuber size (Martin et al., 1992).

Tuber yield is a function of the amount of intercepted light and water availability (Allen and Scott, 1980). Light interception depends on the photosynthetically active leaf area. The larger this area, the larger the surface exposed to transpiration. Drought generally reduces plant growth and canopy size (Deblonde and Ledent, 2001; Tourneux et al., 2003), diminishing the evaporative demand and improving plant survival under water stress. But reduced canopy results in decreased yields due to the smaller leaf area available for photosynthesis. Early leaf appearance combined with the ability to sustain leaf growth under increasing soil moisture deficit would improve productivity in the presence of drought (Jefferies and MacKerron, 1993). Deeper and denser roots would enhance water availability, where water remains available in deeper soil layers.

Maintenance of photosynthetic activity under water stress is a key element of plant drought tolerance. Under water stress, photosynthesis per leaf area is mainly restricted by stomatal and mesophyll limitations, i.e., in how far  $CO_2$  remains available for the photosynthetic apparatus, when stomatal and mesophyll conductance is kept low to avoid excessive transpiration. Only at high stress levels non-stomatal metabolical limitations, such as reduced ribulose bisphosphate carboxylase regeneration and ATP synthesis inflict carbon assimilation under drought. However, at high irradiances RuBP is present in excess and  $CO_2$  should remain the limiting factor for photosynthetic rate (Parry et al. 2007). Low  $CO_2$  availability for the photosynthetic apparatus favors the oxygenase reaction of ribulose bisphosphate carboxylase and photorespiration, resulting in up to 50% reduction in carbon gain compared to well-watered conditions.

Water stress leads to increased production of active oxygen species in plant cells. While active oxygen species have regulative roles, excessive accumulation of these compounds can damage proteins and membranes. Mitigation of oxidative stress therefore can enhance plant survival under water stress.

In the last years we have investigated drought responses in tolerant and susceptible potato cultivars on agronomical, physiological, biochemical and molecular level. These investigations showed variation in water stress sensitivity in potato germplasm and pointed towards mechanisms that confer drought tolerance to this crop.

# Drought tolerance traits of potato

# Water use efficiency

The amount of harvested biomass per unit of transpired water (i.e., water use efficiency - WUE) under drought and well-watered conditions is a genotype-dependent trait. Large variation of WUE has been found in potato (Fig. 1). Appropriate stomatal control is one of the keys for high water use efficiency. Plants with high WUE would be highly suitable to increase yields where water resources are scarce. An important component of WUE is the regulation of abscissic acid biosynthesis and sensitivity of the plant. Together with an array of other factors, abscissic acid co-regulates growth and stomatal behavior (reviewed by Wasilewska et al. 2008). The right balance between abscissic acid accumulation and sensitivity of the plant is crucial for activating tolerance mechanisms against drought without completely inhibiting plant growth. Variation in abscissic acid content in potato germplasm under drought has not yet been systematically assessed.



Figure 1. Variation of water use efficiency under well-watered and drought conditions in improved CIP potato clones. Water use efficiency was determined at CIP according to Krishnamurthy et al. (2007). DM: dry matter.

# Increased harvest index under drought

Stress might alter carbon allocation to different tissues. Under heat, carbon transport to tubers is restricted; instead, above ground tissues act as sink for sugars, resulting in sugar-mediated reduction of photosynthesis and reduced tuber growth (Lafta and Lorenzen 1995, Timlin et al., 2006, Basu and Minhas, 1991). Similarly drought alters carbon partitioning in potato. Allocation of sugars to tubers varies under drought and both increase and decrease of harvest index is observed in a genotype-dependent manner (Fig. 2). For drought prone areas, genotypes with increasing harvest index under drought would be preferred.



igure 2. Variation of harvest index under well-watered and drought conditions in improved CIP potato clones under drought and well watered conditions.

# Increased root size

Some potato genotypes have the capacity to increase root size under drought. Together with reduction of canopy size under water stress, this might lead to dramatically increased root/shoot size ratios in some genotypes. Genotypes with increased root/shoot size ratios support drought better and generally have less yield loss under water stress than clones that lack the capacity to enlarge their root system under water stress conditions.

# Detoxification of reactive oxygen species

Reactive oxygen species are produced by the Mehler reaction and by the antenna pigments, during photorespiration and detoxifying reactions catalyzed by cytochromes, as well as in oxidative processes in mitochondria (Asada and Takahashi 1987). They act as signaling molecules for regulating development and various physiological responses (Miller et al., 2008). Under stress, reactive oxygen species accumulate and can reach toxic levels in the plant. Reactive oxygen scavengers like superoxide dismutase, ascorbate peroxidase, catalase, glutathione peroxidase, peroxiredoxin and others can mitigate the toxic effects of these molecules (Watkinson et al., 2006, Schafleitner et al., 2007, Mane et al., 2008). Under drought, we have observed up-regulation of an array of putative active oxygen scavengers acting in the chloroplast. Moreover, an antiquitin family aldehyde dehydrogenase 7 gene was found induced in drought tolerant potato cultivars only. In model plants this gene is involved in the detoxification of reactive aldehyde species generated by oxidative stress-associated lipid peroxidation (Rodrigues 2006). This gene would be a good candidate for mitigating oxidative stress in mitochondria.

