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PLANT AGRONOMIC PERFORMANCE AND SELECTION FOR GREEN ROOFS UNDER MEDITERRANEAN AND MARITIME CLIMATE CONDITIONS

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A tutti coloro che in questi anni hanno creduto in me,

offrendomi e mostrandomi sinceramente il loro amore,

la loro stima ed il loro supporto;

che hanno saputo gioire, vivere e codividere con me

questo percorso professionale e di vita

'What is essential is invisible to the eye' [...]

' It is the time you have wasted on your rose

that makes your rose so important.'

"The Little Prince" by Antoine de Saint-Exupéry

CONTENTS

APPENDIX

The present thesis is partially based on the following proceedings papers, which are referred to by their Roman numbers.

I. M.E. Provenzano, M. Cardarelli, F. Saccardo, G. Colla, A. Battistelli, S. Proietti (2009) "Evaluation of Perennial Herbaceous Species for their Potential Use in a Green Roof under Mediterranean Climate Conditions", in Proceedings of *2nd International Conference on Landscape and Urban Horticulture*, 9-13 June 2009, Bologna, Italy.

II. M.E. Provenzano, M. Cardarelli, M. Crasso, G. Colla (2010) "Evaluation of potential green roof plant species in a Mediterranean climate", in Proceedings of *World Green Roof Congress: green roofs for a changing climate*, 15-16 September 2010, London, UK.

1 GENERAL INTRODUCTION

During the last sixty years an extreme and rapid increase in the urban population has occurred. It is predicted that the world population will be concentrated in urban areas, reaching by 2050 70% and 86% in more developed countries (Population Division of the Department of Economic and Social Affairs of the United Nations, 2007).

Rapid urbanisation, indeed, has resulted in upgrading of interest in urban green space, and the ways in which this green space can benefit cities and their inhabitants have become key issues in urban planning.

As a matter of fact, the concepts of “sustainable cities”, recently enhanced by the European Commission (2006), and “green building”, which are dominating the contemporary design thinking, are effective expressions of an increasing demand and burden of healthier and more liveable environments surrounding cities and town (Johnston and Newton, 2004).

Under this perspective the so-known environmental “green lungs”, of which green roofs are part, will represent a pivotal element to sustain and preserve life in urban and suburban areas (Johnston and Newton, 2004).

Green roofs (or vegetated roof tops) represent by far the most important tool for building greening and, nonetheless, a significant opportunity to expand the presence of vegetated surfaces in urban areas by replacing impermeable surfaces, with the associated positive impact on the quality of city life (Johnston and Newton, 2004; Banting *et al.*, 2005). In fact, roof greening has been recently recognized as part of greening strategies adopted by several cities to support sustainable urban ecosystems and to improve urban dwellers’ quality of life. Consequently, vegetated roofs are increasingly being incorporated as a sustainable practice in building design (Banting *et al.*, 2005; Snodgrass and Snodgrass, 2006; Newman and Jennings, 2008; City of Los Angeles, 2006).

Green roofs, indeed, offer many advantages, especially on an urban scale. In cities they provide new green areas and reduce the volume of water runoff, heat urban island effect and air and water pollution, while also increasing biodiversity. With regard to buildings, green roofs contribute to lower energy consumption and realize other economic benefits (Oberndorfer *et al.*, 2007; Dunnett and Kingsbury, 2008).

The widely known advantages of green roofs have encouraged the expansion of the roof greening strategy in several countries. In Europe is possible to count over 75 municipalities which currently provide incentives or defined requirements for green roof installation (Peck and Kuhn, 2001).

Despite the growing interest and research on green roofs, this greening technique in Italy is relatively new and the scientific findings are deficient (Fioretti *et al.*, 2010). However, there have been some active areas in Italy, of which Bolzano province and City Council are the main examples. It was in 2004 and in this municipality when green roofs had been included for the first time in a public urban policy in Italy (RIE procedure of Bolzano City Council). More recently, through the initiative of the Italian green roofs association (AIVEP), two important steps in green roofs policing came through the production of a national standard regulation for green roofs (UNI 11235, 2007) and the inclusion of green roofs in the national legislation (D.P.R.59/09). This law represent the adoption of the European directives 2002/32/CE and 2006/32/CE related to energetic aspects, by which people can benefit until up 55% of tax reduction on the cost of the construction.

Finally, although green roofs have been proven to be efficient as a passive cooling building technique, especially in warmer climates (Theodosiou, 2003), the high construction cost, the climate conditions and the scarcity of quantitative data supporting the effectiveness of green roofs in our climate have discouraged the expansion of green roofs in the Italian construction industry.

1.1 RESEARCH OBJECTIVES

Green roof technology is relatively new and when the project started there was relatively little accessible or published information about how different types of succulent and non-succulent species perform in roof conditions in Italy, and more generally in Mediterranean areas. Furthermore, if compared to other research field such as storm water retention and management, very few researches have focused their attention to plant performance and on plant selection for green roofs, where plant selection and survival represent overall a key element for their success.

The main hypothesis was that plant diversity, origin and variability affect the capacity of plant to succeed under high stress environments.

The objectives of the present study are specified as follows:

- To select and evaluate plant species, both cultivated and wild collected and assess their suitability for growth in a green roof environment, through screening programs in both Mediterranean and maritime climates.

- To look at the local flora and test whether or not native species give good or better performance compared to non-native species for green roofing application.

Some additional studies were carried out to monitoring the thermal mitigation effect on the roof surface of green roof systems with different depth and vegetation in a Mediterranean climate, and how this can be affected by the presence of vegetation. Furthermore, in maritime climate conditions a germination study and a vegetation survey study were preliminarily undertaken, in order to provide additional and complementary information on plant performance and growth in green roofs conditions.

The last year of the PhD was conducted at the Landscape Department of Sheffield University, under the supervision of Dr. Nigel Dunnett and Dr. Zoe Dunsiger, this gave me the opportunity to enrich this research and further investigate plant-related aspects of green roofs in a climate different to that of southern Europe.

Since the present research mainly aimed to outline a range of species adaptable to the roof growing conditions, several screening and plant evaluation studies were performed. The objectives of the thesis were achieved through a series of investigations on plant selection started with: preliminary interviews to professionals; a review of precedent studies; and a glasshouse experiment for drought tolerance (Viterbo, Tuscia University) along with laboratory studies conducted at Tuscia University and CNR (National Research Council), Porano. Afterwards, a field collection was created, by which the plant material - including the wild collected - had been further evaluated. Finally, other investigations on the experimental roof set up at Tuscia University (Viterbo) were conducted to evaluate plant growth and adaptability to the roof conditions and the thermal effects of the different green roof systems tested.

