



Master project

Study on the differences in below- and above-ground traits of scots pines (and their relationships) according to water availability

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Table of contents

1	Abstract	2
2	Introduction	2
3	Methodology	7
3.0.1	Sampling site	7
3.1	Data collection	8
3.2	Data analysis	10
4	Results	12
4.1	Environmental influence	12
4.2	Root traits	13
4.3	Aboveground traits	18
4.4	relationships between belowground and aboveground traits	26
5	Discussion	29
5.1	Results discussion	29
5.2	Limits	31
6	Conclusion	33
7	Appendix	34
7.1	Correlations	34

1 Abstract

This study focused on the physiological changes of Scots pine trees grown in three different plots: a dry soil, an irrigated soil, and a soil irrigated for 15 years and then dry for 7 years. Differences between these three plots were studied concerning 20 below- and above-ground traits, as well as their relationships. It was found that these relationships are quite strong, especially between trees in the irrigated plot. There were clear differences between the irrigated and non-irrigated plots, and the PC Analyses gave very clear and distinct spectra. However, some results were surprising, it was found that the roots are more developed in the irrigated plot, and that the foliage is denser in the non-irrigated plots

2 Introduction

Ongoing climate change has the effect of modifying the environment in which trees grow, by increasing the temperature and especially reducing the quantity of soil water. Heat waves or periods of drought are more frequent and intense, and cause deterioration of forest stands.

In this context, in order to understand the risks that droughts pose to trees, it is interesting to define precisely what the death of a tree means physiologically. According to a study published in 2011 which was the first to give a clear definition¹, “Death is defined as thermodynamic equilibrium between the organism and the environment, in which plants no longer have energy gradients to drive metabolism or re-generate”. Applying this to trees under drought conditions, the thermodynamic equilibrium with plant's environment can be summarized by the transport of water from the soil to the atmosphere². Therefore the tree dies when it is no longer possible for the xylem to transport the elements that are essential for the functioning of the organs and for regeneration, i.e. water and nutrients.³ This circulation is driven by the transpiration of the leaves (or needles) which creates a gradient in water potential between the roots and the top of the tree, through a continuum of conducting tissue⁴.

The proper functioning of this system, therefore, depends on the water storage capacity of the plant $Wp = U - ET + S$, where U is the water uptake from the soil to the top, ET is the evapotranspiration, and S is the storage (which includes water in the cells and all water in the duct xylems and phloem).⁵ Under normal conditions, the water storage capacity increases over time. A drought becomes dangerous if it reduces the Wp , which means that the water outflows are greater than the inflows, and that the tree will have to draw on its own reserves in order not to die⁶. If the tree falls below this sustainable level of water reserves, transport and coordination of organs can be disrupted because of many physiological processes, such as the reduction of hydraulic capacitance and water storage

¹N.G. McDowell. “Mechanisms linking drought, hydraulics, carbon metabolism, and vegetation mortality”. In: *Plant Physiol.* 155 (2011), pp. 1051–1059, p. 2.

²William M. Hammond. “Dying on time: traits influencing the dynamics of tree mortality risk from drought”. In: *Tree Physiology* 39 (2022), pp. 906–909, p. 2.

³Hammond, “Dying on time: traits influencing the dynamics of tree mortality risk from drought”, p. 2.

⁴H. Rennenberg. “Physiological Responses of Forest Trees to Heat and Drought”. In: *Plant Biol.* 8 (2006), pp. 556–571, p. 2.

⁵William R. L. Anderegg. “Linking definitions, mechanisms, and modeling of drought-induced tree death”. In: *Trends in Plant Science* 17.12 (2022), pp. 693–700, p. 2.

⁶William R. L. Anderegg, “Linking definitions, mechanisms, and modeling of drought-induced tree death”, p. 2.

capacity, cavitation in xylem vessels, xylem conduit collapse, phloem transport deficiency, rupture of xylem– phloem coupling, inability to catch nutrients from the soil, etc⁷.

In order to avoid such disturbances, trees undergoing drought develop mechanisms to keep the water storage capacity as high as possible. They reduce the water loss caused by evapotranspiration. This is done by closing the stomata (more precisely, reducing their conductance) , so that the water present does not reach the gaseous state and is released into the atmosphere. By reducing the stomatal conductance, CO₂ inputs are also reduced, inhibiting photosynthetic activity. The reaction of photosynthesis itself consumes water, so it is a positive feedback loop, explaining the often drastic physiological reactions.

⁸ The tree thus enters a survival phase, where its development, ensured by photosynthesis, becomes secondary. In fact, cellular respiration becomes more important in periods of drought⁹, as it allows the release of water and provides energy to the cells through the consumption of glucose (produced by photosynthesis).

During severe droughts in which photosynthesis is zero, and metabolism and mobilisation are inhibited, the amount of carbon available to maintain the water system is determined by the level of NSC (non-structural carbohydrate) at the beginning of the drought¹⁰¹¹.

Another way to maintain a good water balance is to lower the potential of the leaves, which helps the roots to better pump water out of the soil. To do this, chemical agents such as proline or other osmolytes accumulate at the top to lower the osmotic pressure of the leaves¹².

Furthermore, it is important to know the effects of repeated droughts, or sustained soil drying, not least because these are the stresses that many of Europe's forests will face in the coming decades, according to the IPCC¹³. Indeed, water availability will decrease on average, and extreme events are likely to become more frequent and intense. Physiologically, the behaviours described above are also valid after a drought acclimation. Photosynthesis is inhibited by lack of water, with the same reasons and consequences as for short drought. This is measurable by fluorescence, stomatal conductance and PSII quantum yield¹⁴. It was found in a pine forest that these parameters have lower values in plots facing drought compared to irrigated plots¹⁵.

It is especially interesting that plots that have been irrigated for a long time and then suddenly faced with droughts show more stress symptoms than plots that have always been faced with drought. This proves that the trees have acclimated. And this acclimation takes place mainly in the first years, but it continues even after 15 years¹⁶. The first adaptation concerns the leaf area. Again with

⁷Ximeng Li. “Unlocking Drought-Induced Tree Mortality: Physiological Mechanisms to Modeling”. In: *Frontiers in Plant Science* 13 (2022), p. 2.

⁸ĽUBICA DITMAROVÁ. “Physiological responses of Norway spruce (*Picea abies*) seedlings to drought stress”. In: *Tree Physiology* 30 (2009), pp. 205–213, p. 2.

⁹Rennenberg, “Physiological Responses of Forest Trees to Heat and Drought”, p. 2.

¹⁰Hammond, “Dying on time: traits influencing the dynamics of tree mortality risk from drought”, p. 2.

¹¹Li, “Unlocking Drought-Induced Tree Mortality: Physiological Mechanisms to Modeling”, p. 2.

¹²DITMAROVÁ, “Physiological responses of Norway spruce (*Picea abies*) seedlings to drought stress”, p. 2.

¹³etc Sina Löschke Komila Nabiyeva. “IPCC Sixth Assessment Report (Group II : Impacts, Adaptation and Vulnerability)”. In: (), p. 2.

¹⁴DITMAROVÁ, “Physiological responses of Norway spruce (*Picea abies*) seedlings to drought stress”, p. 2.

¹⁵Petra d’Odorico. “Drone-based physiological index reveals long-term acclimatation and drought stress responses in trees”. In: *Plant, cell environment* 44 (2021), pp. 3552–3570, p. 2.

¹⁶Arun K. Bose. “Lessons learned from a long-term irrigation experiment in a dry Scots pine forest: Impacts on traits and functioning”. In: *Ecological Monographs* 92 (2022), p. 2.

the aim of inhibiting photosynthesis, some of the leaves will die or not grow back to reduce contact between the tree and the atmosphere as much as possible to minimise evapotranspiration. Thus, the PRI of dry forests is low¹⁷, not only because of the closure of the stomata but also because of the decrease of the leaf surface. Moreover, the branches with an unfavourable water balance are the ones that will die out first because of cavitation problems, which helps to balance and improve the water balance of the rest of the tree, and contributes to the reduction of the leaf area.¹⁸

This is accompanied by a more profound change, which concerns the allocation of biomass, with emphasis on absorbing and conductive elements (roots and stem), to enhance the hydraulic conductance in the soil-leaf continuum. Another plastic and long-term response is the development of a xylem with increased resistance to drought-induced cavitation able to withstand lower water potential.¹⁹

It is important to note that the amount of water in the soil can have indirect effects on the tree through the intermediary of the rest of the ecosystem, whether by the decomposition of litter, the capacity of natural regeneration of woody species), the abundance and diversity of insects, and the amount of biomass and fungi.

20

But apart from that, studying the question of tree adaptation to drought by means of the functional spectrum approach can be interesting. For this purpose, the scientific field decided to divide the tree into several parts, each fulfilling distinct functions (leaves, trunk and branches, roots, etc.), and to study, through the associated traits, the variations in their economic functioning. And the notion of “Spectrum” comes from the fact that it has been found that certain combinations of functional traits have been more viable than others in the course of evolution, generating a continuum of trait trade-offs that can be summarised by a few axes of variation.

21

One of the most studied is the leaf economics spectrum. It was developed as a result of observations that shown correlations between several leaf-related traits²². This is the result of adaptation strategies of trees to their environment, which result in trade-offs between the benefits of a developed leaf system, i.e. carbon fixation and energy input from photosynthesis, and the costs in biomass allocation and water loss through evapotranspiration. The trade-off is related to resource management, between rapid resource acquisition and greater resource conservation. Focusing on traits, the two ends of the spectrum are represented on the one hand by structurally inexpensive but short-lived leaves with high nitrogen content and high conductivity (for fast-growing species). On the other hand by structurally expensive but long-lived leaves with low nitrogen content and low conductivity (for the more resistant species)²³. However, some research argues that the LES is

¹⁷d’Odorico, “Drone-based physiological index reveals long-term acclimatation and drought stress responses in trees”, p. 2.

¹⁸Hafiz Maherali. “Influence of climate-driven shifts in biomass allocation on water transport and storage in ponderosa pine”. In: *Oecologia* 129 (2001), pp. 481–491, p. 2.

¹⁹Nathalie Bréda. “Temperate forest trees and stands under severe drought: a review of ecophysiological responses, adaptation processes and long-term consequences”. In: *EDP Sciences* 63 (2006), pp. 625–644, p. 2.

²⁰Bose, “Lessons learned from a long-term irrigation experiment in a dry Scots pine forest: Impacts on traits and functioning”, p. 2.

²¹Sandra Diaz. “The global spectrum of plant form and function”. In: *Nature* 529 (2016), pp. 167–171, p. 1.

²²Kris R. Kramer-Walter. “Global Leaf Trait Relationships: Mass, Area, and the Leaf Economics Spectrum”. In: *Science* 340.6133 (2016), p. 1.

²³Lisa A. Donovan. “The evolution of the worldwide leaf economics spectrum”. In: *Trends in Ecology & Evolution*

not so closely related to growth form, plant functional type or biome, especially because the LES is limited in its diversity, and mono-directional, describing the management of resources that pass through the leaves.²⁴.

Another economic spectrum focuses on wood (WES). It is based on trade-offs between three parameters: resistance to embolism (Stress), conductive efficiency (competition/resource use) and mechanical strength (disturbance)²⁵. There are, therefore, three trade-offs between these parameters, which support the following relationships: a negative correlation between the resistance to embolism and conductive efficiency, a negative trade-off between conductive efficiency and mechanical strength and a positive relation between resistance to embolism and mechanical damage.²⁶ However, the validity of this economical spectrum (and others) depends on plants optimising growth and survival in all tissues through choices in allocation; this condition is more likely to be true for wood.²⁷.

Finally, the root economic spectrum is used to study the tendency of root systems to either expand to absorb soil resources as efficiently as possible or to avoid capturing too much of the biomass allocation. However, for roots, this theoretical tool is less consistent with field data than it is for leaves and wood. Unlike leaves or wood structures, roots (especially fine ones) are unconstrained in the way they are constructed: plants can construct roots with high or low SRL (specific root length) of any tissue density without this causing problems of mechanical or other strength. Several studies have also found non-linear relationships of RTD and RN (root nitrogen) with root diameter in woody species^{28,29}. Therefore the different root-related traits are less related to each other or less directly related, which makes it more difficult to predict one from the other. They are thought to be related to small but multiple influences, such as the mechanical, chemical and biological properties of the soil, in addition to their functions for the tree and their economic resource management.

Despite some non-linearities, therefore, one of the important functions of leaves, wood and roots is resource management. It is therefore thought that it may be relevant to decompartmentalise the different organs and consider them as an interconnected system (the plant) in order to study how it manages flows and allocations as a whole³⁰. A single spectrum of “fast-slow” plant economy that includes leaves, stems (wood) and roots can help explain individual ecological strategies. “Fast” refers to species that are able to move resources (water, nutrients) quickly and that have low tissue density, short tissue lifetimes and high rates of resource acquisition and flow at the organ and indi-

26.2 (2011), pp. 88–95.

²⁴Ian J. Wright. “The worldwide leaf economics spectrum”. In: *Nature* 428 (2004), pp. 821–827.

²⁵Jerome Chave. “Towards a worldwide wood economics spectrum”. In: *Ecology Letters* 12 (2009), pp. 351–366.

²⁶Hans Verbeeck. “Time for a Plant Structural Economics Spectrum”. In: *Frontiers* (2019).