#### **Osmotic adjustment**

Decrease of osmotic potential in plant tissues as a consequence of solute accumulation is often observed in potato plants under drought stress. Although the contribution of osmotic adjustment for increased water uptake from drying soils is very limited (Serraj and Sinclair 2002), decrease of osmotic potential is considered as a desirable trait in plant drought tolerance (Levy 1983, Jefferies 1993, Heuer and Nadler 1998). Osmotic

adjustment contributes with turgor maintenance under increasing water deficit and the accumulating osmolytes act also as protein and membrane protectors (reviewed by Yancey 2005). Particularly under transient water stress, osmotic adjustment might improve plant survival and regeneration. Many metabolites have been proposed to contribute to osmotic adjustment, including metal ions, organic acids, polyamines, sugars, sugar alcohols, amino acids and their derivates, and proteins (Handa 1983).

#### Stabilization of proteins and membranes

Protein and membrane-stabilizing properties have been attributed particularly to late embryo abundant (LEA) and dehydrin proteins whose expression is strongly induced under drought stress (Schafleitner et al., 2007). Another protein group probably involved in protein stabilization and re-folding are chaperones such as DnaJ family and heat shock proteins. Genes encoding these proteins are highly expressed under drought stress and could be associated with tolerance (Watkinson et al., 2006, Schafleitner et al., 2007).

# Conclusions

The present knowledge on water stress tolerance in potato points towards several candidate traits that could be used for breeding drought tolerant potato varieties. For some of the proposed traits, such as increased root mass and enhanced ROS detoxification, strong evidence for their beneficial effect in drought tolerance is already available, while for other candidate traits, such as osmotic adjustment or protein and membrane stabilization-related mechanisms, direct proof for their function in yield maintenance under stress in potato is still missing. To determine the functionality of candidate genes and traits and to better understand the trade-offs such as the yield penalty caused by a tolerance trait, thorough agronomical analysis of genotypes expressing these candidate traits in different drought environments will be required.

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# References

Allen E.J., Scott R.K. 1980. An analysis of growth of the potato crop. J Agr Sci 94:583-606.

- Asada K., Takahashi M. 1987 Production and scavenging of active oxygen in photosynthesis. In : Kyle D.J. et al (eds) Photoinhibition (Topics in Photosynthesis), vol. 9, Elsevier, Amsterdam, pp227–287.
- Basu P.S., Minhas J.S. 1991. Heat tolerance and assimilate transport in different potato genotypes. J Exp Bot 42:861-866.
- Deblonde P.M.K., Ledent J.F. 2001. Effects of moderate drought conditions on green leaf number, stem height, leaf length and tuber yield of potato cultivars. Europ J Agron 14:31-41.
- Handa S., Bressan R.A., Handa A.K. 1983. Solutes contributing to osmotic adjustment in cultured plant cells adapted to water stress. Plant Physiol 73:834-843.
- Haverkort A.J., Waart M. van de, Bodlaender K.B.A. 1990. The effect of early drought stress on numbers of tubers and stolons of potato in controlled and field conditions. Potato Res 33:89-96.
- Heuer B., Nadler A. 1998. Physiological response of potato plants to soil salinity and water deficit. Plant Sci. 137:43-51.
- Jefferies R.A. 1993. Responses of potato genotypes to drought. I. Expansion of individual leaves and osmotic adjustment. Ann Appl Biol 122:93-104
- Jefferies R.A., MacKerron D.K.L. 1993. Responses of potato genotypes to drought. II. Leaf area index, growth and yield. Ann Appl Biol 122:105-112.
- Krishnamurthy L., Vadez V, Jyotsna Devi M., Serraj R, Nigam S.N., Sheshshayee M.S., Chandra S., Aruna R. 2007. Variation in transpiration efficiency and its related traits in a groundnut (*Arachis hypogaea* L.) mapping population. Field Crops Res 103, 189–197.