Research in maritime climate conditions was performed at Sheffield University in three different roof sites and in a glasshouse. A screening to evaluate how substrate depth can influence plant germination was conducted, other studies were undertaken to test plants with different provenance for their growth on the rooftop environment and in some cases plant growth performance in glasshouse vs. roof conditions were compared. Furthermore, a vegetation survey was conducted to evaluate plant development and diversity in relation with substrate depth on an existing and established green roof.

With a high level of confidence the Intergovernmental Panel on Climate Change (IPCC) reports that "in southern Europe, climate change is projected to worsen conditions (high temperatures and drought) in a region already vulnerable to climate variability" and that "in northern Europe, climate change is initially projected to bring mixed effects, including some benefits, but as climate change continues, its negative effects are likely to outweigh its benefits" (Adger *et al.*, 2007). Under this perspective the studies conducted during this PhD program which have taken place in two different European climates (Mediterranean and maritime) help to outline a more complete picture concerning green roof research on a wider climate-geographical range.

This thesis seeks to extend the plant species used in green roofing, specifically in a Mediterranean area, because of the changes being seeing in the environment due to climate change. In addition, the more extreme climatic conditions experienced in Mediterranean versus maritime climates requires a reconsideration of those species typically used and seen to be successful in north-west European green roof projects.

Described below are some of the core benefits in vegetating surfaces in urban environments from various studies. With the increasing need to consider this environment and potential mitigation, comparison of the maritime experience and possibilities in the Mediterranean regions is made in this study.

1.2 THESIS OUTLINE

In Chapter 2, plant selection and performance in Mediterranean climate conditions are presented through a glasshouse plant screening for drought tolerance and a roof based plant evaluation trial. Furthermore, since the benefits of thermal mitigation from green roofing can be consistent, particularly in warm climates, and because the vegetation structure and its presence may affects also the roof's thermal property, temperature patterns monitored underneath different green roofs system were compared and plant effect evaluated. This work is here presented in terms of complementary and additional to the plant evaluation study in roof condition under a Mediterranean climate, to provide additional information which may help also to highlight plant functional benefit (Chapter 3).

In Chapter 4, plant growth at different stages in roof conditions under a maritime climate was analyzed. Particularly, the attention was mainly focused on genera screening and the effect of the provenance and geographical origin of the plant material on its growth/performance.

In the general conclusion an attempt is made to provide an overall picture of the results obtained in both Mediterranean and maritime climates (Chapter 5).

1.3 OVERVIEW OF GREEN ROOFS

1.3.1 Benefits of green roofing

Green spaces in urban areas are often scarcely represented and this lead cities to have to deal with many environmental, economical and sociological issues. How green roofs may play a role in it?

Promotion of Biodiversity and Habitat Preservation

In high-densely populated urban environments, where green spaces are scarce, green roofs reveal also their vital potential to support life.

Even though green roofs are not a direct replacement of ground-based habitats, they can provide to endangered or displaced flora and fauna alternative restored habitats (Brenneisen, 2003; Grant, 2006).

Dunnett *et al.* (2010) stated that "from both an ecological and an aesthetic viewpoint, there are considerable benefits in promoting plant species-diversity in green roof vegetation". A plant rich roof, particularly characterized by morphologically-diverse functional types, can be less vulnerable to environmental fluctuation and can be more efficient than a low diverse one (Obendorf *et al.*, 2008).

In Switzerland, where more than everywhere else the role of green roofs in enhancing biodiversity and wildlife has been studied, have been found on vegetated roofs numerous endangered species listed in the red data books (Brenneisen, 2003).

In creating habitat the use of native species of local provenance is advised (Grant, 2006). Despite the urban landscape is often rich of non-native plants, because the variability existing within populations and the scarce knowledge about native plants genetics, the use of local resources should provide higher guarantees of suitability and adaptation to the local climate and its potential must not be underestimated.

Screening studies may lead to a wider range of possible plant which grow successfully in the urban environment, and so provide more heterogeneity and diverse habitats. For this reason, plant screening and selection is one of the main research objectives of this thesis which was attempted to develop in both Mediterranean and maritime climates.

Energy Savings and thermal benefits

Green roofs have been shown to impact positively on a building's energy consumption by improving its thermal performance, particularly during the summer. In winter, their insulating effect help to reduce the heat loss from inside the building through the roof, consequently reducing heating needs. In summertime, green roofs reduce roof surface temperatures and ambient air temperatures, thus lowering cooling energy demand (Niachou *et al.*, 2001; Liu and Minor, 2005).

Nevertheless, the insulation properties of green roofs varies through the year and depends on the technical characteristics of the system and on the climate conditions (Niachou *et al.*, 2001; Liu and Baskaran, 2003; Tan *et al.*, 2003). It is also a truism that the insulating effect played by green roofs, by reducing the extreme temperature fluctuations on the roof surface, extends the roof membrane life span (Köehler, 2003; Liu and Baskaran, 2003; Banting *et al.*, 2005, Abram and Salchegger, 2006).

Temperature is one of the main problems connected to the climate change, and in warm climates such as the Mediterranean mitigating temperature may helps to improve quality of life and reduce energy consumption to cool down indoor environments.

This aspect is considered in Chapter 3, where monitoring under Mediterranean climate conditions of the roof surface and underneath vegetated and non-vegetated green roof systems temperatures is carried out in this work.

Air Quality and Carbon Sequestration

In the UK buildings are responsible of the 50% of the CO_2 emissions, which are mainly from heating and cooling the indoor environments (Hassel and Coombes, 2007). Moreover, green roofs help to slow the formation of ground-level ozone by lowering air temperatures. Vegetation behaves as a sink of pollutants resulting in the removal of certain pollutants from the air through dry deposition process and microclimate effects (Emilsson *et al.*, 2006; Yang *et al.*, 2008). As quantified by Yang *et al.* (2008) green roofs can remove ground-level ozone (O_3) by up 52% of the total and also particulate matter (PM) and other gaseous pollutants, including nitrogen oxides (NO_X), sulphur dioxide (SO_2) and carbon monoxide (CO). After these considerations roof greening by reducing buildings' energy consumption, air pollution and greenhouse gas emissions may thus have a positive effect on a global scale on the climate change. In fact, green roofs can represent a tool helping the climate change mitigation, while often undertaken at the local level has global benefits, with possible ancillary benefits at the local or regional level.