²⁷Chave, “Towards a worldwide wood economics spectrum”.

²⁸Kris R. Kramer-Walter. “Root traits are multidimensional: specific root length is independent from root tissue density and the plant economic spectrum”. In: *Journal of Ecology* 104 (2016), pp. 1299–1310.

²⁹Carlos P. Carmona. “Nonlinearity of root trait relationships and the root economics spectrum”. In: *Nature Communications* 10.2203 (2019).

³⁰Carlos P. Carmona. “Fine-root traits in the global spectrum of plant form and function”. In: *Nature* 597 (2021), pp. 683–687, p. 2.

vidual level. Species with a “slow” strategy have the opposite characteristics^{31,32}.

Another way of classifying trees, called “global spectrum of plant form and function”, also based on traits, uses a different approach³³. It was obtained by a less deterministic method than the plant economic spectrum, more stochastic. From important traits trying to describe the variety of plant organs, one tries to find out what trends are obtained. By means of a PSA analysis, this results in a plan whose two axes are the size of whole plants and organs on the one hand, and the construction costs of the biomass growth on the other. It is more or less the same axes as for the economic spectrum, which proves that the economic approach is relevant. The first of these two axes (PC1) is mainly composed of plant height, stem density (SSD), seed mass (SM) and leaf mass per area (LMA), and the negative of leaf nitrogen per mass (Nmass). The second axis (PC2) is composed of Leaf area (LA), leaf nitrogen per mass (Nmass), diaspore mass and the negative of leaf mass per area (LMA). But unlike the spectra mentioned above, the GSPFF is occupied in two dimensions, with some parts denser than others, each corresponding to different taxonomic types, but without any trend emerging as to the relationship between the two axes. This is mainly due to the huge diversity of species used, which are supposed to represent all woody and non-woody plants.

This report will focus on a Scots pine forest. In the field studied by the GSPFF, this species belongs to the gymnosperm (monophyletic group which contains the conifers). As can be seen from the figures, they have a high PC1 and a low PC2 compared to the rest of the plants. If we compare them to the woody-angiosperm, they have the same PC1 but a lower PC2. This means roughly that softwoods have a lower resource cost for an equivalent development. Therefore they are by definition better adapted to more complicated environmental conditions.

Numerous studies have already compared the spaces occupied by a set species in the different functional spectra. It would be interesting to put the comparison between the functional areas of the same species (here, Scots pine), but growing in different environments (dry or irrigated soils over the long term).

Therefore, this report will focus on the differences in below- and above-ground traits of scots pines (and their relationships) according to water availability. The below-ground traits studied here are root length, diameter, nitrogen concentration, and tissue density, as well as root density in the soil, and the proportion of roots at a depth of less than 50 cm. For the above-ground traits, we will be interested in the tree height, the crown radius, the stem density, the seed mass, the leaf area index, the specific leaf area and the leaf mass per area. That would allow to understand better what underlies the development of these two parts of the tree, which a priori have fairly distinct functions, but actively participate in the survival of the tree in times of drought by adapting their functioning.

Since lack of water should lead to foliage atrophy and root development, the relationship between below and aboveground traits should diverge in dry versus irrigated soils. It will also be interesting to find out which root traits are strongly correlated with above-ground traits, in each plot. Furthermore, we will check that the results correspond to the state of the art described in this introduction for each of the traits. Finally we will try to determine if the effects of drought are determining on

³¹J Peter B. Reich. “The world-wide ‘fast-slow’ plant economics spectrum: a traits manifesto”. In: *Journal of ecology* 102 (2014), pp. 275–301.

³²Carmona, “Fine-root traits in the global spectrum of plant form and function”, p. 2.

³³Diaz, “The global spectrum of plant form and function”.

the studied traits on the short, medium or long term.

3 Methodology

3.0.1 Sampling site

The data useful for this study were collected in Pfywwald. This is a Scots pine forest located in the Rhone valley between Brig and Sion, in the canton of Valais. It is one of the driest valleys in the Alps. The mean annual temperature is 11,03°C (20,4°C for June–August) and mean annual precipitation is 563mm (172mm for June–August) for the period 2003–2021 (data from the MeteoSwiss station Sion; MeteoSwiss, 2022). The soil volumetric water content is low, fluctuating from 0.11 to 0.47 with an average of 0.27 (based on the data from 2003 to 2014). According to the pedotransfer function of Puhlmann et al. (2009), the soil is characterized by a low available water-holding capacity of 135mm until 0.8 m rooting depth³⁴. Since 2003, WSL researchers have been using part of this forest to study physiological changes in pines as a function of soil irrigation.

For this, three different areas are studied, in a part of Pfywwald dedicated to the WSL research center. The Scots pines are 100 years old with a density of 730 individuals/ha. The first area is the control plot : here, there is no treatment, and the trees that grow there undergo the climatic conditions described above. It is located in the plot 6 of the area dedicated to WSL. The second area was an irrigated plot (located west of plot number 6). The treatment of 2022 started on 11 May. The amounts of precipitation and irrigation are shown in the Figure 1 (until July 19th). The total amount of water added each year by irrigation varies, but it approximately double the average annual precipitation (600mm/year).

The third zone was also watered from 2003 to 2015, but the treatment was stopped 7 years ago. It is located east of the irrigated plot (in plot number 5).

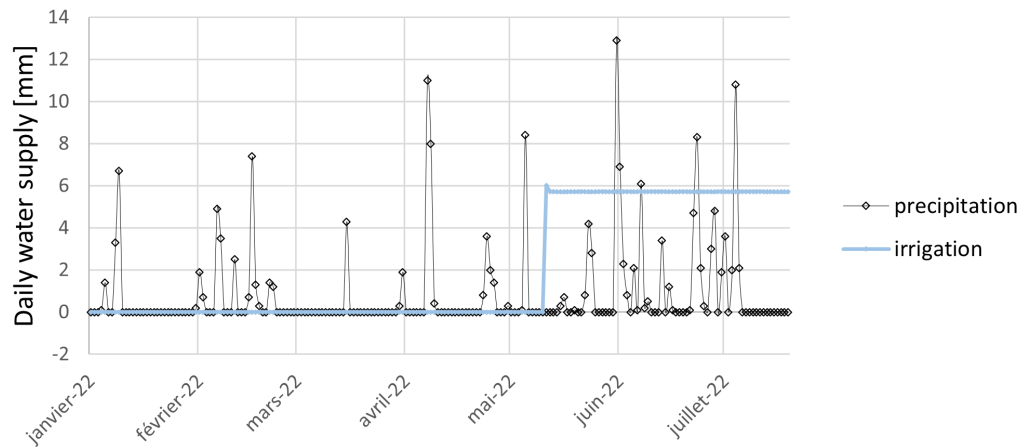


Figure 1: Daily precipitations and irrigation from 1 January to 19 July 2022 [mm/d]

Since this system has been in place for 19 years now, it is relevant to use it to study the

³⁴Bose, “Lessons learned from a long-term irrigation experiment in a dry Scots pine forest: Impacts on traits and functioning”.

physiological differences between trees benefiting from an advantageous water regime and trees facing a dry environment over the long-term (as a model of a future climate).

3.1 Data collection

The data collection has been framed by the protocol TraitDivNet. It is a worldwide project aiming to obtain information on selected key traits for a significant portion of the world's ecosystems to reveal the general mechanisms determining functional diversity across and within ecosystems, by the aggregation of tree traits data in every type of ecosystem.

The below- and above-ground traits are summarized in the table below :

	Traits	Unit
Below-ground traits	Root length mean	cm
	Root diameter mean	cm
	Root length/diameter ratio	-
	Root nitrogen concentration	%
	Root tissue density	g/ml
	Soil root density	mg/cm ³
	Root mass ratio over 50cm	-
Above-ground traits	Tree height	m
	Growth of vegetativ height	m/y
	Crown radius	m
	Growth of crown radius	m/y
	Stem density	t/m ³
	Needles dry mass (per leaf)	g
	Leaf area index (LAI)	-
	Needles nitrogen concentration	%
	Seed mass (2016)	g
	Specific leaf area	cm ²
	Needles length mean	cm
	Density needles (on the branch)	-
	Leaf mass per area (LMA)	g/cm

TraitDivNet was helpful in selecting the different studied traits. It also served as a protocol for collecting samples in the field.

The data for tree height (and its growth), crown radius (and its growth too), specific stem density and seed mass has been retrieved from pre-existing databases, as their sampling were too long or complex. The tree height and crown radius were not all obtained by direct measurements but by calculation from other measurements. These two data files contain values for several years on each tree (2002, 2009, 2014 and 2019), which makes it possible to find the growth of tree height and crown radius by differential calculation. Crown densities were only measured for a few trees, so the value used in this study is an average of the trees in the plots concerned. The soil root concentration was sampled this summer by a member of the WSL who passed it on to me. He took

volumes of soil at several depths (100 cm³ each time), and weighed the roots contained in these volumes

However, not everything was done exactly as prescribed by Traitdivnet. First of all, because of the subject of the study and the composition of the forest, we only took our samples from Scots pines (and not from the five most abundant species as told by Traitdivnet).

Before starting the sample collection campaigns, we had to select trees. As explained before, we first selected plots with different irrigation regimes, irrigated, stop and control, respectively plots 6 west, 6 east and 5. Then in each of these three areas, we selected three or four trees (5 at the beginning, but some trees were removed from the selection), meeting certain criteria: they had to be Scots pines, tall enough to reach direct sunlight (to avoid differences in results due to sun obstruction) and placed close to the scaffolding (to facilitate the collection of samples of above ground traits).

For traits related to leaves (needles for Scots pines), samples were collected from the top of the tree in the part exposed to direct sunlight for the reasons mentioned earlier. As TraitDivNet recommends, we took a minimum of 3 bunches of thorns (or 4cm²) per tree, sometimes more, trying to select relatively young but fully expanded and hardened needles that don't show signs of pathogen or herbivore attacks.

Fine roots have to be traced from the stem of their parent plant in order to assign root traits to individual plants and avoid taking roots from other species. For this, it is follow the primary roots, and cut the secondary ones to which several fine ones are attached.

Once these samples were collected, some of them required subsequent analyzes in order to calculate the various parameters. This is the case of needles and roots, whose nitrogen levels have been calculated by a CHN analysis, thanks to which we can know elemental concentrations in a given sample with accuracy and precision. For the length of fine roots and needles (as well as their specific area), we scanned the samples and used the Pixsat software.

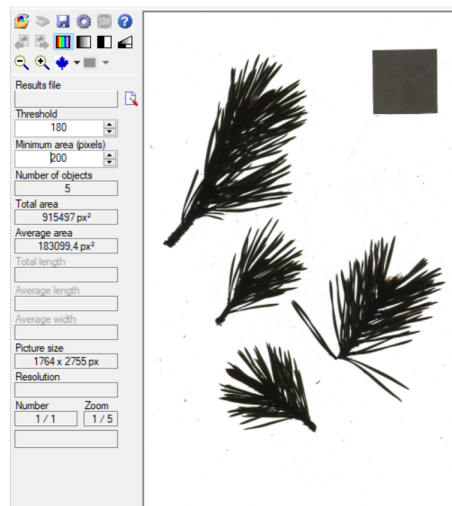


Figure 2: specific leaf area calculation, with the software “Pixstat”

The leaf area has been calculated separately. We took emisspheric pictures in the different plots, then processed and analyzed them using a software called Hemisfer. It makes a separation between the pixels corresponding to the sky or the foliage (and branches); then it calculates the LAI, with Lang's method³⁵, which does not take into account the angle of the leaf (so it corresponds well to pine needles). Knowing that the irrigated and stop plots are both on plot number 6, their LAI has been calculated by taking the LAI of the three photos of plot 6, and putting weights according to their location (and the proportion of each image occupied by the irrigated area and the stop area)

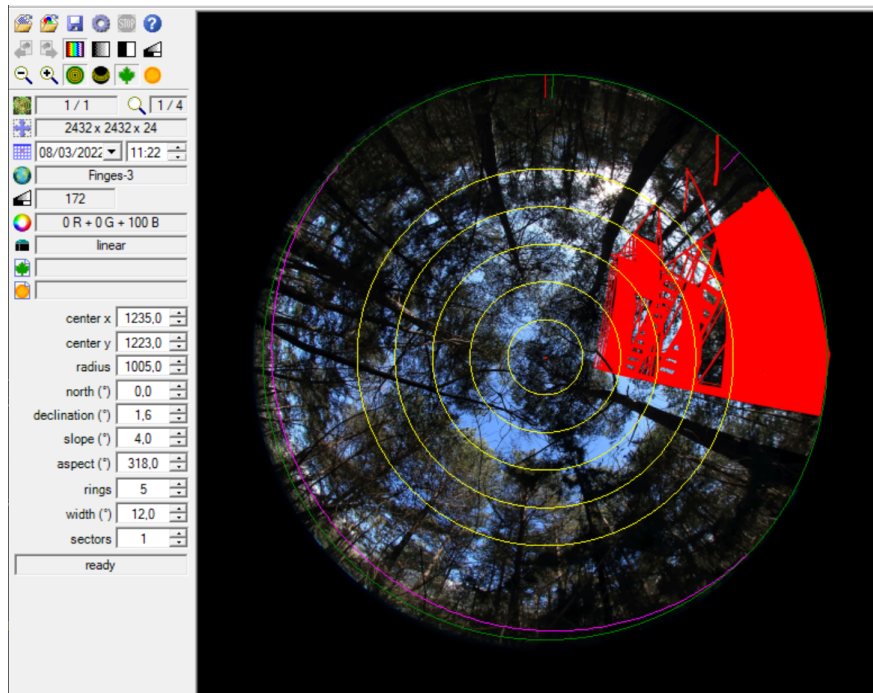


Figure 3: LAI calculation by the software “Emisfer” (the red part is highlighted scaffolds)

However, in order to explain any indirect factors of differentiation between the plots, the biodiversity of each of them was apprehended by observation and census. In addition, as Traitdivnet recommends, we probed the soil of the three plots in order to know their physico-chemical structure. For this, it was necessary to dig three profiles down to the source rock in order to be able to observe the soil as a whole.