- Lafta A.M., Lorenzen J.H. 1995. Effect of high temperature on plant growth and carbohydrate metabolism in potato. Plant Physiol 109:637-643.
- Levy D. 1983. Varietal differences in the response of potatoes to repeated short periods of water stress in hot climates. 1. Turgor maintenance and stomatal behaviour. Potato Res 26:303-313.
- Mane S., Vazquez Robinet C., Ulanov A., Schafleitner R., Tincopa L., Gaudin A., Nomberto G., Alvarado Carlos., Solis C., Avila L., Blas R., Ortega O., Solis., Panta A., Rivera C., Samolski I., Carbajulca D., Bonierbale m., Pati A., Heath L.S., Bohnert H.J., Grene R. 2008. Molecular and physiological adaptation to prolonged drought stress in the leaves of two Andean potato genotypes. Funct Plant Biol 35:669-688.
- Martin RJ, Jamieson PD, Wilson DR et al (1992) Effects of soil moisture deficits on yield and quality of 'Russet Burbank' potatoes. New Zealand J Crop Horticultural Sci 20:1-9
- Miller G., Shulaev V., Mittler R. 2008. Reactive oxygen signaling and abiotic stress. Physiol Plantarum 133: 481-489.
- Parry M.A.J., Madgwick P.J., Carvalho J.F.C., Andralojc P.J. 2007. Prospects for increasing photosynthesis by overcoming the limitations of Rubisco. J Agric Sci 145:31–43.
- Rodrigues S.M., Andrade M.O., Soares-Gomes A.P. et al 2006. *Arabidopsis* and tobacco plants ectopically expressing the soybean antiquitin-like ALDH7 gene display enhanced tolerance to drought, salinity, and oxidative stress. J Exp Bot. 57:1909-1918.
- Schafleitner R., Gutierrez R.O., Gaudin A. Alvarado Aliaga C.A., Nomberto Martinez G., Tincopa Marca L.R., Avila Bolivar L., Mendiburu Delgado F., Simon R., Bonierbale M. 2007. Capturing candidate drought tolerance traits in two native Andean potato clones by transcription profiling of field grown plants under water stress. Plant Physiol Biochem 45:673-690.
- Serraj R., Sinclair T.R. 2002. Osmolyte accumulation: can it really help increase crop yield under drought conditions? Plant, Cell & Envir 25:333-341.
- Timlin D., Lutfor Rahman S.M., Baker J., Reddy V.R., Fleisher D., Quebedeaux B. 2006. Whole plant photosynthesis, development, and carbon partitioning in potato as a function of temperature. Agron. J. 98:1195–1203.
- Tourneux C., Devaux A., Camacho M.R., Mamani P., Ledent J.F. 2003. Effect of water shortage on six potato genotypes in the highlands of Bolivia (II): water relations, physiological parameters. Agronomie 23: 181–190.
- Trebejo I., Midmore D.J. 1990. Effect of water stress on potato growth, yield and water use in a hot and a cool tropical climate. J Agric Sci 114:321-334.
- Vasquez-Robinet C., Mane S.P., Ulanov A.V., Watkinson J., Stromberg V.K., De Koeyer D., Schafleitner R., Willmot D.B., Bonierbale M., Bohnert H.J., Grene R. 2008. Physiological and molecular adaptations to drought in Andean potato genotypes. J. Exp. Bot 59:2109 2123.
- Wasilewska A., Vlad F., Sirichandra C. 2008. An update on abscisic acid signaling in plants and more. Mol Plant 1: 198–217.
- Watkinson J.I., Hendricks L., Sioson A.A., Heath L.S., Bohnert H.J., Grene R. 2008. Tuber development phenotypes in adapted and acclimated, drought-stressed Solanum tuberosum ssp. andigena have distinct expression profiles of genes associated with carbon metabolism. Plant Physiology and Biochemistry 46, 34–35.
- Yancey P.H. 2005. Organic osmolytes as compatible, metabolic and counteracting cytoprotectants in high osmolarity and other stresses. J Exp Biol 208: 2819-2830.

# Early detection of drought stress in potato (*Solanum tuberosum* L.) and grapevine (*Vitis vinifera* L.) crops through multifractal analysis applied to remotely sensed data

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# Abstract

Drought stress is a growing agricultural concern, since climate variability and change are affecting the water cycle worldwide, and an increment of drought periods in some regions of the world have been forecasted for the upcoming years. Several methodologies are being developed for assessing, monitoring, and managing water availability in order to supply the accurate water amount to crops attaining the highest possible water use efficiency. In order to determine the ability of remote sensing for identifying and monitoring drought stress in potato and grapevine crops, continuous measurements of multispectral reflectance of plant canopies have been analyzed, and compared with measurements of physiological parameters simultaneously registered using gas exchange techniques, and existing spectral vegetation indices. The results evidenced that multifractal analysis of reflectance data in potato did discriminate between the Control (well irrigated) and Drought treatments around 6 days before the conventional gas-exchange assessment detected any difference. The difference was also shown by the split-reflectance spectrum. In grapevine, multifractal analysis did discriminate among treatments around 2 days prior to its discrimination by either gas-exchange or sap-flow. Therefore, multifractal analysis applied to multispectral remotely sensed data might become a useful tool for early drought stress detection to improve the water use efficiency by crops.

**Keywords:** Multispectral reflectance, Potato crop, Grapevine crop, Drought stress, Multifractal analysis, Wavelet transform.

# Introduction

Several attempts have been carried out in order to find a reliable methodology to assess drought stress in plants, since some common methods are becoming old-fashioned e.g. validity of the Scholander pressure chamber awaits new experimental evidence (Cochard *et al.*, 2001). Gas-exchange techniques have demonstrated high success in measuring the main physiological parameters of plants such as stomatal conductance (*g*) and photosynthesis ( $A_N$ ), but their disadvantage is the requirement of expensive complex equipments and well-trained technicians. Remote sensing, largely improved during the Second World War (Campbell, 1996; Lillesand *et al.*, 2004), has been used for monitoring the status of crops and natural vegetation on the basis of changes in plant reflectance patterns.