Reducing the Urban Heat Island Effect

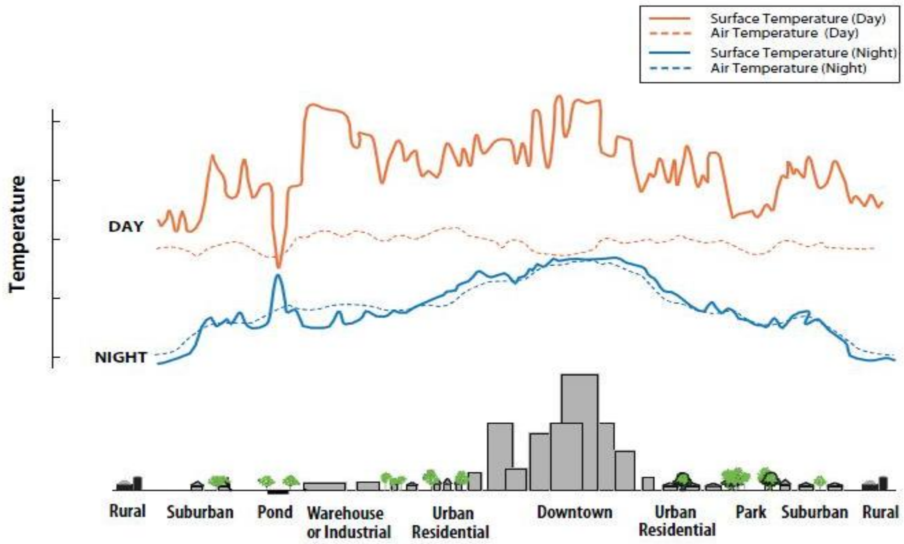
In cities, a high portion of the incoming solar energy is absorbed by the hard, heat-absorbent surfaces (Fig.1.1). As a consequence, the local climate in the city is altered, causing a significant rise of the urban temperature and other alterations, known as the heat island effect (Alexandri and Jones, 2008; U.S. Environmental Protection Agency's Office, 2010).

The lack of vegetation in the urban landscape seems to be the main cause of the temperature`s increasing in urban areas (Alexandri and Jones, 2008; U.S. Environmental Protection Agency's Office, 2010). In fact, as observed by Takebayashi and Moriyama (2007), a vegetated surface tends to have a small sensible heat flux, similarly to a highly reflective white surface, due to the effect of the evapo-transpiration process. This means that the "heat island" phenomenon is a result of ground-level air temperature being much higher than in surrounding rural areas, where the vegetation and plant life cool the air through moisture retention and subsequent water evaporation from soil and transpiration from leaves.

Bass *et al.* (2002) using a sophisticated model found that if only 5% of the roof area of Toronto had green roofs the temperature at mid-day in summer would be reduced by 1-2 °C.

The Environmental Protection Agency's Office of Atmospheric Programs and also the Urban heat island group suggest green roofs as one of the possible strategies to mitigate the heat island in cities, as green roofs can help to increase the number of green surfaces in the urban areas.

Fig 1.1 Variations of Surface and Atmospheric Temperatures depending on the land use



(*Source:* http://www.epa.gov/heatislands/about/index.html)

Enhanced Stormwater Management and Runoff Quality

Notably, another pivotal element which makes green roofs interesting for built environments is their effectiveness in the reduction and the management of stormwater runoff. Indeed, in urban areas the water balance changes compared to the rural areas, because the proportion between the various elements of the water balance are considerably different.

The dominance of impermeable surfaces in cities increases the amount of runoff lost as surface runoff, while the lack of vegetated areas and ground-surfaces is the cause of a lower water loss trough evapo-transpiration and ground storage processes (Hathaway *et al.*, 2008). It has been given evidence by several studies (VanWoert *et al.*, 2005; Teemusk and Mander, 2007; Cappelletti *et al.*, 2009) that green roofs can retain great amount of stormwater up to 100%, thus avoiding the rapid water runoff and high peak flow which are typical in cities and may cause floating problems (Villarreal and Bengtsson, 2005). Because roofs represent a large area of impermeable surfaces in town and the beneficial effect of green roofs on stormwater management for a city can reach 38% in savings (Banting *et al.*, 2005), is understandable why so much research has been done on this field.

Plants and growing medium not only retain and delay the release of stormwater but also act as a filter of water contaminant, especially nitrogen contaminant, when the green roof maintenance is appropriately managed (Emilsson *et al.*, 2006).

A Raw Benefits/Costs Comparison

A green roof might have higher initial costs than most conventional or cool roofs, however a full life-cycle analysis can identify how the roof benefits at the same time both the building owner and the community (Tab.1.1). In many cases, these advantages justify the cost of green roofs, particularly in densely populated areas (Kosareo and Ries, 2007). As illustrated by Banting *et al.*, 2005, in Toronto city green roofs through their positive benefits can generate a cost reduction by up to 38% for the stormwater management, 25% for the urban heat island effect and 22% for the building energy consumption.

Tab.1.1 Benefits and costs of green roofs

Benefits/Costs	Energy, hydrology, UHI benefits	Other benefits
Private Benefits	– Reduced energy use – Extended service life	– Noise reduction – Aesthetic value – Food production
Public Benefits	– Reduced temperature – Reduced storm water – Reduced installation costs (from widespread technology use)	– Reduced air pollutants – Reduced greenhouse gases – Human health benefits
Private Costs	– Installation Architecture/Engineering – Maintenance – Program administration	N/A
Public Costs		N/A

Sources: Grant *et al.*, 2003; Banting *et al.*, 2005; Dunnett and Kingsbury, 2008.

1.3.2 From the traditional roof gardens to the contemporary types of green roofs

A brief history of green roofs

The origin of greening roofs is likely to be connected with the development of the first important civilizations. It is true that by up to X-IXth centuries B.C. green roofs were popular and well established in the Eastern countries (Matteucci *et al.*, 1994). Probably, the most famous roof gardens of all the time are the hanging gardens of Babylon, XIth century B.C., which also were considered one of the Seven Wonders of the World (Osmudson, 1999).

In Europe, remains of earth-sheltered huts, dating from the Viking period, have been found in Ireland and Scottish islands (Grant *et al.*, 2003). Scandinavians have used sod (or turf) to cover pitched roofs and the practice continues to the present day (Osmudson, 1999; Grant *et al.*, 2003).