3.2 Data analysis

To analyze all the data collected, the first step is a comparison between the three plots for each of the parameters, in order to see certain trends emerge and get a general idea of the differences. The visual representation can be made for each tree, but it will be interesting to calculate the average

³⁵A.R.G.Lang. “Simplified estimate of leaf area index from transmittance of the sun’s beam”. In: (December 1987).

and standard deviation of the dry and irrigated plots, and to compare them with the third.

The second step is to establish the correlations between the different traits (or groups of traits) for each tree and in each plot to clearly visualize the pine's adaptation in relation to water availability.

It will also be interesting to place the trees (and the plots) in a functional space such as GSPFF, if this is statistically relevant, in order to clearly visualize the adaptation of the pine in relation to the availability of water. To do this, a PCA (principal component analysis) has been done with the software R studio

4 Results

4.1 Environmental influence

As mentioned in the introduction, it is important to study the ecosystem contexts in which drought occurs, in order to identify some of the influences on outcomes and indirect effects of drought

-Vegetation

In the control plot the vegetation is quite dense, with a profusion of bushes and robust shrubs. It comprises alisiers, viburnum, alders and privet, all between 1 and 3 metres. As for the trees, there are oaks and boudots, the tallest being 2.5m and 5m, respectively. This plot therefore has three distinct levels of developed vegetation: the dominant trees (Scots pine), the dominant trees (oaks, boudots), and the undergrowth (bushes and shrubs, and rare seedlings).

The vegetation of the irrigated plot is a little denser than in the control plot, especially because there are many young Scots pine trees, distributed in patches. In addition to this, there is a great biodiversity, since all the species of the control plot are present (an abundance of oaks, birches, trunks, viburnum, etc.), but with the addition of young shoots of honeysuckle, wild cherry and elm, as well as tall hawthorns. The plants are generally healthy, much straighter than in the other plot, and some of the shrubs are reaching a larger size. It is also interesting to note the presence of grass on the ground (*Equisetum sylvaticum*, an acidophilous plant like pine)

In the stop plot, the vegetation is totally atrophied, very sparse, with no plants over 2m (and generally under 1m). These are not young seedlings, however, but rather small, stunted shrubs, such as honeysuckle, alizier, viburnum, barberry, privet and juniper. On the other hand, there are no pine seedlings.

- Soils :

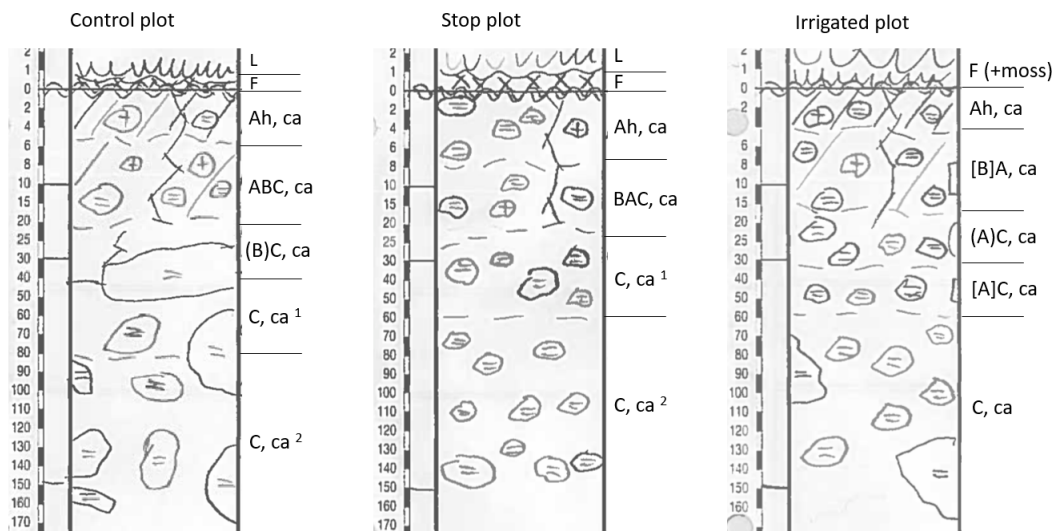


Figure 4: Soils horizons

As shown on the Figure 4, the irrigated soil is the only one of the three that does not have an L-horizon (unprocessed organic debris), which is a sign of slow decomposition by microorganisms. This corroborates the intuition of the previous section about the higher microbial activity in the irrigated soil compared to the other two.

The Ah horizons represent the topsoil, mainly composed of organic matter (humus). In these three soils, it contains more than 5% CaCO_3 (and this is the case for all horizons of the three soils). The C horizons are mainly made up of materials from the weathering of the parent rock. The irrigated soil has less Ah and C horizons than the other but there is a deeper and richer intermediate zone.

4.2 Root traits

1. Root diameter and length

The fine roots diameter and length are often very positively correlated; that is why they are represented together. We can see that the diameters are, on average, higher in the irrigated plot, with 0,14 (and lower in the control plot, with 0,11cm), which may mean that the irrigated trees have fewer very fine roots (cf Figure 5). Concerning the length of these fine roots, it is in the stop plot that the roots are the shortest : 23,6 cm on average, compared to 25,3 for the control plot and 28,7 for the irrigated one (cf Figure 6).

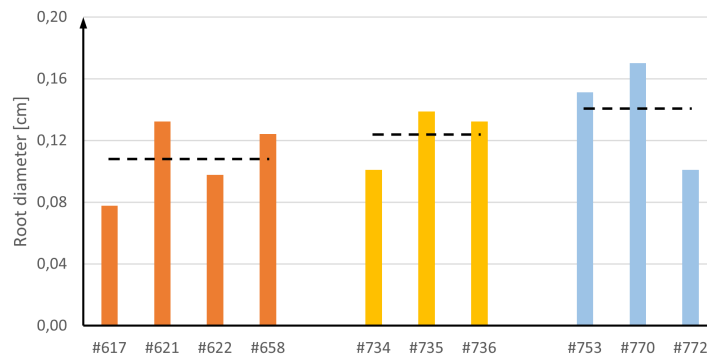


Figure 5: Fine roots diameter (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

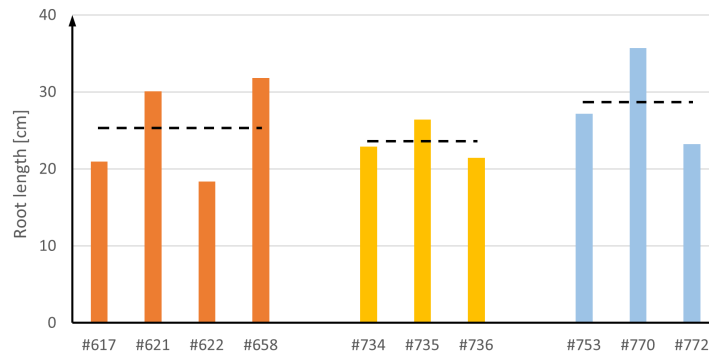


Figure 6: Fine roots length (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

As shown in the Figure 7, the roots of the irrigated plot follow the global relationship, the roots of the control plot are above and those of the stop plot are below. This means that for the same diameter, the fine roots of the stop plot are shorter than those of the irrigated plot, and the roots of the control plot are longer.

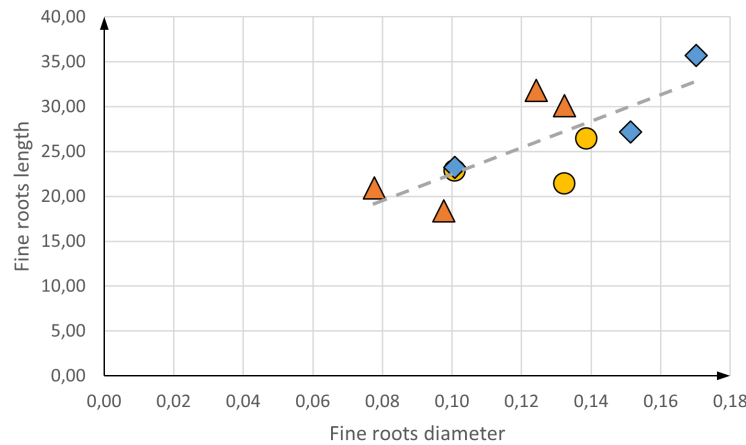


Figure 7: Relationship between diameter and length of fine roots (orange triangles = control plot individuals; yellow circles = stop plot; blue squares = irrigated plot; dotted line: trend curve)

The Figure 7 shows an almost linear relationship between the diameter and length of the fine roots. Therefore, it is interesting to calculate the ratio between these two parameters for each tree. As shown on the Figure 8, the roots of the control plot have the largest roots for a given diameter (ratio = 234.1), followed by the roots of the irrigated plot (ratio = 202.1), then those of the stop plot (187.8).

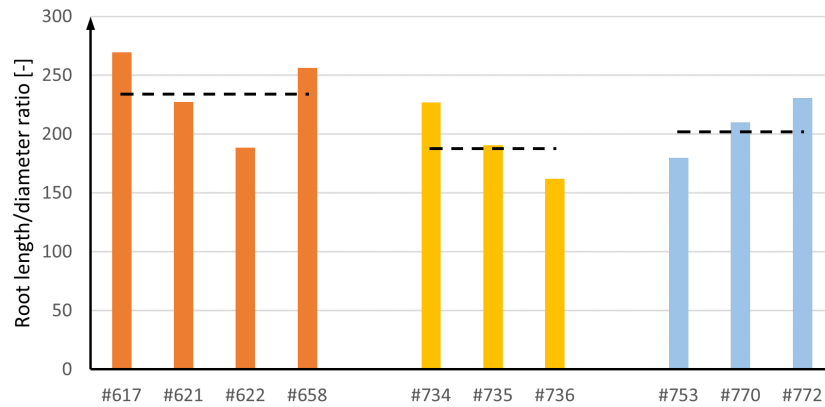


Figure 8: Fine roots length/diameter ratio [-] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

2. Fine root tissue density

The average density of fine roots tissues is identical for the stop plot and the control plot (at 0.65g/ml), below the irrigated roots which are at 0.75g/ml (cf Figure ??). Here, the variation between different individuals is low, especially because this trait is less dependent on chance or selection bias than diameter and length (when taken separately).

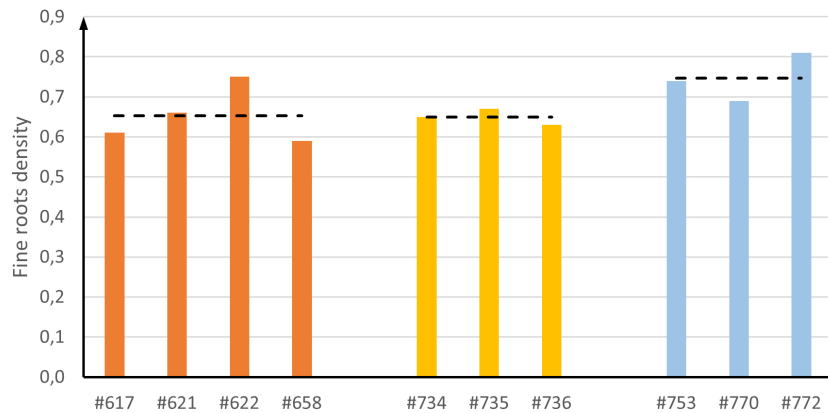


Figure 9: Fine roots density [g/ml] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

3. Soil root density

Soil root density expresses the mass of root present in a given volume of soil at several depths (cf Figure 10). For each of the three results, the distribution of root density according to depths is not necessarily the same throughout the plot, depending on the soil at the sampling location. But on the sample taken for this study, it can be seen that except for the irrigated

plot, the vast majority of roots are in the upper part of the soil (beyond 10cm, and 50cm quite clearly). Taking the totals over the whole depth, the irrigated plot is the densest in roots, with $22,5 \text{ mg/cm}^3$, the stop plot is at $19,7 \text{ mg/cm}^3$ and the control one at $9,2 \text{ mg/cm}^3$.