Fast metabolic changes occurring in plants, alter the reflectivity and propagation of solar radiation inside plant tissues, where a fraction is absorbed and other reflected in all directions (Gilabert *et al.*, 1997). Remote sensing captures details of reflectivity and absorption of light which are linked to the biochemical and structural components of the plant, such as chlorophyll, other pigments, water content, proteins, leaf thickness, cellular structure and cell wall and other biochemical materials (Ritchie, 2003; Blackburn and Ferwerda, 2008). All these components are affected by stresses, resulting in differences in the spectral signature of healthy and stressed plants that can be assessed through reflectance measurements (Chávez *et al*, 2009a). However, interpreting these patterns is not an easy task since most of the compounds of plant tissues act as radiation filters affecting the resultant spectrum, thus converting the reflectance response into a complex signal. Most of the meaningful spectral information is contained in small non-visible changes of the signal that conventional analysis had always obviated since they just focus on a small number of optimal wavebands while discarding the majority of the spectrum. Therefore, mathematical tools such as wavelets and multifractal analyses have the potential to capture much more of the information contained in reflectance spectra.

The aim of this work was to assess the value of remotely sensed reflectance as a reliable technique for early detection of metabolic changes caused by drought in plants, instead of destructive conventional methods. Also, the relevance of multifractal analysis to improve the accuracy and earliness of reflectance data for drought-stress diagnosis is presented.

# Materials and methods

# Plant material and treatments

**Potato.** Two experiments with potato (*Solanum tuberosum* L.) plants were carried out in a greenhouse in Mallorca, Spain, during the winter and autumn of 2006. Thirty plants of the cv. Marfona for the first experiment and 18 plants cv. Marie Spears for the second one, were grown in 10 litres pots of vegetal substrate (90%) and perlite-stone (10%). Three watering treatments were applied: *Control* (Ctrl), 100% of the daily measured evapotranspiration (dme), *moderate-drought* (D75), accounting for 75% of dme, and *severe-drought* (D50), 50% of dme. Management was similar for all plants. The treatments were initiated some 2 weeks after emergence. Thereafter, pots were weighted every day to determine water availability from evapotranspiration and irrigation was applied according to the treatment of every potted plant. Leaves from the same canopy strata were detached from sampled plants and their respective fresh weight (FW) registered immediately. Leaves were then kept in distilled water during 24 hours for determining their turgid weight (TW). Their dry weight was obtained after 48h at 60°C in a forced air oven. The relative water content was determined as RWC = [(fresh weight – dry weight)] to 0. Four replicates per treatment were obtained from different individuals.

**Grapevine**. An experiment was performed in six-year old grapevine (*Vitis vinifera* L.) plants under outdoors conditions, in Mallorca, Spain, during the summer of 2006. 12 plants cv. Tempranillo grafted on R-110 rootstock were grown in 60 litres pots containing a 20:80 v/v mixture of organic matter and sandy loam soil. Plants were irrigated daily until the end of July, and gas-exchange parameters and reflectance of solar radiation were measured. Plants were then submitted to three treatments (4 per each one) that lasted until the second week of September. The drought treatments were caused by stopping the irrigation of plants during 5 days per week. For treatment 1) *severe-drought* (D1), irrigation was re-initiated at the evening of the 6<sup>th</sup> day; and for treatment 2) *moderate-drought* (D2), irrigation was re-initiated at the early-morning of the 6<sup>th</sup> day. The *Control* (Ctrl), was normally irrigated at field capacity. Management was similar for all plants before application of the treatments. A 2cm layer of perlite and a thermal-keeper film was extended over each pot to diminish direct soil evaporation.

# Reflectance measurements

During the first indoor experiment with potato, canopy reflectance was measured with a USB2000 spectrometer (Ocean Optics, USA) covering the 350-800nm wavelength region, with a resolution of 0,5nm. During the second potato experiment and the grapevine experiments, a high resolution spectrometer HR-2000 (Ocean Optics, USA) covering the 180-1100nm wavelength region, resolution 0.065nm, was used. In both experiments, a white Spectralon® panel was used for calibration measurements, resulting in relative reflectance values. Three reflectance measurements per sample were taken and averaged to estimate its spectral variability. Measurements were performed every 1 or 2 d for grapevines and 2 or 3 d for potatoes, at noon, when solar radiation was projected vertically to the Earth. So, reflection was measured under an angle of 15° from nadir, minimizing shadow effects. The aperture angle of the fore optics of the spectrometer was 25°, placed at a distance of 2 cm from the leaf surface, resulting in a projected Field of View of around 1cm in diameter.

#### Gas exchange measurements

In both potato and grapevine, gas exchange measurements were performed using a portable infrared gas analyzer (IRGA) Li-6400 (Li-Cor Inc., Lincoln, USA).

# Sap flow measurements

Sap flow measurements were made using the thermo heat balance method (THB), which applies external heat into one point of the conductive xylem and sensed internal temperature in another near point of the same branch. Then, the sap flow was calculated according to the heat losses and flux-gradient theory (Lindroth *et al.*, 1995).