In Italy the oldest practice of roof greening goes back to the Etruscan age (IXth century B.C), who used to cover their tombs with greenery (Abram and Salchegger, 2006). The practice of greening roofs was known and used also during the Roman age and the middle age up until the present years, assuming through the time different values and functions (Osmudson, 1999; Abram and Salchegger, 2006).

It is only on the XXth century when few vanguard architects as Gropius, Frank Lloyd Wright and particularly Le Corbusier - who promoted and re-established this technology in the modern architecture - that green roofs started to assume their current role in architecture up to arrive to the modern-engineered green roof systems and to the first roof landscaping guidelines, created by the German Landscape Research & Construction Society (FLL) by the end of the XIXth century (Matteucci *et al.*, 1994; Osmudson, 1999; Appl, 2009).

Types of green roofs

A general description of a modern green roof system includes five basic elements, from the top to the bottom: the vegetation layer; the substrate (a lightweight soil media); the filter layer (a geotextile); the drainage layer (engineered coarse media or plastic profiled elements); the waterproof-root barrier. Green roofs are also known as eco-roofs, vegetated roofs, brown roofs, spontaneous roofs etc. All these definitions express different features of a unique technology.

As defined by the FLL guidelines (2008) green roofing is classified into three main types on the basis of use, construction method and maintenance requirements: extensive; simple-intensive or semi-intensive and intensive (Tab.1.2).

Extensive green roof systems are the most lightweight and require a minimal level of maintenance. They are often not accessible by general public, and may be designed for flat or sloped roofs (Dunnett and Kingsbury, 2008; FLL, 2008). Due to the substrate shallowness the range of planting options is not wide and usually includes hardy drought-tolerant perennials, typically *Sedum* spp., grasses and alpine species.

Simple-intensive green roofs can be designed to be accessible, they are slightly deeper than normal extensive and similarly to the extensive they require low maintenance. The plant choice for these systems is wider and diverse. Because of these characteristics simple-intensive green roofs can be considered as a combination of the other two systems, extensive and intensive (Dunnett and Kingsbury, 2008; FLL, 2008).

Finally, intensive green roofs are more close to the concept of conventional gardens thus are often referred as roof gardens (Grant *et al.*, 2003; Dunnett and Kingsbury, 2008). They require a high level of maintenance which includes fertilizing, weeding and cutting. Compared to the other systems, the intensive are heavier because of the substrate depth, which is generally at least 15 cm deep. They can support any kind of vegetation, from lawn and herbaceous perennials through shrubs and even trees (Grant *et al.*, 2003; Dunnett and Kingsbury, 2008).

Tab.1.2 In the following table types and principal characteristics of green roofs are summarized.

Type	Depth	Accessibility	Maintenance requirements	Planting
Extensive	Usually 2 to 10 cm (can be up to 20 cm)	Generally accessible only for maintenance	Low	Mosses, succulents, grasses and some forbs.
Simple intensive	Usually 12 to 50 cm (can be up to 100 cm)	Accessible to public or only for maintenance	Regular	Grasses, forbs and shrubs
Intensive	15 to 200 cm	Accessible	High	All vegetation

Sources: Grant *et al.,* 2003; Hassel and Coombes; 2007; Dunnett and Kingsbury, 2008; FLL, 2008.

2 STUDIES ON PLANT SELECTION AND PERFORMANCE FOR EXTENSIVE AND SIMPLE-INTENSIVE GREEN ROOFS IN A MEDITERRANEAN CLIMATE

2.1 BACKGROUND

The top of a roof in a city may be one of the most physically stressful places in which plants can grow. In this habitat, plants are exposed to several environmental stresses (high solar radiation, low nutrient supply, drought, heat, freezing, wind speed etc.).

It is also well-known that green roofs provide important environmental benefits such as storm water retention, summer cooling, increasing urban biodiversity as well as a large number of economical, sociological and ecological advantages (Dunnett and Kingsbury, 2004). Furthermore, it is also true that the performances and benefits of a green roof can vary significantly depending on the type and depth of the growing medium used, on the vegetation density and selection (Compton and Whitlow, 2006; Dunnett *et al.*, 2008; Lundholm *et al.*, 2010) on the structural features of the entire system (Simmons and Lady Bird Johnson Wildflower Center, 2010) and, finally, on the climatic and geographical conditions (Getter and Rowe, 2008).

Although these benefits have long been investigated by researchers, relatively few studies have been conducted to investigate one of the key factors for green roofs to be successful: the plant selection (Monterusso *et al.*, 2005, Dunnett *et al.*, 2008b, Getter and Rowe 2008). Particularly, there is a lack of information concerning Mediterranean areas, as most of research and projects have been carried out in temperate summer rainfall countries or in other continents, and when vegetation is concerned works do usually focus on survival and growth of commonly used taxa, principally Sedums and other succulents (Emilsson, and Rolf, 2005; Monterusso *et al.,* 2005; Durhman *et al.,* 2006). Furthermore Dunnett and Kingsbury (2004) pointed out the need of screen plant material for different green roof functions.

As stated by Getter and Rowe (2008) “Plants chosen for a specific location must be able to survive local geographical climate extremes as well as harsh microclimate conditions often found on rooftops”.

Because of the shallow substrate depth and the consequently limited moisture content, the presence of drought-tolerant vegetation is essential for the long-term success of extensive green roofs (Koehler, 2003; Durhman *et al.*, 2006; Dunnett and Kingsbury, 2008; Getter and Rowe, 2008).

Dunnett and Kingsbury (2004) considered the role of natural plant community as model for green roof vegetation. As suggested by the authors one key to identify new suitable species and to expand the potential diversity of plants is looking at some drought- and exposure-tolerant characteristics combined with aesthetic qualities such as: low-mat forming or cushion-forming growth; gray or silver foliage - a plant biological adaptation to avoid water loss which is also a valuable aesthetic character; rooting stems; leaves with thick cuticle or curved leaves surfaces which helps to reduce the heat and solar radiation; and finally shallow roots.