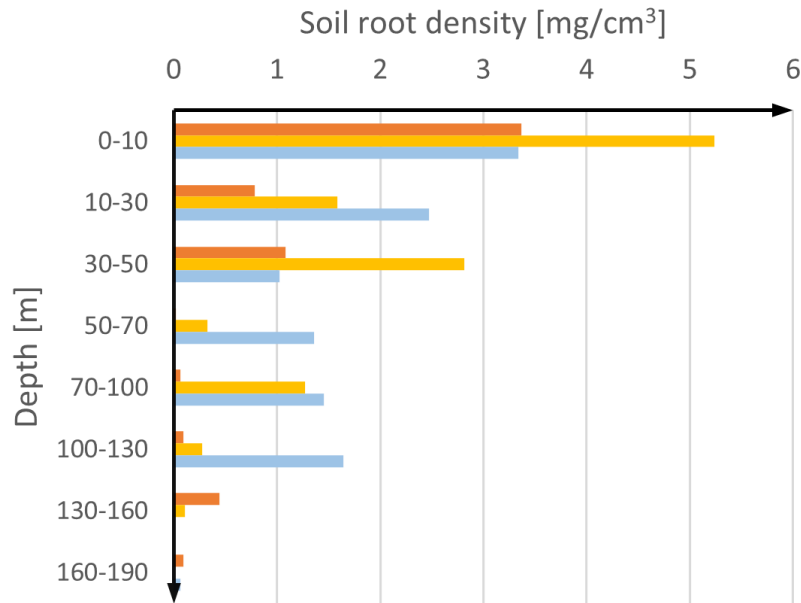


Figure 10: Soil root density, per depth intervalls [mg/cm^3] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals)

4. Fine root nitrogen content

The stop plot is the one where the fine roots have the highest amount of nitrogen, with 0,78% on average. Then, the control plot's fine roots are at 0,68% and 0,57% for the irrigated ones (cf 11). The variations between the different individuals are quite unequal between the different plots, with relative standard deviations of around 10% for the stop and control plots but almost 25% for the irrigated plot.

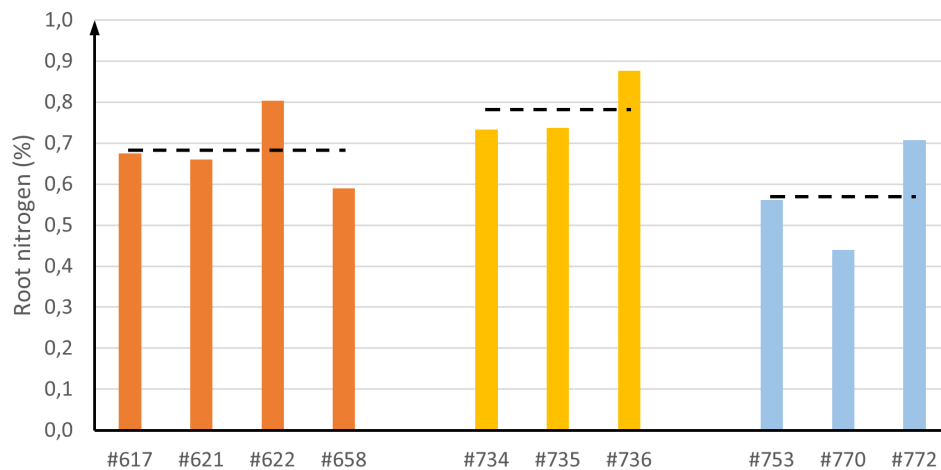


Figure 11: Fine roots nitrogen content [%] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

5. Below ground trait relationships

The below-ground traits are considered multidimensional, but the results still show links between the different traits, in addition to those already observed between root diameter and root length. To summarize these links, the proportion of correlation coefficients of all the above ground traits between them whose absolute value is greater than 0.5 (indicating that the links are significant), if we consider the three plots together, is 33%; it is 60% for the control plot, 50% for the stop plot and 0.70% for the irrigated plot (cf appendix figures).

Some of these links are more notable than the others. For example, root nitrogen content (which is lowest in the irrigated plot) is negatively correlated with fine root length in all three plots; soil root density is positively correlated with root tissue density, which makes sense, but negatively correlated with root length/diameter ratio, as well as with the “proportion of roots at a depth of less than 50 cm”. There are many other correlations, but they do not appear significantly in our data. However, some other relationships are paradoxical, such as that of root tissue density with nitrogen content and length/diameter ratio; here, depending on the plots, the relationships are completely reversed, going from a coefficient close to 1 to close to -1. We will discuss these phenomena in another section.

Overall, it can be said that the roots of the non-irrigated plots are less developed than those of the irrigated plot; they are a bit thinner and slightly shorter. Although they contain more nitrogen, their tissues are often less dense, and it is less abundant close to the surface but mostly at depth.

This is exactly what shows the PCA spectra (Figure 12). The spectra of the control and irrigated plots are almost symmetrical, along an axis slightly skewed to the PC1. The spectrum of the irrigated plot is far from that of the control plot mainly due to a higher density of root. Traits related to this (soil root density, root tissue density, Root mass ratio over 50cm) are the ones that will vary the least between individuals of each of the two plots, unlike all

the others. The stop plot is placed between the two others, but more reduced in its length on the side of the high nitrogen concentration

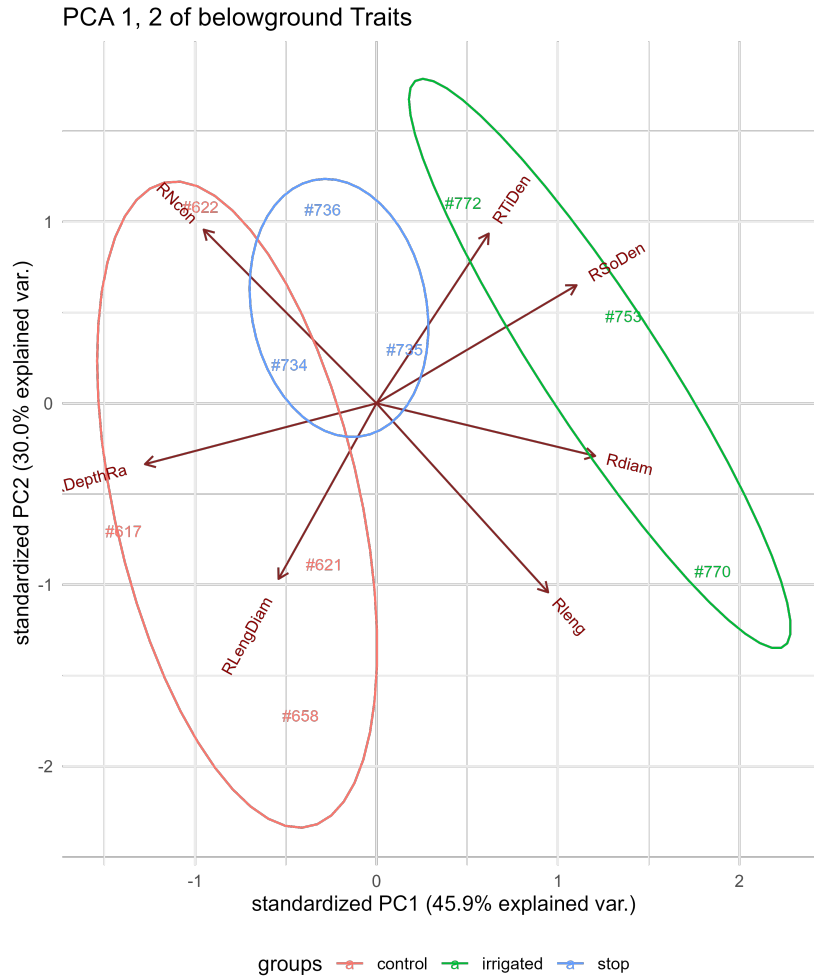


Figure 12: below-ground traits spectrum, from PCA
(RTiden = fine roots tissue density ; RSoden = roots soil density ; Rdiam = fine roots diameter ; Rleng = fine roots length ; RLenDiam = fine roots length/diameter ratio ; RNcon = fine roots nitrogen concentration)

4.3 Aboveground traits

1. Leaf area index

Due to the method used, the leaf area index cannot be individualised. It was calculated to be 1.8 in plot 5, and about 1.94 in the area chosen as control plot (which is denser in Scots pine than the rest). The results obtained by hemisfer in plot number 6 gave an LAI of 2.39 for the stop plot, and 2.57 for the irrigated plot.

2. Needles nitrogen content

The results for needles nitrogen content are quite similar. It averages 1.29% for the control plot, 1.26% for the stop plot and 1.17% for the irrigated plot (cf Figure 13). The variation between individuals within each plot is small, with relative standard deviations of between 3% and 7%.

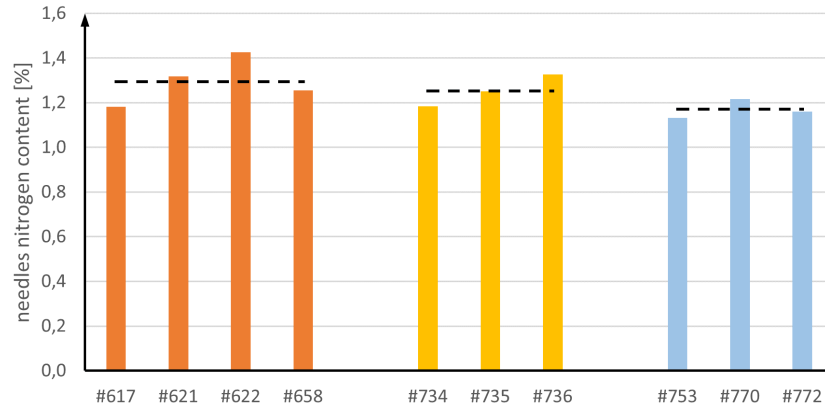


Figure 13: Nitrogen content [%] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

3. Specific leaf area

The so-called “leaf area” here is the area of the needles clusters at the top of the branches. The results are therefore expressed both in cm^2/leaf and in cm^2/cm , the latter unit taking into account the length of the branch on which the thorns are held (Figures 14 and 15). This is close to needle length, but it takes into account the density of needles along the branch. The stop plot is the one with the highest specific area with $24,9 \text{ cm}^2/\text{leaf}$, followed by the irrigated one ($20,6 \text{ cm}^2/\text{leaf}$), and finally the control one ($15,7 \text{ cm}^2/\text{leaf}$). The differences are considerably reduced for leaf area per branch length, and in this case the results of the irrigated plot are very slightly below those of the control one.

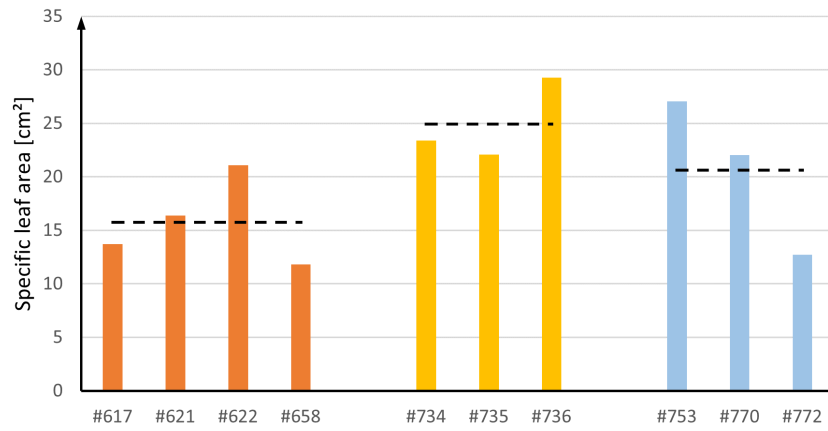


Figure 14: specific leaf area [cm^2] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

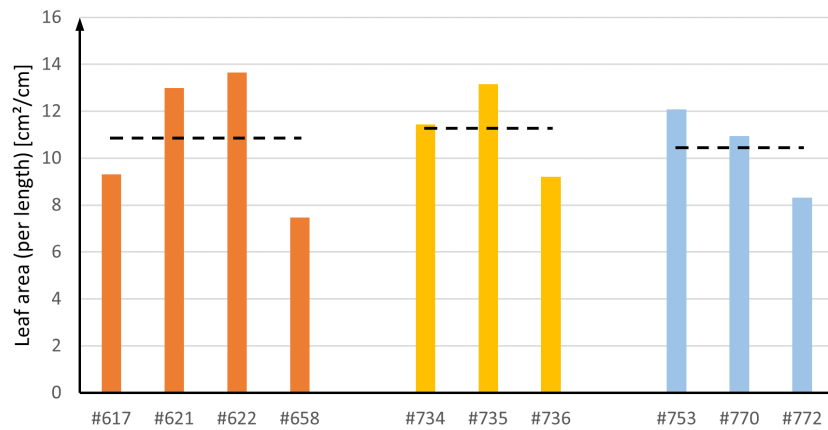


Figure 15: leaf area (per branch length) [cm^2/cm] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

4. needles length

The plot with the largest needles was the one where irrigation was stopped, with an average of 4.97 cm, followed by the irrigated plot with 4.01 cm, and finally the control plot with 3.48 (cf Figure 16). The standard deviations vary from 8% to 22% (respectively for the stop plot and the control plot).