#### Wavelet and multifractal data analysis

The analysis described by Chávez *et al.* (2009b) was slightly modified by splitting the spectra into 4 bands to mimic those of Landsat TM: blue (450-520nm), green (520-600nm), red (630-690nm) and NIR (760-900nm). The proportion of the reflected radiation per band was calculated as a function of time (growth), resulting in heterogeneous reflectance spectra displaying singularities through time. This assessment was carried out to contrast the information it provided against the results of the wavelet-multifractal analysis of the entire spectra.

A software that describe the canonical method of wavelet multifractal modulus maximum (WTMM), originally implemented by McAteer *et al.* (2007) was modified to analyze the reflectance data. The methodology described by Arneodo *et al.* (1988) was followed and adapted to run in IDL6.3 for Windows. Data were processed with the Continuous Wavelet Transform (CWT) and the WTMM. The mother wavelet analyser used was the second derivative of the Gaussian function (Mexican hat). For more details, review Arneodo *et al.* (1995), McAteer *et al.* (2007) and Chávez *et al.* (2009b).

#### Data pre-processing

The pre-processing consisted of two sequential steps. In the first one, a background correction was required to account for variations in the signal due to both natural changes in the atmosphere within a sampling date. In the second step, the anomalies over moving averages of 41 wavelengths (see equations 2 and 3) were calculated. Anomalies reduce the signal to noise ratio thus making small fluctuations in the physiology of the plant more perceptible to the analyses. The background correction of the first step was performed by adjusting the measured response (G) of individual plant signals to a reference response through fitting a linear regression (Equation 1; Yarlequé, 2009).

Background correction:

 $S(t_i) = A.G(t_i) + B$ 

$$\hat{S}(t_i) = \sum_{k=i-20}^{i+20} \frac{S(t_k)}{41},$$
(2)

where *t* is the wavelength (ranging from 390 to 1020 nm), *i* is the time at the middle of the analyzed moving window, and *k* is the counter that allows the analyzed window to move from -20 to +20.

Anomalies:

Moving average:

$$S'(ti) = S(ti) - \hat{S}(ti)$$
(3)

# Statistical analysis

Differences between treatments means were analyzed with time-repeated measurements analyses performed with the SPSS 12.0 software package (SPSS Inc., Illinois, U.S.A.).

# **Results and discusion**

#### Multifractal spectra of reflectance

The raw reflectance spectra measured from plants did not allow a clear discrimination of the treatments, although some regions of the spectra regions demonstrated different reflectance values. However, discrimination was noticeable after pre-processing and multifractal analysis of data from the 8<sup>th</sup> day post

(1)

treatments application (dpt) (data not shown) for the first experiment, and from the 6<sup>th</sup> dpt for the second experiment (Figure 1), i.e. 7 and 5 days earlier than the gas-exchange measurements, respectively.

In grapevine, the resultant multifractal singularity spectra did show that after only 6 dpt, both D1 and D2 were completely differentiated from the control (Figure 2), and differences among treatments were held throughout the rest of the experiment. Differences were noticed 2 days earlier than showed by the gas-exchange measurements.



Figure 1. Passive reflectance of potato plants (*left*) obtained by the high resolution spectroradiometer HR2000, and their correspondent multifractal singularity spectra (*right*). Second experiment



Figure 2. Passive reflectance of grapevine plants (*left*) obtained by the high resolution spectroradiometer and their correspondent multifractal singularity spectra (*right*)

# Reflectance spectrum by regions and Vegetation Indexes

In potato, stress in plants was evidenced by their distinct reflectance pattern from around 7 and 5 dpt for the first and second experiment, respectively (P<0.05) (Figure 3). In grapevine, such differences were evident around 7 dpt (P<0.05). The main bands for detecting drought stress were the blue (P<0.01) followed by NIR, red and finally the green region of the electromagnetic spectrum (P<0.05).

In contrast, the spectral vegetation indexes (SVI) tested did show an inconsistent response (data not shown), even those specifically developed to retrieve water content of plants. SVI demonstrated to be an unreliable method for real-time monitoring of water status at whole plant level, probably due to the focusing on a few optimal wavebands while discarding the majority of the spectrum (Blackburn and Ferwerda, 2008). These findings confirm the statement of Gamon *et al.* (1990), Peñuelas *et al.* (1995) and Dobrowski *et al.* (2005), that SVIs are less useful for this dynamic monitoring.

# Gas exchange measurements

The first potato experiment showed differences among treatments for photosynthesis rate ( $A_{N}$ ) and stomatal conductance (*g*) from the 8<sup>th</sup> and 10<sup>th</sup> dpt, respectively (*P*<0.01) (Figure 4). During the second experiment, daily  $A_{N}$  and *g* did not show a clear trend. In grapevine, daily gas-exchange measurements did show rates with differences among treatments for  $A_{N}$  and *g* from the 8<sup>th</sup> dpt (*P*<0.01).

# Potato Relative Water Content (RWC)

During the first experiment the RWC for D50 was lower than for both D75 and Ctrl (P<0.05) at the 6<sup>th</sup> dpt, reaching their maximum difference at 13<sup>th</sup> dpt and onwards (P<0.01). During the second experiment, differences among treatments occurred at 10<sup>th</sup> dpt (P<0.05). However, at the 17<sup>th</sup> dpt RWC appeared similar (P≥0.05) probably due to a sudden increase of relative humidity (%), but later turned different again until the end of the trial, 26<sup>th</sup> dpt (P<0.01).