Plants have developed many strategies of adaptation to stressful environments. Rozema and Verkleij (1991) sustained that tolerance to naturally occurring stresses have evolved over a long period of time, while plant response in adaptation to stresses due to human activity is more recent and can develop very rapidly. Water is an essential component of plant life and because it plays a large role in physiological processes, its availability is often limiting. Mechanisms that enable plants to survive water deficit are many and can be divided into two categories: physiological adaptations and morphological adaptations. Physiological adaptations mainly concern transpiration control and metabolic pathway of photosynthesis:

- Carbon fixation that ultimately dictates plant growth and development reflects the balance between photosynthesis and respiration. Plants are subjected to water loss from leaves surface, which occurs mainly through stomata. To carry out metabolic processes as photosynthesis, plants need to exchange resources with the surrounding environment (light, gasses, water and nutrients), and plant respiration and CO2 uptake require that specific structures like stomata stay opened to allow the gas exchanges. The result is that when stomata are opened water particles pass from leaf cells to the atmosphere, because of a vapour pressure deficit between the mesophyll environment and the atmosphere. To control transpiration, thus water loss, plants can respond with stomatal closure, reduced stomatal conductance and leaf abscission during summer (Werner et al., 1999; Taiz and Zeiger, 2002);

- Plants from different environments have developed different photosynthetic metabolisms. Most of plants from temperate climates have the so called C_3 photosynthetic pathway, in which CO2 is assimilated leading to the synthesis of C_3 compounds (triose-phosphates), while species adapted to high temperature and irradiance conditions have C_4 metabolism, in which the first product of CO_2 fixation is a 4-carbon intermediate. Finally, CAM biochemical pathway is undoubtedly an adaptation to drought, since CAM plants can close their stomata during most of the day, when environmental conditions are not favourable, and is found in succulents from arid and semi-arid regions and epiphytes from tropical and subtropical regions (Lambers et al., 2008). CAM pathway is characterized by spatial and temporal separations of different processes: $C0_2$ uptake; organic acid formation and accumulation; organic acid decarboxylation and $C0_2$ fixation through Calvin cycle. There are three main metabolic phases as distinguished by Lüttge (2004): (phase I) nocturnal uptake of CO_2 via open stomata, fixation by phosphoenolpyruvate carboxylase (PEPC) and vacuolar storage of CO_2 in the form of organic acids, mainly malic acid, and (phase 2) daytime remobilization of vacuolar organic acids, decarboxylation and refixation plus assimilation of CO_2 behind closed stomata in the Calvin-cycle (phase III).

Morphological adaptations involve aspects of plant anatomy and architecture at different levels as:

- Leaf anatomy (i.e. changes in the number of layers of mesophyll, in cells dimensions, presence of non-glandular leaf hairs) where anatomical variations in leaves development induced by environmental conditions can directly affect photosynthesis and transpiration process increasing plant water use efficiency (Turner and Kramer, 1980) and photoprotection when drought and high light intensity are combined. Moreover many plants have adapted to xeric environments by eliminating leaves and replacing those with thorns or by reducing leaves area and size to reduce transpiration (Taiz and Zeiger, 2002);

- Root anatomy and extension, and root/shoot biomass ratio. In order to ensure optimal water absorption and storage plants have developed different strategies as: roots with water storage cells, fibrous root system with a large surface area (typical of annuals and ephemeral desert plants) or extensive root system (typical of perennials). Furthermore it is well known that xeric plants invest a large amount of energy allocating carbon to develop the root system more than the shoot biomass (Turner and Kramer, 1980). The ratio of root/shoot biomass changes depending on plant origin and its evolutionary adaptation and on environmental conditions. Root and shoot biomass ratio of

Mediterranean plants tend to be higher than in plants of more temperate ecosystems, possibly as an adaptation to summer drought (Lloret et al., 1999; Pausas, 1999). Pace et al. (1999) observed that the tropical young cotton plants grown under drought conditions had higher root/shoot biomass ratio than control plants, as well as Barbour (1973) indicated that perennial desert plants possess a fast growing root system and a high root/shoot biomass ratio.

In the Mediterranean basin, as observed by Mehta and Yang (2008), the main rainy season extends from October to March, with maximum rainfall occurring during November–December. On the other hand, during summer temperature increases, while rainfall results scarce or, in some cases, even absent. The Mediterranean is thus a type of climate characterized by hot summer drought and wet winter.

Due to these considerations, drought can be considered as an important factor affecting plant performance and limiting the species range in these regions. Therefore water availability is also one of the most limiting factors for plant growth as it is for green roofs, especially for the extensive systems which mainly rely on precipitation events (Grant *et al.*, 2003; Dunnett and Kingsbury, 2008). Also, the global issue of water shortage, which is increasing and in some Mediterranean region is already a matter of urgency (Ministero dell'Ambiente, 1999), will presumably have influence on the roof greening expansion in these areas, unless ad hoc studies to widening the planting options for and to adjust the green roof technology to dryer environments are conducted. In Southern European countries different plant species and engineered solutions need to be identified alternatively to the ones already tested and developed in Northern countries.

Although green roofs are living systems, this artificial and stressful environment is challenging for life. Hence the use of native species has been also investigated as a planting design strategy to enhance the green roof ecological function and to ensure a better success in these difficult conditions (Hauth and Liptan, 2003; Dunnett, 2006; Licht and Lundholm, 2006; MacDonagh *at al*., 2006; Martin and Hinckley, 2007; Sutton, 2008). However, most of the published studies have been performed in non Mediterranean-type climates and the use of non-native species has to be taken in consideration since it may have positive advantages first of all the possibility to wider the plant range for these demanding environments (Dunnett and Kingsbury, 2008).

Recently, Benvenuti and Bacci (2010) published an interesting work which tested several xerophytes from the Mediterranean area for their performance on green roof-like ground-based beds. Preliminary results from this study seemed to satisfy expectations on the plant material used, most of native species performed well. However, substrate depths lower than 20 cm are not advised. The lack of findings related to the Mediterranean area suggest the needs of further vegetation studies and green roof plant evaluation trials in real roof conditions for the Mediterranean environment.

Besides the fact that environmental conditions on a roof may be significantly harsher than a natural environment, it is clear that native and adaptable plants need to be reviewed for heat and drought tolerance, also on account of the fact that most systems are designed to be low-input and low-maintenance.

The Mediterranean flora presents a high level of endemic species richness, which contributes to the maintenance of a high biodiversity in such ecosystems. In Italy there is a tendency to preserve the landscape strictly connected to its historical background and to take in consideration the use of native species for biodiversity and cultural landscape preservation (Pardossi et al., 2007). In this thesis has been considered that the species richness and the high biodiversity of Mediterranean flora have great potential which need to be evaluated and promoted.