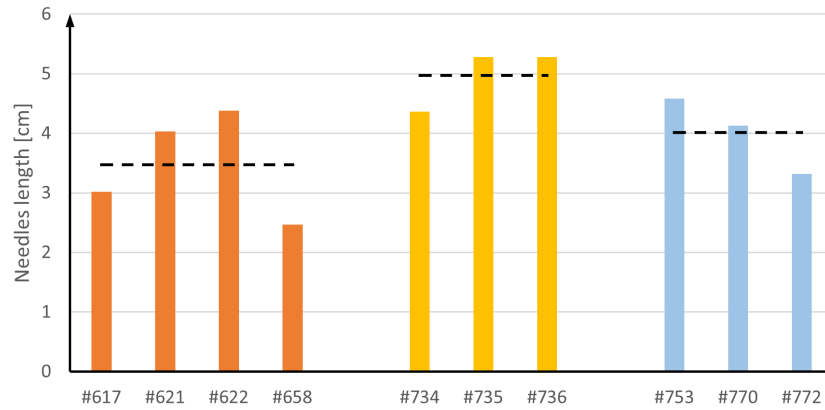


Figure 16: Needles length [cm] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

5. needles density

Needles density is the leaf area divided by the product of the average needle length and the shoot length. The results are greater than 1 because the needles are on both sides of the stem, and the area covered by the spines does not stop at the stem tip. The needles of the control plot are the densest, with a ratio of $3.11 \text{ cm}^2/\text{cm}^2$; those of the irrigated plot are at 2.60, and those of the stop at 2.29 (cf Figure 17). The relative standard deviations are very low (between 2 and 3%), except in the irrigated plot, where the standard deviation is 17%, due to a remote value.

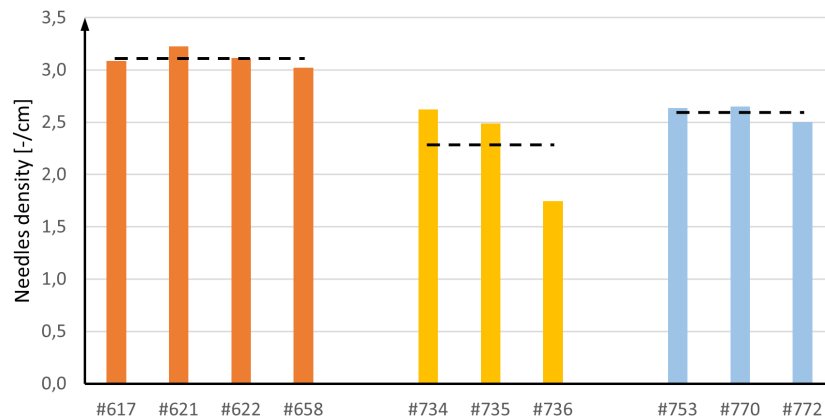


Figure 17: Needles density [-] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

6. Leaf mass per area

As shown on the figure 18, the highest leaf mass per area is for the trees of the stop plot, with $0.0758 \text{ g}/\text{cm}^2$; trees in the irrigated plot have a leaf mass per area of $0.0711 \text{ g}/\text{cm}^2$, and 0.0647

for the control plot.

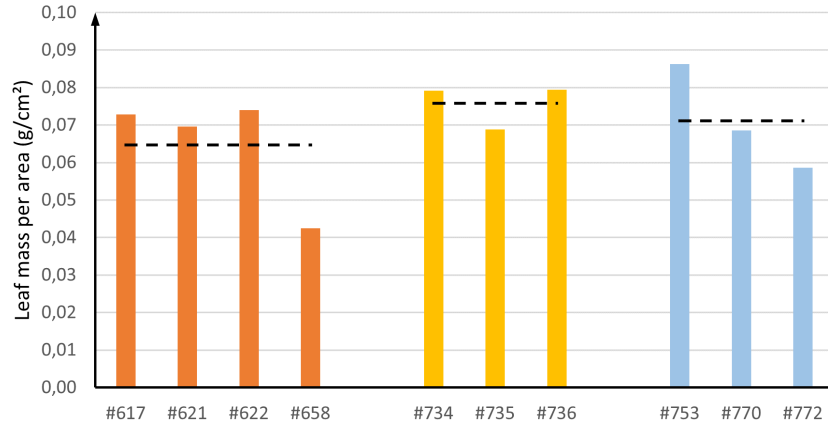


Figure 18: Leaf mass per area [g/cm^2] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

7. Tree height (and growth)

The stop plot is the one where the selected trees are the tallest with an average of 19.3m, but with the lowest growth (4.6 cm/year on average); the trees in the irrigated plot are 16.6m tall on average and grow by 13.4 cm/year; and 13.5m tall in the control plot for 8.4 cm/year. The standard deviations of the tree height are about 2m in the irrigated and control plots, and about 4m in the stop plot (cf figures 19 and 20).

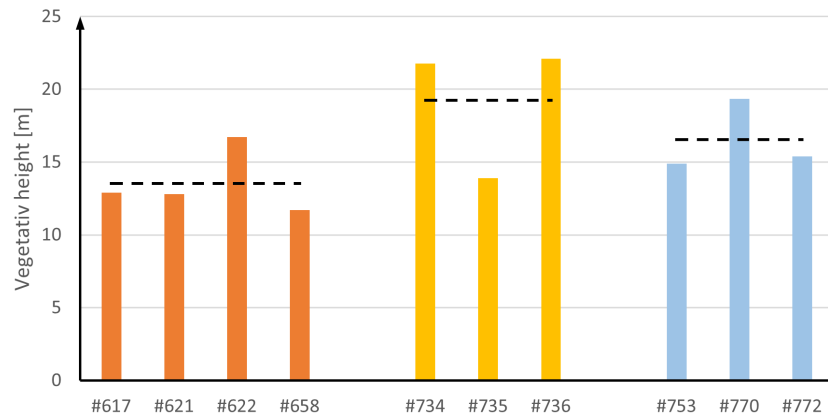


Figure 19: tree height [m] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

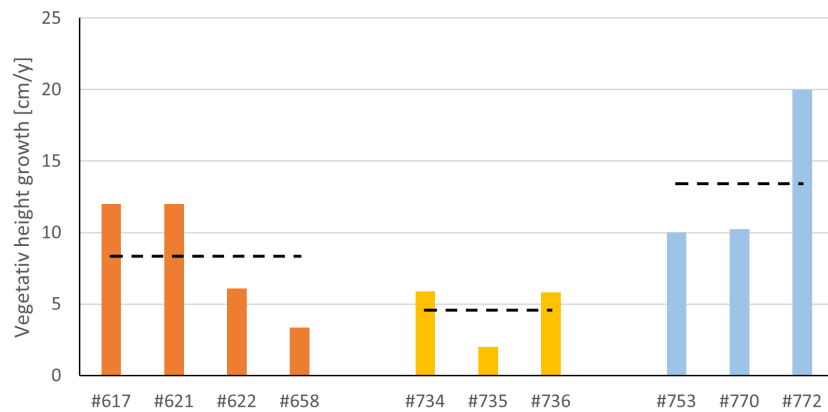


Figure 20: tree height [cm/y] (see caption to Figure 19)

8. Crown radius (and growth)

As for height, the trees of the stop plot have the largest crown radius, on average 2.7m, and grow the slowest, by 0.8 cm/year. Trees in the irrigated plots have an average crown radius of 2.5m, growing at 1.2cm/year, and those in the control plot are at 2.1m on average and 1cm/year (cf figures ?? and 22).

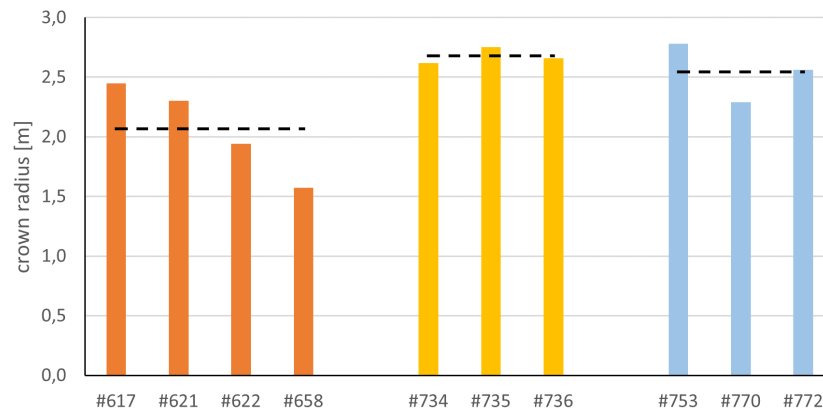


Figure 21: crown radius [m] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

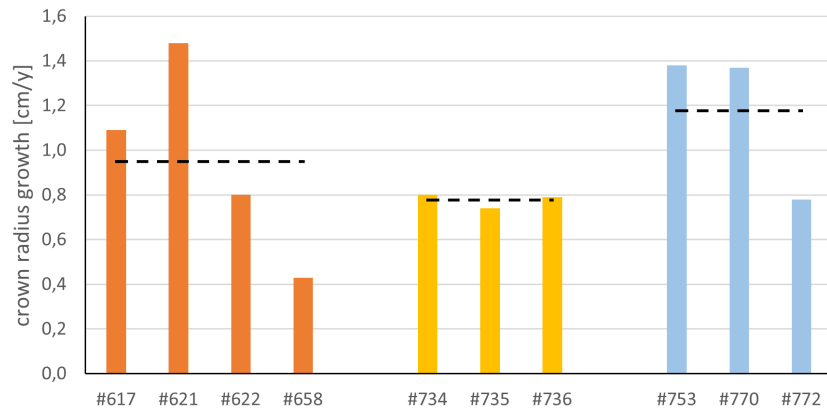


Figure 22: tree height [cm/y] (see caption to Figure ??)

9. Stem density

The stem density dataset does not include trees from the stop plots, and only a few trees from plots 5 and 6. From this, the values for the control plot and the irrigated plot can be approximated to be 0.53 t/m^3 and 0.59 t/m^3 respectively.

10. Seed mass

The seed mass is extremely variable between individuals as can be seen in the Figure 23, but it can be indicative of the stress experienced by the trees. The loblolly pines selected in the stop plot had the highest seed mass on average (in 2016), with 22.1g on average, 14.8g for the irrigated plot and 11.62g for the control one.

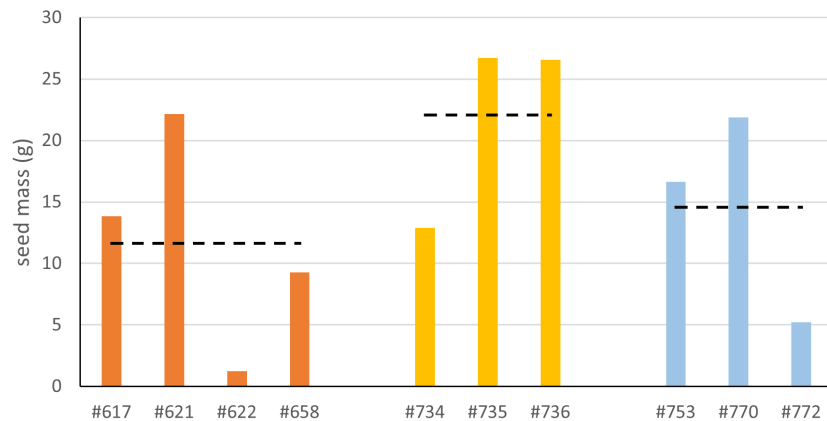


Figure 23: tree height [cm/y] (red bars : control plot ; yellow bars : stop plot individuals, blue bars: irrigated plot ; the bars represent the individuals ; dotted lines : averages of each plot)

11. Above-ground traits relationships

Above-ground traits are very clearly and quite robustly linked to each other. The proportion of correlation coefficients of all above-ground traits with each other whose absolute value is greater than 0.5 considering all trees is 42%; 58% in the control plot, 65% in the stop plot and 0.60% for the irrigated plot. So there are less differences between plots than for below-ground traits.

This can be seen in the graph of the PCA spectrum of the above ground traits (Figure 24), where the parameters are much more aggregated than in the below-ground traits. The three plots studied show fewer differences between them (in terms of links between traits), and the correlation coefficients are often closer to 1 or -1. This is for example the case between the traits that have to do with the leaves: needles dry mass per leaf, specific leaf area, needles length, and leaf mass per area (and LAI to a lesser extent) are all highly positively correlated, and negatively correlated with needles density on the branch.

Stem density (higher in the irrigated plot than in the control one) is interesting, as it is correlated with almost all other traits: positively with tree height and its growth, crown radius, and LAI (with a coefficient of 1); and negatively with nitrogen concentration and needles density on the branch. However, it has no value for the stop plot, and only one for each of the other two plots, and should therefore be interpreted with caution.

The growths of crown radius and tree height are rather correlated (positively). Let's recall that they both increase in the irrigated plot and decrease in the stop plot. More surprisingly, tree height growth are negatively correlated (or close to 0) with the leaf area-related traits mentioned above (needle dry mass per leaf, specific leaf area, needle length, and leaf mass per area), as well as with needle nitrogen concentration.

This last parameter varies little between plots ; it is negatively correlated with tree height growth as mentioned above, but also with crown radius, stem density and LAI.