Figure 3. Reflectance of potato plants (2<sup>nd</sup> experiment) divided according to the bands of satellite Landsat TM



Figure 4. Gas exchange rates of plants of the potato (1) and grapevine (2) experiments. Daily photosynthesis (*a*) and stomatal conductance (*b*)

#### Sap flow measurements

A high similarity was observed between daily transpiration rates measured by the sap flow and gas-exchange in grapevine. During a typical sunny day (about 1500 µmol.m<sup>2</sup>.s<sup>-1</sup>, average temperature at midday around 33°C and 55% average of relative humidity), a high slope of sap flow was observed during the first hours in the morning. As the drought stress augmented, the sap flow slope progressively decreased day by day. Then, a continuous cycle of watering followed by progressive dehydration was observed weekly (Figure 5).



Figure 5. Continuous records of sap flow and details of readings. At the beginning of the experiment, detail in *a* indicates that there are not differences among treatments. Similar behaviour is shown in *b*. In contrast, difference among control and drought treatments is observable in *c* from day 8 post treatment and onwards

# Tuber yield

Potato is sensitive to drought with reductions in yield (Jefferies, 1993; Gregory and Simmonds, 1992; Jefferies and Mackerron, 1987; van Loon, 1981). However, our results indicated that potato was almost insensitive to moderate drought. Indeed, Ctrl and D75 obtained the same production per plant during the first experiment  $(0.79\pm0.05 \text{ kg plant}^{-1} \text{ for Ctrl} and 0.80\pm0.05 \text{ kg plant}^{-1} \text{ for D75})$ . On the other hand, plants under D50 did reduce their production  $(0.66\pm0.03 \text{ kg plant}^{-1})$ . Likewise, during the second experiment there was a similar trend among treatments  $(0.74\pm0.17 \text{ kg plant}^{-1})$  for Ctrl and  $0.725\pm0.031 \text{ kg plant}^{-1}$  for D75) whereas D50 presented the lowest yield  $(0.653\pm0.054 \text{ kg plant}^{-1})$ . Jefferies (1995) has pointed out that yield reduction under drought stress is mainly due to a reduction in canopy expansion, which may delay tuber initiation and bulking.

# Conclusions

Multifractal analysis of plant reflectance data enhanced the precision and earliness of diagnosis of drought stress compared to gas-exchange and relative water content measurements. The average difference for potato and grapevine was 4 days.

Multifractal analysis provided more reliable and stable results than splitting the spectrum into the main regions. Spectral vegetation indexes (SVI) were ineffective indicators for water stress in plants.

These findings show the advantage of using multifractal techniques to extract features from the complex signals generated by potato and grapevine plants under water stress. The challenge is to replicate these findings under commercial field conditions.

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# References

- Arneodo, A.; Grasseau, G.; Holschneider, M. 1988. Wavelet transform of multifractals. Phys. Rev. Lett., 61(20), 2281-2284.
- Arneodo, A.; Bacry, E.; Graves, P.V.; Muzzy, J.F. 1995. Characterizing long-range correlations in DNA sequences from wavelet analysis. Phys. Rev. Lett., 74(16), 3293-3296.
- Blackburn, G.A.; Ferwerda, J.G. 2008. Retrieval of chlorophyll concentration from leaf reflectance spectra using wavelet analysis. Rem. Sens. of Environ., 112(4), 1614-1632.
- Campbell, J.B. 1996. Introduction to remote sensing. The Guilford Press, NY, USA, p 622.
- Cochard, H.; Forestier, S.; Améglio, T. 2001. A new validation of the Scholander pressure chamber technique based on stem diameter variations. Journal of Experimental Botany 52(359), 1361-1365
- Chávez, P.; Zorogastúa, P.; Chuquillanqui, C.; Salazar, L.F.; Mares, V.; Quiroz, R. 2009(a). Assessing Potato Yellow Vein Virus (PYVV) infection using remotely sensed data. International Journal of Pest Management, 55(3), 251-256.
- Chávez, P., Yarlequé, C., Piro, O., Posadas, A., Mares, V., Loayza, H., Chuquillanqui, C., Zorogastúa, P., Flexas, J., Quiroz, R. 2009(b). Applying multifractal analysis to remotely sensed data for assessing PYVV infection in potato (*Solanum tuberosum* L.) crops. Remote Sensing Journal. Under revision.
- Dobrowski, S.Z.; Pushnik, J.C.; Zarco-Tejada, P.J.; Ustin, S.L. 2005. Simple reflectance indices track heat steadystate chlorophyll fluorescence at the canopy scale. Rem. Sens. of Environ., 97(3), 403-414.
- Gamon, J., Field, C., Bilger,W., Bjoerkman, O., Fredeen, A., Peñuelas, J. 1990. Remote sensing of the xanthophyll cycle and chlorophyll fluorescence in sunflower leaves and canopies. Oecologia, 85(1), 1-7.
- Gilabert, M. A.; García-Haro, F. J.; González Piqueras, J. 1997. Acerca de los índices de vegetación. Revista de Teledetección, 8, 1-10.
- Gregory, P.J., Simmonds, L.P., 1992. Water relations and growth of potatoes. In: Harris, P. (Ed.), The Potato Crop. Chapman & Hall, London, pp. 214-246.
- Jefferies, R.A., 1995. Physiology of crop response to drought. In: Haverkort, A.J., MacKerron, D.K.L. (Eds.), Potato Ecology and Modeling of Crops Under Conditions Limiting Growth. Kluwer Academic Publishers, The Netherlands, pp. 61-74.