Moreover, the importance of enhance vegetation richness and diversity on the roof has been recently better recognized for several reasons. Although, it is well-ascertained that sedums fulfil the characteristics required for shallower green roofs, mainly due to their wide variability and their related adaptability to a wide range of conditions, on the other hand the use of plants belonging to a same genus, which thus share basic and common distinctive elements, such us shape of the flowers, succulence, and susceptibility to certain pathogens (Stephenson, 1994) is not recommended. Indeed, even if is not orthodox the use of term monoculture (Snodgrass and McIntyre, 2010), avoiding planting design exclusive of sedums or of any particular genus and species is desirable, either for ecological than for more technical and esthetical reasons. Over than improving and support biodiversity, by varying vegetation may also vary the system’s performances (Dunnett *et al.*, 2005; Lundholm *et al.*, 2010) since plants have greater performances (Nagase and Dunnett, 2010).

Therefore it can be maintained that widening the range of plant choice for green roofs design is necessary, particularly in environments such as the Mediterranean.

Because of the importance of the vegetation for green roofs the plants used in the experiments here described were selected through different steps and criteria described as follows. The plant selection was focused on perennials since these are the dominant species on green roofs and may better ensure the requisite of longevity required (Snodgrass and Snodgrass, 2006).

1. The lists of species identified over the years as suitable and successful for green roof planting in Northern countries and in U.S.A. with the contribution of roof greening companies, horticulturalist and researchers were taken from Dunnett and Kingsbury (2008) and Snodgrass and Snodgrass (2006). Moreover a list of the species used by green roofs companies in Northern Italy was attempted to be done through a combination of information gained from personal professionals' interviews and the literature (Matteucci *et al.*, 1994; Abram and Salchegger, 2006; Devecchi and Assone, 2007).

2. A wide range of non commercial and native species was considered on the basis of the habitat, soil, sun exposure and watering requirements; and screened for aesthetically valuable characters (flowering season, flowers and foliage feature) and for growth capacity. They were grouped according to the life cycle (annual, biennial or perennial) and their life-form, all these information were provided by the Italian Flora (Pignatti, 1986). The final species were choosed within perennials and selected to be easy to find and to propagate. They were collected from the wild in Central Italy regions, the same area where the experiments were performed.

3. The range of commercial and native species screened for the Mediterranean climate conditions was narrowed to about 38 species for a total of about 50 taxa which were planted and further evaluated in a field collection. Evaluation was based on the observation of plant maximum growth, flowering times and plant aspect in winter and in summer.

The core of selected species and taxa was drawn on a very wide range of native and non-native species. Several criteria were adopted to screen plants and narrow the range selection, as it is also partially shown in the descriptive tables found in the following chapters. Most of these criteria are described and summarized by Dunnett and Kingsbury (2008). First of all were analyzed plant drought- and exposure-tolerant characteristics (low-mat forming, stems that root into the substrate, succulent leaves, grey-silver foliage, pubescent foliage, leaves with thick cuticle etc.) along with growth and groundcover capacity, hardiness and species origin. Secondly, visual criteria of selection were adopted such as winter season interest (foliage with attractive colour or texture/form), evergreen foliage, flowering times and duration, flower shape and colour and production of fruits with aesthetically valuable shapes.

Other criteria of selection are represented by plant growth, establishment and survival in green roofs conditions, as these are desiderable feature for the green roof success. Plant growth is estimated in different ways, since there are different measurements directly related to the plant growth. Measuring plant height, diameters (spread) or calculating a growth index is a quite diffuse method (Monterusso et al., 2005; Compton and Whitlow, 2006; Nagase and Dunnett, 2008a). Shoot dry biomass and the relative growth rate have been spreadly used to highlight plant growth performance in green roof conditions as well as under stress conditions (VanWoert *et al*., 2005; Durhman *et al.*, 2006; Nagase and Thuring, 2006; Nagase and Dunnett, 2010; Thuring *et al*., 2010) Concerning plant growth in terms of ground cover depending on the research purpose and set up some authors use a visually estimation and some others count the number of plants of each specie using transects (Monterusso *et al.,* 2005; Martin and Hinckley, 2007; Emilsson, 2008; Getter and Rowe, 2008; Benvenuti and Bacci, 2010). Another method used to determine the coverage rate is the analysis of periodical digital photographs with image analysis softwares by which it can be established the exact area of coverage of each plant (Sendo *et al*., 2010). Visual estimation is often used to detect also plant survival (Getter and Rowe, 2007).

In the present studies several growth and physiological parameters were used to describe plant response to different environmental situations. Other specific measurements which were taken and are not mentioned here will be described in the specific chapters.

The experiments presented in this chapter had the following objectives. First of all, these studies were set up to evaluate a range of potential plant taxa to be used on green roof systems in the specific context of the Mediterranean Italian climate. Secondly, they aimed to test whether native species perform better than non-native species, but also how material collected from local wild areas (wild forms) and the relative commercial material (commercial forms) would perform in green roof conditions.

2.2 EXPERIMENT 1

2.2.1 Greenhouse screening of green roofs potential perennial groundcovers for drought tolerance under Mediterranean climate conditions

ABSTRACT

In Mediterranean climates, the establishment and survival of vegetation on extensive green roofs can be limited by drought stress. The aim of this research was to test eight succulent and sixteen non-succulent taxa for their tolerance to drought. For both succulent and non-succulent species, a randomized-complete block design with three replicates was used to test the factorial combination of genotypes and irrigation scheduling (W1= watering when substrate water content was reduced by 10% of the container capacity; W2 = watering when substrate water content was reduced by 65% of the container capacity). The tolerance to drought stress was different among the non-succulent group. In general, low growing taxa tolerated longer periods of drought showing a low water loss during the time and as *Armeria maritima* also good level of plant water status. *Delosperma cooperi* and *Sedum rupestre* were more tolerant to drought stress compared to the other succulent plants. Results seem to confirm the hypothesis that low growing species are the most suitable for green roofs while partially confirm that native species can be more tolerant to drought conditions than non-native.

Keywords: drought tolerance, green roof, perennials, succulents, growth.

2.2.2 INTRODUCTION

The lightweight extensive green roofs are the wider diffuse method to grow plants on the rooftops (Snodgrass and Snodgrass, 2006). These systems require minimal maintenance, are designed for long term sustainability and are intended to be self-sustaining. Despite their lower cost and the lower capacity required for building holding, they can support a limited range of plants to grow, as they are characterized by shallow substrate depths. Specifically, extensive green roofs have usually between 2 and 15 cm of growing medium to support plant life. Thus the planting choice normally falls on succulents and some drought-tolerant forbs or grasses (Durhman *et al.*, 2007; Dunnett & Kingsbury, 2008; FLL, 2008).

However, also stonecrops in very shallow substrates without supplementary irrigation do not always survive (Durhman *et al.*, 2007), while in deeper substrates seems to do not benefit of additional water as other species do (Dunnett and Nolan, 2004; Thuring *et al.*, 2010).