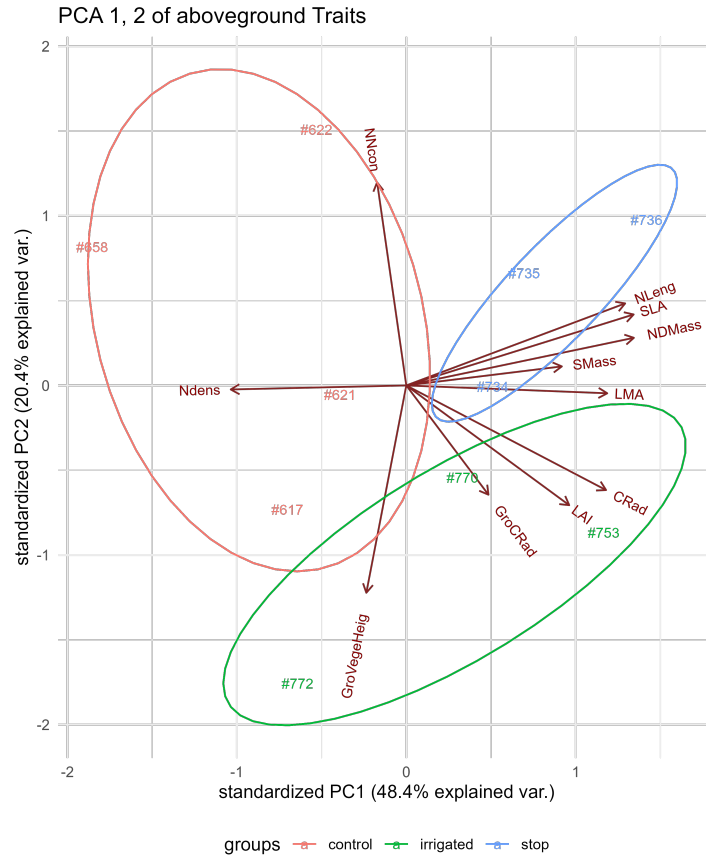


Figure 24: Above ground traits spectrum, from PCA (VegeHeig = tree height ; GroVegeHeig = tree height growth ; CRad = Crown Radius ;GroCRad = Crown Radius Growth ; NDMass = Needles dry mass (per “leaf” ; LAI = leaf area index ; NNcon = Needles nitrogen concentration; SMass= seed mass ; SLA = specific leaf area ; NLength= Needles Length ; Ndens = Needles density ; LMA = Leaf Mass per area)

4.4 relationships between belowground and aboveground traits

The correlations between above-ground traits and below-ground traits are quite high (in absolute value). They are in the same order of magnitude as for the above ground traits with each other and for the below-ground traits with each other. The correlation coefficients are shown in the figures in Appendix The proportion of correlation coefficients of all above-ground traits with an absolute value greater than 0.5 when all trees are considered is somewhat lower, at 22%, but rises to 49% when only trees in the control plot are considered, 71% for those in the stop plot and 78% in the irrigated plot. Some of these relationships are quite clear. This is the case for the relationships between the ratio root length/diameter and the traits “needle dry mass per leaf”, “specific leaf area”, and “needles length”, all negative and lower than -0.8, for all plots and for each of them. These relationships are therefore independent of the water availability. In the same scheme, LAI is also positively correlated with soil root density (close to 1) and negatively with “proportion of root above 50cm”. As for stem density and root nitrogen concentration, their correlation coefficient was

close to -1.

Many other correlations between traits have high values, but are opposite between the different plots (the coefficients don't have the same sign). This is for example the case between needles nitrogen, specific leaf area, needles length, needles density on the branch and LMA on one side, and root tissue density and root nitrogen concentration on the other side. For the relationship of these four above-ground traits with these two below-ground traits, the correlations are higher than 0.5 for the control plot, and lower than -0.5 for the irrigated plot.

The PCA spectra (Figure 25) allows us to understand them in their globality, thanks to the inclination of the eclipses. This is not exactly the case between the control plot and the irrigated plot. Overall the trees in the irrigated plot are on the side of the graph where Needles and roots nitrogen concentration are low as well as Root mass ratio over 50cm, and where root length, growth of tree height and crown radius are high. These parameters are therefore orthogonal to the spectrum of the irrigated plot and react differently in their relationships to the other traits than for the other plots. Trees within the irrigated plot will be differentiated by their specific leaf area, LMA, and root length diameter. The stop has a spectrum with a position and a direction between those of the other two. However, it is on the side of high root nitrogen concentration and needle length, and low root length/diameter ratio and tree height growth

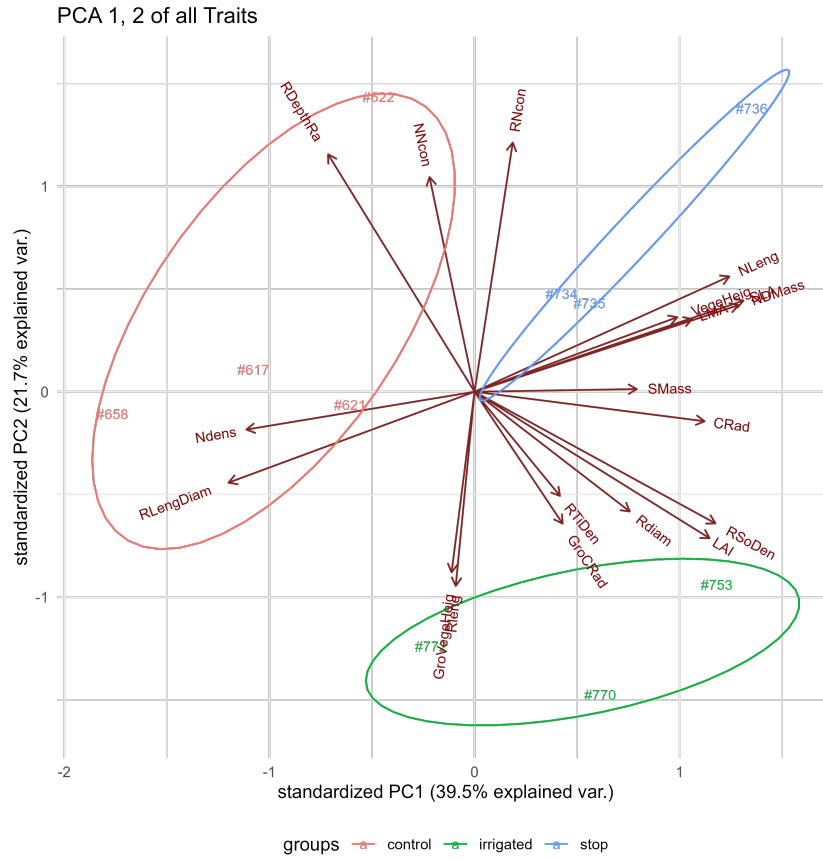


Figure 25: All traits spectrum PCA (RTiden = fine roots tissue density ; RSoDen = roots soil density ; RDepthRa = Root mass ratio over 50cm ; Rdiam = fine roots diameter ; Rleng = fine roots length ; RLengDiam = fine roots length/diameter ratio ; RNcon = fine roots nitrogen concentration ; VegeHeig = tree height ; GroVegeHeig = tree height growth ; CRad = Crown Radius ; GroCRad = Crown Radius Growth ; NDMass = Needles dry mass (per “leaf” ; LAI = leaf area index ; NNcon = Needles nitrogen concentration; SMass= seed mass ; SLA = specific leaf area ; NLength= Needles Length ; Ndens = Needles density ; LMA = Leaf Mass per area)

5 Discussion

5.1 Results discussion

The vegetation is quite different between the plots, and it has probably an indirect influence on it. The vegetation of the irrigated plot is very dense, diversified, and with a real renewal and strong, straight seedling. That of the stop plot is very sparse and stunted, and the control plot vegetation is in between. This does not seem quite intuitive, as the stop plot received more water than the control one, which should have allowed the vegetation to develop. This is probably what happened during the irrigation phase, but the plants were not adapted to the drier soil and ended up withering and dying. This gives us information on the ecosystem dynamics in each of the soils around the pine trees studied.

The developed vegetation in irrigated plot implies more concurrence for water uptake. However, this is probably negligible as the competing vegetation has a very low biomass compared to the pines (trees over 3m are very rare outside the pines). Regeneration may be inhibited by lack of water, as competing vegetation is stronger than seedling pines for water uptake, but this does not concern the mature pines studied in this study. The information that this observation of biodiversity gives us is more about soil activity. The network of micro-organisms and fungi is more important in dense and biodiverse areas. A dynamic soil, in which the rhizosphere of the trees is more efficient, can improve water uptake. The radius of the roots can also be affected by the other trees around, as well as their depth, as shown by the soil analysis.

In addition to this, the irrigated soil is richer in nutrients assimilable by the roots. This is a negative indirect effect for pine trees planted in dry soils, which suffer from the inhibition of the decomposition of coarse organic matter. However, the upper part of dry soils is deeper and therefore allows more moisture to enter the soil and be absorbed by the upper roots. Therefore, there may be indirect effects related to the ecosystem in which the pines grow. However, it remains limited, because the three plots are finally quite similar, especially because they are very close geographically (a few tens of meters)

Let us now focus on the results of below- and above-ground traits. For each trait, the rank of the means of the plots in relation to each other is consistent with the type of phenomenon that causes the differences (between the plots): long, medium or short term phenomena . When the irrigated plot and the control one are at the terminals, it seems to mean that this trait changes in the medium and long term, as for exemple the root length and diameter. This is probably also the case for stem density, but we have no data for the stop plot. It is surprising that this is not the case for leaf traits, such as SLA, LMA, LAI. This may mean that the dynamics influencing the foliage are multiple, and do not develop only on the medium term. We will see later that it can also be attributed to other factors. If the averages of the irrigated and the stop plot are at the first and the third place (or in the opposite order), the variable is often stress-dependent (short-medium term); the control plot is, therefore, better suited than the stop one, and the irrigated plot does not face it. It is the case for the nitrogen concentration, which will come to compensate a lack of development of the roots or the needles. Finally, if the control plot and the stop plot are at the limits, it may mean that the phenomenon governing this trait acts on the very long term, and that the period since the stop plot is no longer irrigated is not sufficient to change the order. This is the case for the vegetative height, and this is why it is more interesting to look at the increase to compare the stop

plot to the other two because in this case, the height does not give a good account of the dynamics in progress.

Looking at the three PCA graphs, (figures 12, 24 and 25), we can see the spectrum of the stop plot folded in between the other two. This result is very clear, and shows that irrigation has a real influence on the observed traits. We can also deduce from what has been said above that the phenomena that allow us to differentiate the three plots act on the traits in the medium term, because of this concordance and the follow-up of irrigation-stop-control.

The spectra of the control and irrigated plots for the below-ground traits are two ellipses aligned and parallel to each other, which means that the trade-off strategies (at the root level) are similar between individuals inside plots for irrigated and non-irrigated roots (because the links between the fine roots traits are unchanged). These results go against the non-linear character of fine root traits. This may be due to the fact that soils have very similar structure and composition compared to the very large panels often used for studies, so that the external constraints on the roots are not so different and make them converge.

Besides that the lack of water mechanically decreases their development (low fine roots diameter and length, low soil root density, large “root mass ratio over 50cm”...). Therefore, the biomass allocation is not optimized to improve water capture. This is in contradiction with other studies on this topic³⁶. Again, soil type probably plays a role.

One of the most surprising points of the results is probably the leaf development; Indeed, there seems to be almost an inhibition of several needle-related traits (such as specific leaf area, needles length and needles density) in the irrigated plot compared to the control one. The foliage of conifers is quite different from that of hardwoods, it is long, thin and acicular needles. It is adapted to dry conditions : their tapered shape reduce the exchange surface, their thick and impermeable cuticle isolates the needle from its environment. and their stomata sunk in wells or grooves reduce water loss³⁷. This may be the reason why certain traits such as specific leaf area and needles length are not reduced by lack of water. Apart from that, let's remember that at the time of sampling, the dry season had not really started; the effects of lack of water on foliage, observed in other studies, may occur during the dry season each year. This could mean that there would be no memory from one year to the next for these traits.

However, there is a higher LAI in the irrigated plot. This trait determines the capacity of the tree for photosynthesis. Trees in dry environment, therefore, have a greater SLA but a lower LAI (partly due to a smaller crown radius), maybe as a strategy to avoid cavitation in the branches, while maintaining activity (useful for survival over the medium and long term). Leaf mass per area is also lower for non-irrigated pines. This is probably due to a decrease in biomass production, which is reflected in the foliage but also in other organs (such as the roots).

According to the results, there is an opposition between the nitrogen concentration and the development of leaves and roots. On the PCA (figure 25), root and needles nitrogen concentration are approximately on the same axis but in the opposite direction to the growth of the tree height and crown radius. Nitrogen is a chemical element used in the composition of proteins, and also

³⁶Bréda, “Temperate forest trees and stands under severe drought: a review of ecophysiological responses, adaptation processes and long-term consequences”, p. 2.

³⁷Raven. “Biology”. In: (2007), p. 721, p. 1.

of chlorophyll. It is therefore probably used to repair the damage caused by drought, and to compensate for the lack of photosynthetic activity. It is also very important for respiration, which increases when photosynthesis as decreases³⁸. This is also consistent with the results of other studies³⁹

The stem density could not be taken into account for the PCA because the data did not allow it (there was no data for the stop plot). However, if we look at the averages of the irrigated plot and the control plot, we can see that the lack of water leads to a lowering of the stem density. It is known that a low stem density increases the risk of cavitation⁴⁰. The pines are therefore weakened by the decrease of their stem density due to the succession of droughts. The allocation of biomass is not optimized to avoid death by cavitation.

Overall, the above-ground traits are highly correlated with each other. This was expected, as the above-ground traits are influenced by less external constraints than the root traits and are generally more readable.