- Jefferies, R.A., 1993. Responses of potato genotypes to drought. I. Expansion of individual leaves and osmotic adjustment. Ann. Appl. Biol. 122(1), 93-104.
- Jefferies, R.A., Mackerron, D.K.L., 1987. Aspects of the physiological basis of cultivar differences in yield of potato under droughted and irrigated conditions. Potato Res. 30, 201–217.
- Lillesand, T.M.; Kiefer, R.W.; Chipman, J. 2004. Remote sensing and image interpretation. John Wiley & Sons Inc, p 704.
- Lindroth, A., Cermak, J., Kucera, J., Cienciala, E., Eckersten, H. 1995. Sap flow by heat baance method applied to small size *Salix*-trees in a short-rotation forest. Biomass and Bioenergy, 8(1), 7-15.
- McAteer, R.T.J.; Young, C.A.; Ireland, J.; Gallagher, P.T. 2007. The bursty nature of solar flare x-ray emission. The Astrophysical Journal, 662(1), 691-700.
- Peñuelas, J.; Filella, I.; Lloret, P.; Muñoz, F.; Vilajeliu, M. 1995. Reflectance assessment of mites effects on apple trees. Int. J. Remote Sens. 16(14), 2727-2733.
- Ritchie, G.L. 2003. Use of ground-based canopy reflectance to determine radiation capture, nitrogen and water status, and final yield in wheat. Thesis for Magister in Science academic degree, Utah State University, USA.
- van Loon, C.D. 1981. The effect of water stress on potato growth, development, and yield. American Journal of Potato Research, 58, 51-69.
- Yarlequé, C. 2009. Análisis de campos de biomasa del altiplano usando wavelet y parámetros universales multifractales. Tesis de Licenciatura en Física. Universidad Nacional del Callao, Perú, p 202.

# Factors related to potato productivity

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# Abstract

Potato is the fourth most important crop in the world and it is an essential food resource worldwide. Phosphorus uptake, which is closely related to the potato crop yield, is both influenced by the soil nutrient concentration and the oxygen supply to roots. To evaluate the factors associated to phosphorus uptake, it was conduced a complete randomized blocks experiment in factorial (3x4) scheme with three grass crops (*Brachiaria brizantha* cv Marandu, *Panicum maximum* cv Tanzania and *Zea mays*), cultivated prior to potato cv. Atlantic, and four levels of phosphorus (100, 400, 700, 1000 kg.ha<sup>-1</sup> of P<sub>2</sub>O<sub>s</sub>) applied to soil before the sowing of the grasses. The residual phosphorus affected significantly the potato productivity, which was not influenced by the previous crop.

**Keywords:** Crop rotation, organic material, phosphorus uptake.

# Introduction

Potato is the fourth most important crop in the world and it is an essential food resource worldwide. The potato protein production per unit of area is 52% and 78% higher in comparison to wheat and rice, respectively. Few crops can produce that quantity of proteins and carbohydrates in the same area (Stevenson *et al.*, 2001). For the food security, the establishment of efficient production methods is essential, such as the crop rotation and the phosphorus (P) application. Phosphorus is an essential element to the starch synthesis and, consequently, to the potato productivity (Marschner, 1995). The potato P absortion efficience is lower in comparison to other crops, such as cassava and pumpkin (Dechassa et al. 2003; Vander Zaag et al. 1979). Pursolove & Sanders (1981) observed that the potato crop recovered only 4% of the applied phosphorus, which makes potato a crop highly sensible to the factors associated to the absorption of that nutrient. Both the soil nutrient concentration and the plant metabolic activity affect the nutrient absorption (Marschner, 1995), which relies on the oxygen supply to the generation of energy and to support the cell activity. The adequate oxygen supply relies on the establishment of soil macropores and water stable aggregates formation. Crop rotation affects the soil physical properties by the roots action, which releases organic material to the system. Silva & Mielniczuk (1998) observed that grasses affect positively to the soil aggregation, which is due to its high root production. Towards the root system production capacity, Brachiaria brizantha cv. Marandu is enhanced among the grasses (Kanno et al, 1999), while *Panicum maximum* cv. Tanzania presents the highest shoot biomass production. Considering the importance of the soil physical, chemical and biological attributes, this research was conducted to evaluate the influence of the previus crop, associated to different phosphorus levels, on the potato crop productivity.