In general, supplementary watering results in promoting plant growth at any depth and also deeper substrates ensure higher plant survival and establishment (Dunnett and Nolan, 2004; Monterusso *et al.*, 2005; Durhman *et al.*, 2007; Thuring *et al.*, 2010).

When selecting plants for green roofs two main factors have significant influence: the geographical climate and the severe microclimate conditions often found on rooftops (Getter and Rowe, 2008). In Mediterranean areas, where plants experience drought and high temperature during the entire summer season, plant selection is fundamental.

In shallow extensive green roofs stonecrops are thus the most successful and popular species. Their success in roof greening is mainly due to two distinctive elements: on one hand their succulent habit, which allow them to store relatively large amount of water in their tissues, on the other hand they have the potential capacity to optimize, through CAM (Crassulacean acid metabolism), the photosynthetic performance and carbon assimilation in potentially limiting environments (Cushman and Borland, 2002; VanWoert *et al.*, 2005).

Looking at the complexity and variability existing between species in CAM expression (Kluge and Ting, 1978; Cushman, 2001; and Lüttge, 2004), several studies has demonstrated that many *Sedum* and *Delosperma* species have CAM photosynthesis. Particularly, some of the species examined exhibit CAM in stressful conditions as in normal conditions, while others exhibit C_3 metabolism or CAM-cycling in normal conditions whilst they can transit to CAM-cycling or CAM when under non-optimal conditions (Gravatt and Martin, 1992; Cushman *et al.*, 1990).

Essentially, CAM metabolism is advantageous for plants since it increases the plant water use efficiency more than C_3 and C_4 metabolisms. Through this metabolism plants fix carbon during the night when environmental factors - as light and temperature - are not limiting and evapo-transpiration rates are low, resulting in the accumulation of Malic acid and decreased level of storage carbohydrates. When plants experience light they assimilate the previously stored carbon dioxide (malate form) through the Calvin-cycle behind closed stomata (Kluge and Ting, 1978; Taiz and Zeiger, 2002; Lüttge, 2004).

As mentioned before, *Sedum* and *Delosperma* over than the CAM characteristic have also some anatomical and morphological features that minimize water loss, including low surface-to-volume ratios, large cells and vacuoles with enhanced water storage capacity (i.e. succulence), and reduced stomata size and/or frequency (Cushman, 2001).

Besides the physiological adaptations to water stress (as CAM can be considered), plants may exhibit also morphological adaptations as for instance leaves pubescence, a character clearly related to the habitat aridity as well as leaves' succulence or reduced plant dimension and growth (Turner and Kramer, 1980).

Since water availability in green roof systems is limited, experiments conducted on green roof plant selection frequently concern tests for drought stress (Durhman et al., 2004; VanWoert *et al.*, 2005; Durhman et al., 2006; Nagase and Thuring, 2006; Wolf and Lundholm, 2008; Nagase and Dunnett, 2010; Thuring *et al.*, 2010). The watering treatments are generally programmed on a day basis (Durhman et al., 2004; Nagase and Thuring, 2006; Wolf and Lundholm, 2008; Nagase and Dunnett, 2010), whereas in the experiment carried out by VanWoert *et al*. (2005) more consideration was given to the soil moisture content and the watering scheduling was determined through weight and lysimetric approaches.

Declining plant water status negatively affect new growth, through limiting cell expansion, and also physiological responses such as stomatal closure and reduction of photosynthetic rates are associated with it (Taiz and Zeiger, 2002; Gimenez *et al.* 2005). Carbon fixation and supply is directly connected to plant development and growth since the CO_2 fixed through photosynthesis becomes part of the structural compounds of plant cells and tissues (Taiz and Zeiger, 2002). Due to these considerations the analyses of plant growth and of physiological responses have been widely used to measure indirectly plant water status and response to drought (Hsiao, 1973; Durhman et al., 2004; VanWoert *et al.*, 2005; Durhman et al., 2006; Nagase and Thuring, 2006; Nagase and Dunnett, 2010; Thuring *et al.*, 2010).

Several measurements were taken in the present study to detect plant response to drought: leaf relative water content, shoot dry biomass, growth index, leaf temperature, leaf chlorophylls and carotenoids content.

It is arguable that overall plant water availability and plant water content are the most important factors affecting many physiological processes which have direct effect on plant growth (Tezara *et al*., 2003; Gimenez *et al*., 2005). Transpiration affects the leaf water potential, and leaves are the plant organs where transpiration and photosynthesis mostly occurs (Taiz and Geiger, 2002; Gimenez *et al.*, 2005). The leaves relative water content is defined by Hsiao (1973) as one of the most commonly used direct indicator of plant water status as confirmed by other studies (Castillo, 1996; Ramirez-Vallejo and Kelly 1998; Jones, 2007).

Shoots dry biomass accumulation, growth index, water-use efficiency and infra-red thermometry are all considered indirect indicators of plant water status (Jones, 2007). As well as chlorophyll and carotenoids content, since water deficit affect their formation and increase the possibilities of photodamage (Tezara et al., 2003).

Shoot dry biomass and growth index are parameters which have been already used in other green roofs experiments (Durhman *et al.*, 2004; Monterusso et al., 2005). Representing the volume of the plant canopy, the growth index is a good growth parameter, it reflects, indeed, the differences in plant architecture and growth between taxa, differences which may vary when environmental conditions are limiting (i.e. drought). Monterusso *et al.* (2005) and Leclerc *et al.* (2006) used for their experiments the growth index as it is calculated by this formula (height + width at the widest point + width 90° to the first width/3). However in the present study the growth index as calculated by Sloan *et al.* (2003) was chosen, being a tridimensional value it gives a picture of the plant form and growth more detailed and close to the reality than a one-dimensional value. Leaf temperature has long been identified as one indicator of the plant water status (Hsiao, 1973; Singh and Singh, 1988; Golestani Araghi and Assad, 1998). Therefore at low substrate water potential a close association between the increase of canopy temperature and the plant moisture stress could be expected from the reduction in transpirational cooling of the plant canopy upon stomatal closure (Singh and Singh, 1988).

Thus, differences in plant growth have a physiological basis and the water status of plants influence many physiological processes as plant growth and photosynthesis. Since in green roofs substrate depth is limited and this may lead plant to experience severe environmental conditions, first of all the water deficit, the drought effect on plant growth response is analysed in order to help the understanding and the selection of species potentially suitable for the green roof application in Mediterranean climates.