But the above- and below-ground traits are also highly correlated together. The PCA of the traits all together also shows that there is no dichotomy between these two parts of the Scots pine. They are all confused, which does not make their interpretation obvious. But this reveals the complexity of the links, or perhaps a blurring related to the differences between the plots. The correlations are stronger within the irrigated plot than within the others. This may be caused by less a regular water supply to the trees of the control and stop plots; they may react differently to each other because the external elements that influence them are more chaotic.

To disentangle all this it would be interesting to make a PCA for each treatment, but it is not possible with three or four trees in each.

5.2 Limits

The results and analysis of this study must be qualified because of certain flaws in the way it was conducted. First of all, the results do not represent a general truth, which can be transposed to other species or to other regions of the world. As mentioned in the last section, the development of Pfywald pines is dependent on a set of environmental variables that have not been well disentangled here.

Then sampling hasn't been done on the same dates for all traits. A large number were in April, just before the start of the irrigation period. This has the merit of observing only the effects of long term adaptation, and not to mix it with short term adaptation. The disadvantage is that some of the effects are lost. In addition, other samples were taken later in the year, at a time when one of the three plots was irrigated. This may distort some of the relationships between traits. Other results were recovered from sampling campaigns dating back several years. Another reason to look at these results with a necessary distance is the size of the data set: 10 individuals, 3 or 4 per plot. There should have been a few more individuals, but they were discarded because they were not present in the WSL databases and therefore lacked a number of traits. The results are therefore

³⁸A.J. Burton. "Root respiration in North American forests: effects of nitrogen concentration and temperature across biomes". In: *ecosystem ecology* 131 (2002), pp. 559–568, p. 1.

³⁹Patrick J. Temple. "Nitrogen allocation in ponderosa pine seedlings exposed to interacting ozone and drought stresses". In: *New Phytologist* 130 (1995), p. 1.

⁴⁰Bréda, "Temperate forest trees and stands under severe drought: a review of ecophysiological responses, adaptation processes and long-term consequences", p. 2.

not as reliable as they could be with a large sample. For some traits the standard deviations within plots exceed the differences between the plot means. These traits have not been highlighted in the “Results” and “Discussion” sections, but the other findings of this survey cannot be corroborated by clearly significant statistical results either. In addition to this, the sampling of leaves, but especially roots, has the same problem: for each tree, only three or four leaf samples are taken, and only a few grams of roots. Given the differences between each sample from the same tree, there is probably a selection bias. Furthermore, the selection was very complicated because the pine roots can be confused with those of the surrounding plants. It is therefore not impossible that some of the roots collected do not correspond to the right tree.

Finally, some of the methods for converting field measurements into analysable data are imprecise. For the LAI calculation, for example, the system based on hemispheric photos is biased by several constraints: firstly, the scaffolding prevents a good part of the canopy from being seen from the ground. Although they have been highlighted to avoid confusion, part of the canopy cannot be taken into account. In addition, the LAI calculations for the irrigated and stop plots required weighting based on visual estimation, which may result in inaccuracies. As these photos are hemispherical, a small part of the analysed areas may have belonged to other plots. The calculation of leaf area is also subject to inaccuracies. Some of the samples were scanned with pine cone embryos, which artificially increases the results. In addition, the deployment of the needles probably caused biases. Although the needles normally also overlap, the way they are arranged on the scans is probably not completely consistent with reality.

6 Conclusion

This work focused on a description and analysis of data from Pfywald Scots pine, and the conclusions cannot be generalized to all species and ecosystems. However, it may provide a better understanding of the root and crown evolution of Scots pine and their relationships in drier environments. Indeed, the results obtained from the sampling campaigns revealed that the relationships between below-ground and above-ground traits change with the amount of water present in the soil, although some remain strong and unchanged. A study under the same conditions but with a larger number of trees, and sampling of all the samples several times during the year would allow to corroborate or deepen some of the results of this study. It is in any case a very interesting subject on which much researches could be done, to learn how trees and forest ecosystems are likely to react to future droughts.

7 Appendix

7.1 Correlations

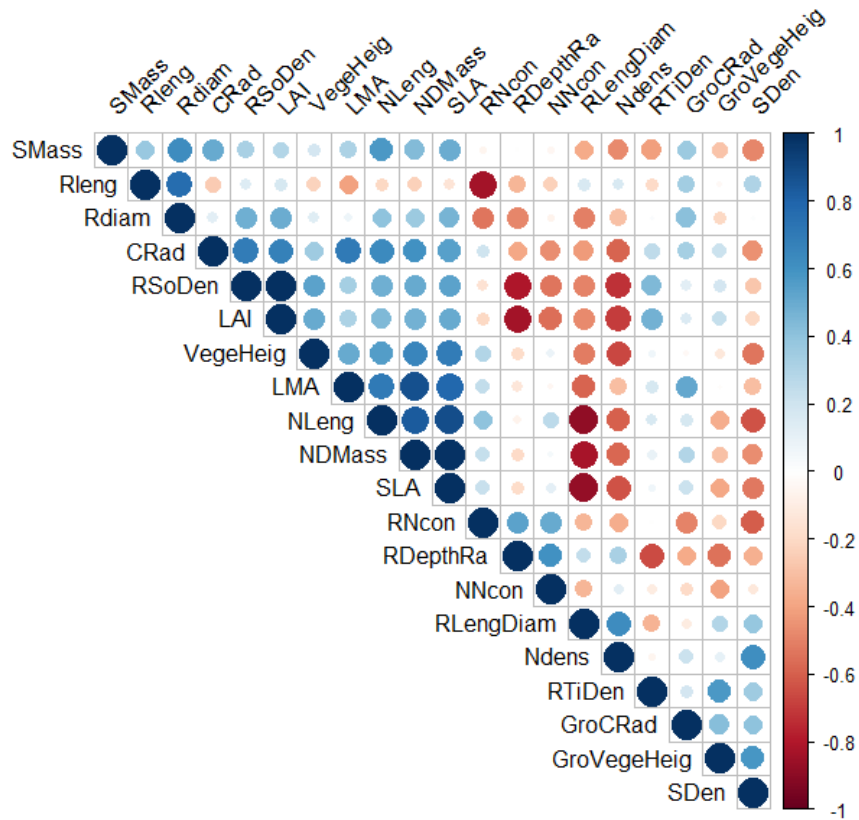


Figure 26: Correlations between all traits

(RTiden = fine roots tissue density ; RSoDen = roots soil density ; RDepthRa = Root mass ratio over 50cm ; Rdiam = fine roots diameter ; Rleng = fine roots length ; RLengDiam = fine roots length/diameter ratio ; RNcon = fine roots nitrogen concentration ; VegeHeig = tree height ; GroVegeHeig = tree height growth ; CRad = Crown Radius ; GroCRad = Crown Radius Growth ; NDMass = Needles dry mass (per “leaf” ; LAI = leaf area index ; NNcon = Needles nitrogen concentration ; SMass = seed mass ; SLA = specific leaf area ; NLength = Needles Length ; Ndens = Needles density ; LMA = Leaf Mass per area)

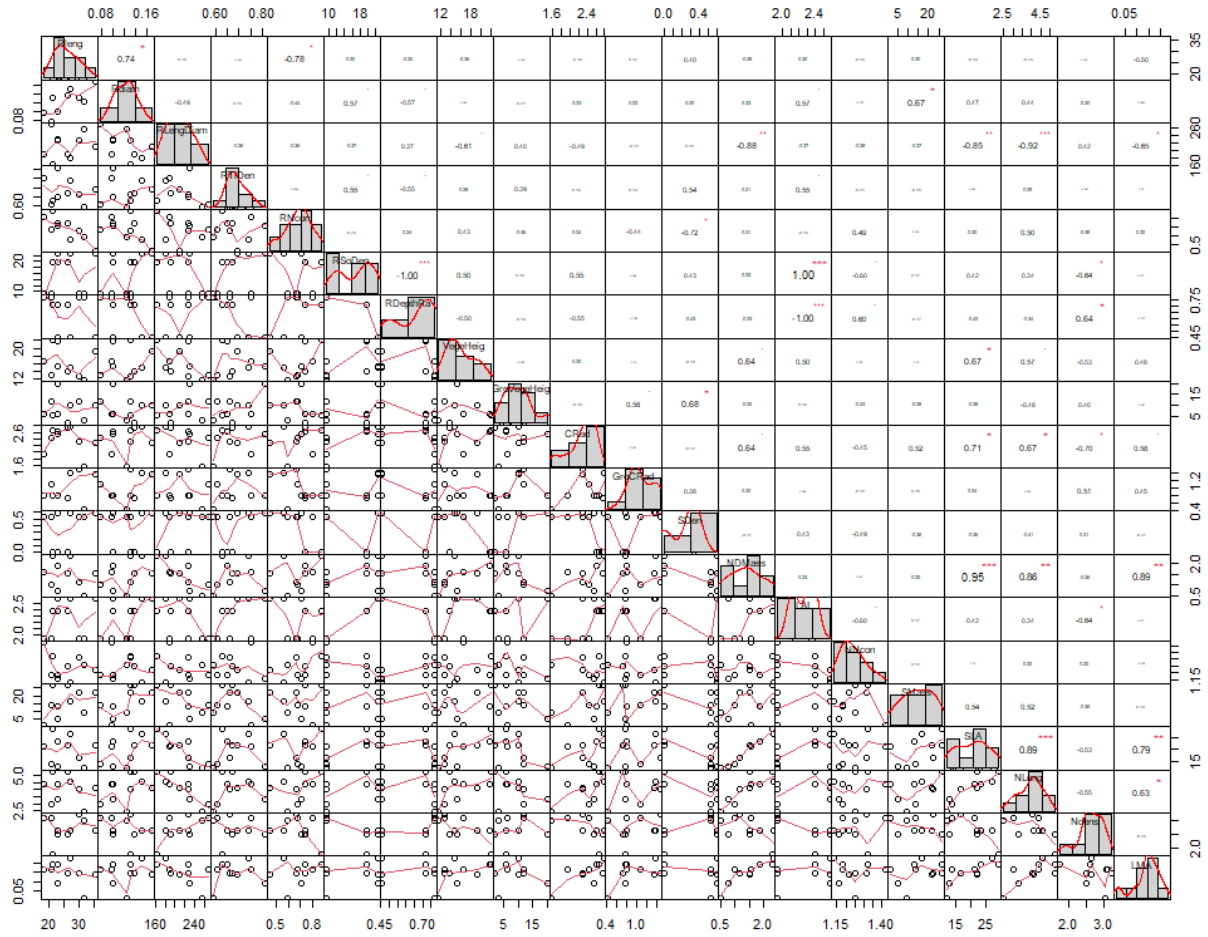


Figure 27: Correlations between all traits

(RTiden = fine roots tissue density ; RSoden = roots soil density ; RDepthRa = Root mass ratio over 50cm ; Rdiam = fine roots diameter ; Rleng = fine roots length ; RLenDiam = fine roots length/diameter ratio ; RNcon = fine roots nitrogen concentration ; VegeHeig = tree height ; GroVegeHeig = tree height growth ; CRad = Crown Radius ; GroCRad = Crown Radius Growth ; NDMass = Needles dry mass (per “leaf” ; LAI = leaf area index ; NNcon = Needles nitrogen concentration ; SMass = seed mass ; SLA = specific leaf area ; NLength = Needles Length ; Ndens = Needles density ; LMA = Leaf Mass per area)