# **Material and methods**

The experiment was a fatorial (3x4) scheme arranged in complete randomized blocks with six replicates. The treatments were combinations among three grasses (*Brachiaria brizantha* cv Marandu, *Panicum maximum* cv Tanzania and *Zea mays* L.), cultivated prior to potato cv. Atlantic, and four P levels (100, 400, 700 e 1000 kg . ha<sup>-1</sup>  $P_2O_5$ ). On November, 2007, just before the sowing of the grasses, a deep soil tillage was proceeded up to 80 cm. On April, 2008, the grasses were cut and milled and the straw remained on the soil surface for 30 days. Then, the soil was prepared and, on May, the potato crop was installed. On September, productivity was evaluated by F and Duncan tests at 5%.

# **Results and discussion**

The residual phosphorus affected significantly the potato productivity and the highest yield was obtained with 700 kg.ha<sup>-1</sup>  $P_2O_5$  (Figure 1).

Figure 1. Potato yield affected by different levels of phosphorus applied to soil. Different letters mean significant difference (Duncan test at 5%) among treatments. Variation coefficient: 26.3%



The P absorption efficiency ranges among the potato varieties, due to the plant physiology and its interaction with the environment (Sattelmacher et al, 1990; Harris, 1992). The level of P which proportioned the highest potato cv. Atlantic yield (700 kg.ha<sup>-1</sup> P<sub>2</sub>O<sub>5</sub>) resulted in a leaf P content of 4,8 g of P per kg of dry leaf tissue (Figure 2), which is higher than the optimum P leaf content determined to the potato cv. Aquila, which ranged from 2,5 to 2,7 g.kg<sup>-1</sup> (Gallo et al, 1970); it is, also, higher than the standard value of 3,5 g.kg<sup>-1</sup> suggested by Malavolta (2006). It indicates the potato cv. Atlantic is highly dependent on phosphorus. The P<sub>2</sub>O<sub>5</sub> level of 1.000 kg.ha<sup>-1</sup> led to a reduction of the leaf zinc content in comparison to the P level of 700 kg.ha<sup>-1</sup> (Figure 2), possibly due to the competition between those nutrients during the uptake process (Malavolta, 2006), which may have let to the observed decrease in potato yield.





The effect of the grasses cultivated prior to the potato crop were not detected but, nevertheless, Bishop & Grimes (1978) obtained increases in the potato productivity due to the improvement of soil macroporosity, achieved by the adoption of deep tillage and zero traffic systems. In this work, the cultivation of grasses with different characteristics of shoot and root production proportioned distinct effects to the soil macroporosity (Rocha et al, 2008), but, instead of it, the potato yield was not affected.

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# References

- BISHOP, JC; GRIMES, DW. 1978. Precision tillage effects on potato root and tuber production. American Potato Journal 55, 65-71.
- DECHASSA, N; SCHENK, MK; CLAASEN, N; STEINGROBE, B. 2003. Phosphorus efficiency of cabbage (*Brassica oleraceae* L. var. capitata), carrot (*Daucus carota* L.), and potato (*Solanum tuberosum* L.). Plant and Soil 250, 215-224.
- GALLO, JR; HIROCE, R; NÓBREGA, A. 1970. Nutrição nitrogenada, fosfatada e potássica da batatinha, revelada pela análise química foliar. Bragantia 29, 1-5.
- KANNO, T; MACEDO, MCM; EUCLIDES, VBP; BONO, JAM; SANTOS JÚNIOR, JDG; CORRÊA, RM; BERETTA, LG. 1999. Root biomass of five tropical grass pastures under continuous grazing in Brasilian Savannas. Grassland Science 45, 9-14.
- MALAVOLTA, E. 2006. Manual de nutrição mineral de plantas. São Paulo: Editora Agronômica Ceres.
- MARSCHNER, H. 1995. Mineral Nutritional of higher plants. London: Academic Press.
- SATTELMACHER, B; KLOTZ, F; MARSCHNER, H. 1990. Influence of the nitrogen level on root growth and morphology of two potato varieties differing in nitrogen acquisition. Plant and Soil 123, 131-137.
- HARRIS, P. 1992. Mineral nutrition. In: HARRIS, P. ed. The potato crop. London: Chapman & Hall.
- PURSGLOVE, JD; SANDERS, FE. 1981. The growth and phosphorus economy of the early potato (*Solanum tuberosum*). Communication in Soil Science and Plant Analysis 12, 1105-1121.
- ROCHA, MG; RAGASSI, CF; FAVARIN, JL. 2008. Macroporosidade do solo cultivado com diferentes gramíneas em sistema de preparo profundo do solo. In. Simpósio Internacional de Iniciação Científica. Piracicaba: ESALQ. Suplemento CD-ROM.
- SILVA, IF; MIELNICZUK, J. 1998. Sistemas de cultivo e características do solo afetando a estabilidade de agregados. Revista Brasileira de Ciência de Solo 22, 311-317.
- VANDER ZAAG, P; FOX, RL; DE LA PENA, RS; YOST. 1979. P nutrition of cassava, including mycorrhizal effects on P, K, S, Zn and Ca uptake. Field Crops Research. 2, 253-263.
- YAMADA, T. 1994. Uso eficiente de fertilizantes. Piracicaba: Esalq. p. 19. (palestra apresentada na disciplina de problemas e fertilidade do solo).