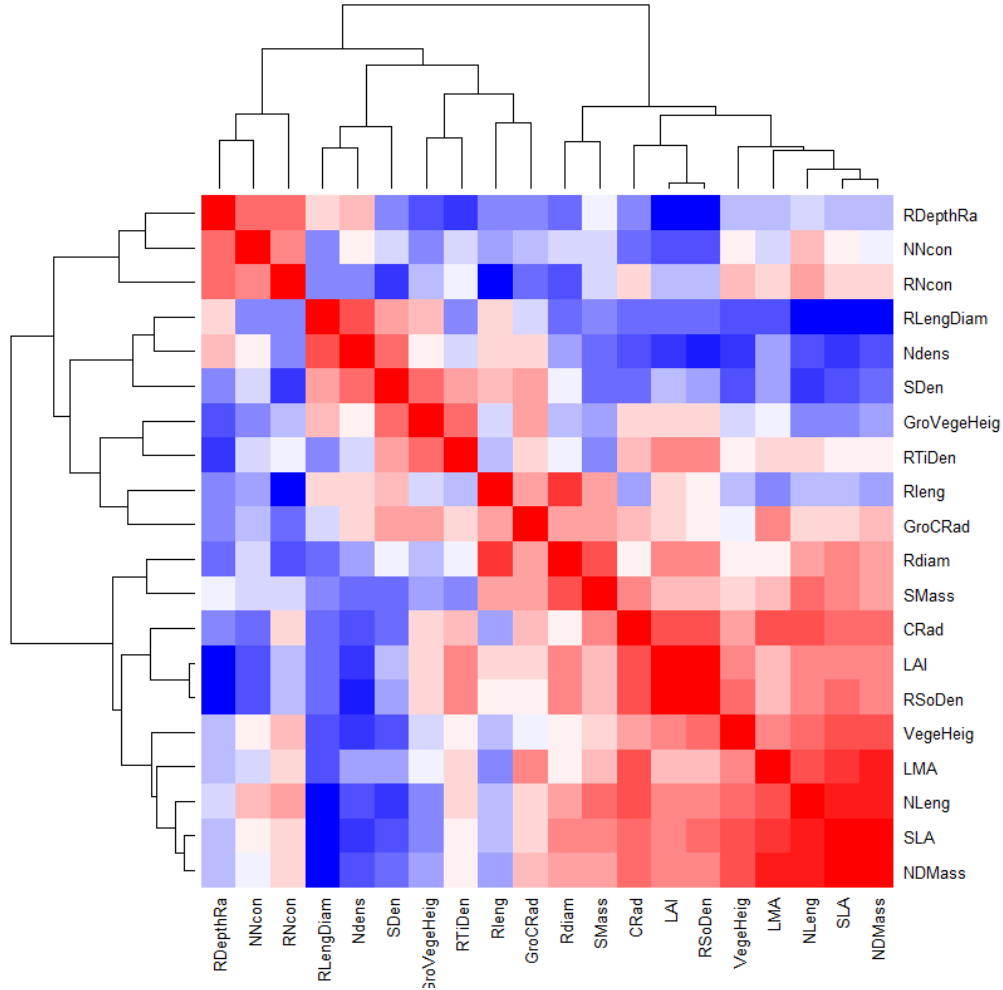


Figure 28: Correlations between all traits

(RTiden = fine roots tissue density ; RSoDen = roots soil density ; RDepthRa = Root mass ratio over 50cm ; Rdiam = fine roots diameter ; Rleng = fine roots length ; RLengDiam = fine roots length/diameter ratio ; RNcon = fine roots nitrogen concentration ; VegeHeig = tree height ; GroVegeHeig = tree height growth ; CRad = Crown Radius ; GroCRad = Crown Radius Growth ; NDMass = Needles dry mass (per “leaf” ; LAI = leaf area index ; NNcon = Needles nitrogen concentration ; SMass = seed mass ; SLA = specific leaf area ; NLeng = Needles Length ; Ndens = Needles density ; LMA = Leaf Mass per area)

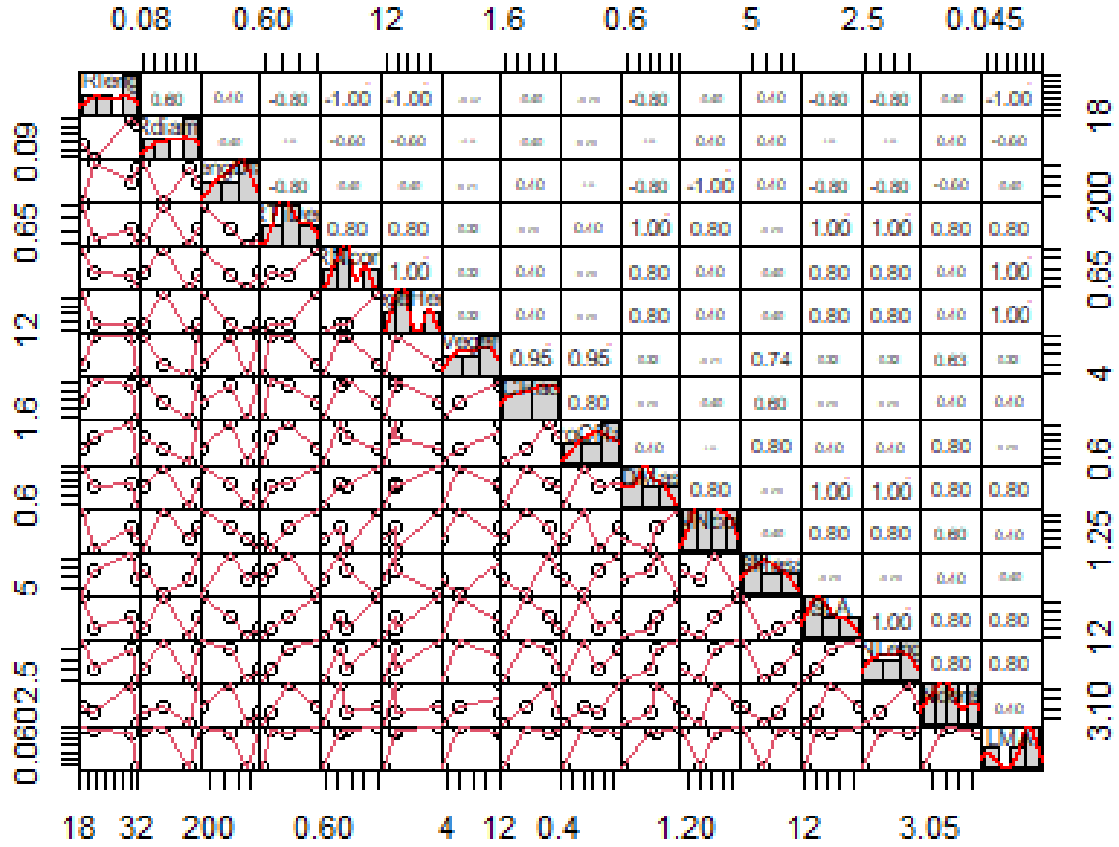


Figure 29: Correlations in control plot

(RTiden = fine roots tissue density ; RSoden = roots soil density ; RDepthRa = Root mass ratio over 50cm ; Rdiam = fine roots diameter ; Rleng = fine roots length ; RLenDiam = fine roots length/diameter ratio ; RNcon = fine roots nitrogen concentration ; VegeHeig = tree height ; GroVegeHeig = tree height growth ; CRad = Crown Radius ; GroCRad = Crown Radius Growth ; NDMass = Needles dry mass (per “leaf” ; LAI = leaf area index ; NNcon = Needles nitrogen concentration ; SMass = seed mass ; SLA = specific leaf area ; NLength = Needles Length ; Ndens = Needles density ; LMA = Leaf Mass per area)

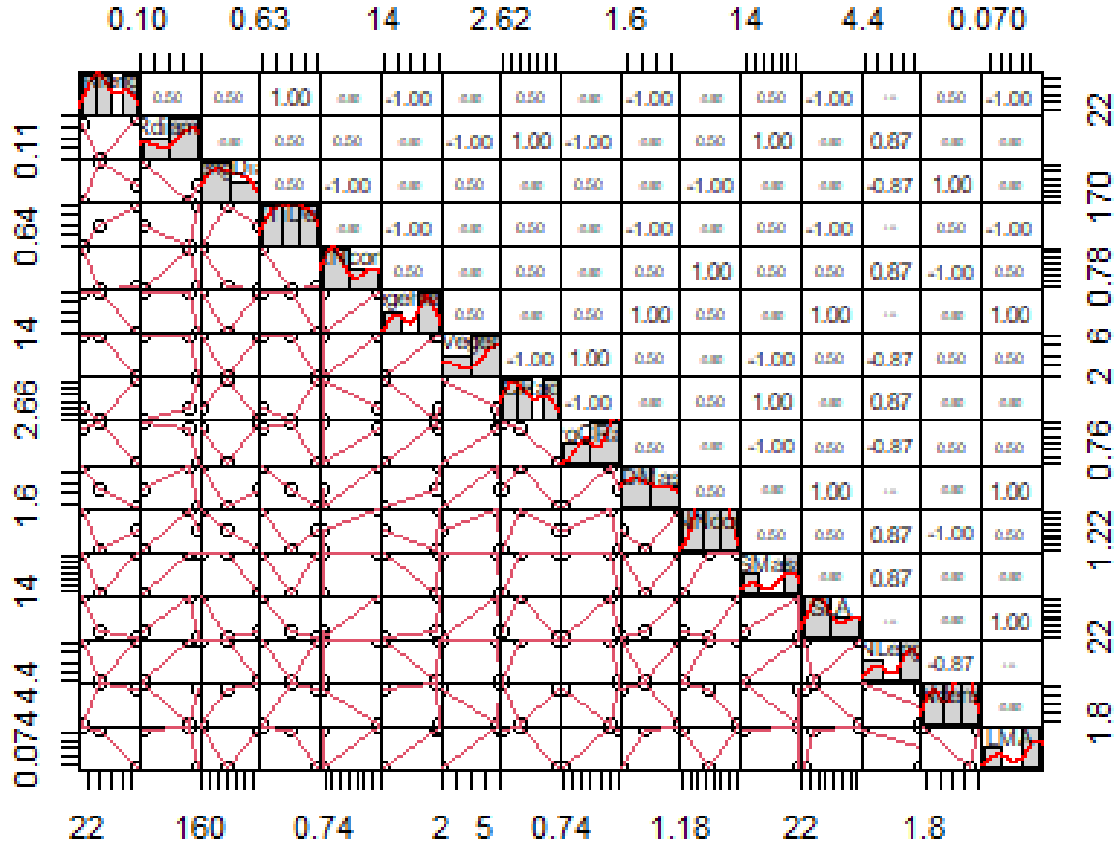


Figure 30: Correlations in stop plot

(RTiden = fine roots tissue density ; RSoden = roots soil density ; RDepthRa = Root mass ratio over 50cm ; Rdiam = fine roots diameter ; Rleng = fine roots length ; RLenDiam = fine roots length/diameter ratio ; RNcon = fine roots nitrogen concentration ; VegeHeig = tree height ; GroVegeHeig = tree height growth ; CRad = Crown Radius ; GroCRad = Crown Radius Growth ; NDMass = Needles dry mass (per "leaf" ; LAI = leaf area index ; NNcon = Needles nitrogen concentration ; SMass = seed mass ; SLA = specific leaf area ; NLength = Needles Length ; Ndens = Needles density ; LMA = Leaf Mass per area)

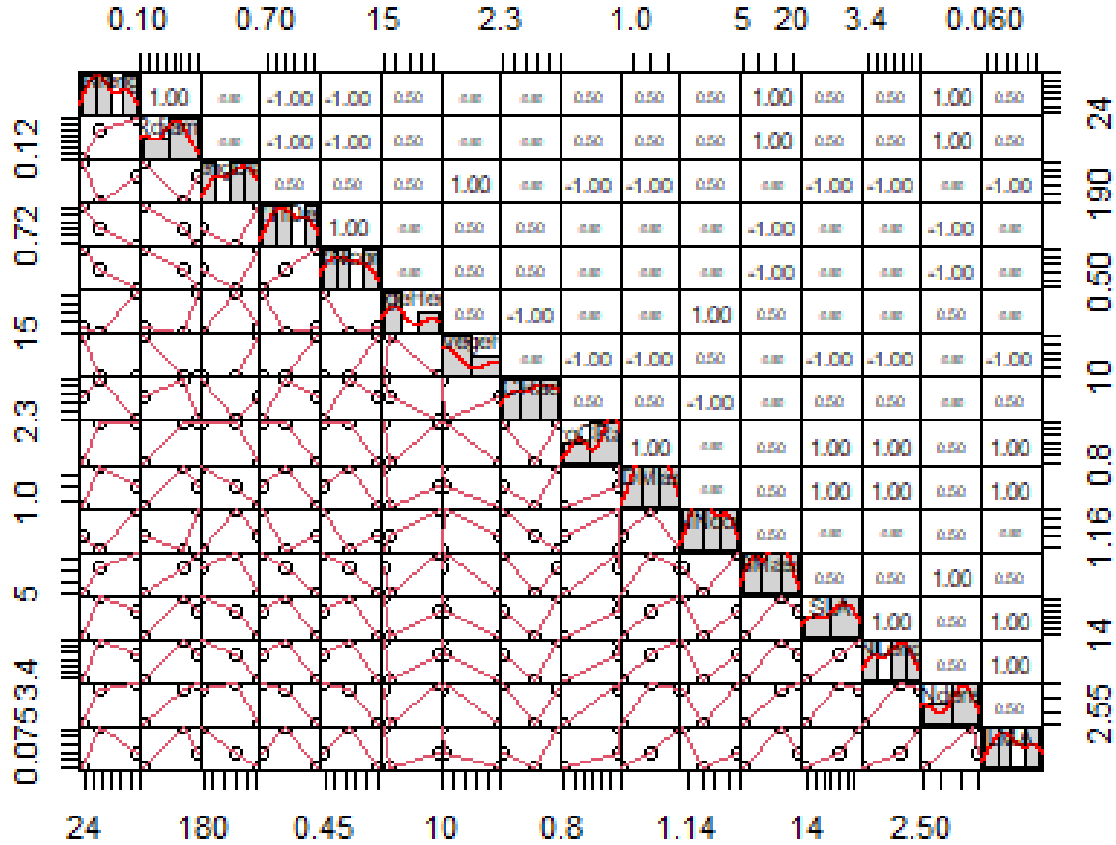


Figure 31: Correlations in stop plot

(RTiden = fine roots tissue density ; RSoden = roots soil density ; RDepthRa = Root mass ratio over 50cm ; Rdiam = fine roots diameter ; Rleng = fine roots length ; RLenDiam = fine roots length/diameter ratio ; RNcon = fine roots nitrogen concentration ; VegeHeig = tree height ; GroVegeHeig = tree height growth ; CRad = Crown Radius ; GroCRad = Crown Radius Growth ; NDMass = Needles dry mass (per "leaf" ; LAI = leaf area index ; NNcon = Needles nitrogen concentration ; SMass = seed mass ; SLA = specific leaf area ; NLength = Needles Length ; Ndens = Needles density ; LMA = Leaf Mass per area)