Starting from the hypothesis that plants from local flora and taxa which show drought-tolerance feature can be more adaptable to the roof environment under Mediterranean climate conditions, the following objectives were pursued in this study: (1) in the present study, the response of some perennials to drought conditions in a glasshouse screening was determined in terms of biomass accumulation, chlorophyll and carotenoids content, growth index, leaf relative water content and temperature, daily transpiration and water use efficiency; and (2) to verify the potential suitability of a core of perennials for extensive green roofs in a Mediterranean climate.

2.2.3 MATERIALS AND METHODS

Study site

The experiment was conducted from August to November 2008 in a polyethylene greenhouse located at the experimental farm of Tuscia University, Central Italy (latitude 42°25’ N, longitude 12°08' E, altitude 310 m).

Plant material and growth conditions

Plant species examined in this study were selected starting from an initial investigation among the species most frequently used on green roofs by green roofing companies based in Northern Italy, where there is actually an established market for green roofs. We also evaluated species characteristics (Habitat, hardiness, drought and heat tolerance, flowering and aesthetic value) by which the selection was narrowed to about 30 taxa, and of these 24 were screened. The plant taxa chosen for the experiment are all originated from dry habitats, mostly typical of the Mediterranean flora or which tolerate drought and sun exposed conditions, and exhibit ornamental and drought-adaptive features, either through flower or foliage.

In the following table (Tab.2.1) the selected taxa are described according to Raunkiaer life form system (Pignatti, 1986) and to the criteria of selection adopted. Plant material consisted of herbaceous and woody groundcover perennials mostly native from Europe and the Mediterranean areas. Nine are native, one is endemic and three are naturalized in Italy. Furthermore, in order to simplify the discussion and to distinguish treatment’s effect on different plant types, taxa were grouped according to the drought-related morphological character of leaf succulence. Thus, in total 16 non-succulent and 8 succulents were examined.

Plants were grown under natural light conditions; ventilation was provided automatically when the air temperature exceeded 26°C. A Campbell scientific weather station connected to a CR-10X data logger provided ambient air temperature.

Plants were placed in plastic pots (Φ 14 cm). Concerning non-succulent plants each pot was covered by plastic lid of which the upper side was white to reduce heat absorption and to avoid evaporation from the substrate surface. The circular lid was fixed all around the pot side through an elastic. To adjust it around the plant stem an hole and two separate halves were made. Pots were filled with growing medium consisting of 92% pumice (Ø 2-4 mm) and 8% peat by volume, additionally 4 g/L of a slow release fertilizer (Nitrophoska Blu, Basf, Italia) was added in order to ensure and induce plants to develop their root system in a highly draining substrate, differently composed from that of cultivation. Water was delivered to the plants with a drip-irrigation system (one emitter of 2 L/h per pot). Plants were watered daily for at least 15 days before irrigation scheduling was imposed.

Experimental design

The experiment was designed, separately for succulent and non- succulent species, in a randomized split-plot layout involving species as subplot, treatments and watering levels as main-plot treatments in three replications. Each subplot consisted of five pots.

Watering treatments were two irrigation schedules: W1 where plants were watered when substrate water content was reduced by up to 10% of the container capacity and W2 where plants were watered when substrate water content was reduced by 65% of the container capacity.

Measurements

Plant transpiration (non-succulent), evapo-transpiration (succulent) and pot water content was measured by gravimetric method or potted plant weighing method (one plant per plot) (Franco and Magalhães, 1965).

The interval between two irrigation events changed according to the transpiration rates of the plants. With regard to the W2 irrigation scheduling these intervals will be referred in the discussion as drought cycles.

At the end of the experiment, two plants from each sub-plot were harvested to determine the fresh and dry weight of shoots (oven-drying for 72 h at 80°C).

Plant water use efficiency expressed as ratio between shoot dry biomass production (grams) and kilograms of water transpired or evapotranspired (succulents) was estimated.

Plant height and plant diameters were measured every 15 days on three plants per plot. Height (H) was determined as the distance from the surface of the medium to the top of the plant, while width (diameter, W) as the average of two measurements (one perpendicular to the other). According to Sloan *et al.* (2003) plant growth index (GI) was calculated as:

$$GI = \pi\left(\frac{W}{2}\right)^2 H$$

The leaf temperature was measured using an infra-red thermometer during morning times and before every irrigation event. At the same time an amount of leaves corresponding to an area of 5 cm^2 were taken from each plot to determine the relative leaf water content (RWC) according to Castillo (1996):

$$RWC = \frac{(Fresh\ weight - Dry\ weight)}{(Turgid\ weight - Dry\ weight)} * 100$$

Chlorophyll and carotenoids analysis was done at the end of trial on young-fully expanded leaves. The samples were immediately frozen in liquid N_2, and stored in a -80°C freezer until up the analysis. Chlorophyll and carotenoids were extracted in acetone solution (80% v/v) and spectrophotometrically estimated by measuring the absorbance at 470, 647 and 664 nm wavelengths with a UV-vis spectrophotometer (Beckman DU-50 spectrophotometer). Formulae and extinction coefficients used for the determination of chlorophyllous pigments (total chlorophyll and carotenoids) were described by Lichtenhaler and Wellburn (1983).

Statistical analyses

Eight species showed growth problems due to biotic stress and were not included in the analyses.

All data were statistically analyzed with SPSS 16 for Windows, 2001.

General Linear Model both Univariate and Repeated Measures were used to detect treatment effects, with percentage data arcsine square root transformed as appropriate, and back-transformed for display. Combined analysis of variance was performed using irrigation schedule and species as fixed variable and block as random.

Duncan's multiple range test was performed at P = 0.05 on each of the significant variables measured.

Correlation analysis was performed to measure whether the variables examined exhibited strong relationship or not.

Tab. 2.1: Main characteristics of the selected taxa

FAMILY	BOTANICAL NAME	FOLIAGE AND FLOWER CHARACTERISTICS	HARDINESS (USDA)	HEAT (AHS ZONE)	ITALIAN STATUS	CHOROLOGICAL TYPE	LIFE FORM	FLOWERING TIME
Brassicaceae	*Aurinia saxatilis* (L.)Devs.	Grey-green leaves, bright yellow flowers.	3 - 10	1-8	Native	Europe (incl.Turkey)	Ch. suffr	III - V
Plumbaginaceae	*Armeria maritima* Willd. 'Splendens'	Green, narrow-linear leaves, pink flowers.	3 - 9	1-9	Cultivated	-	H.ros	V - VII
Caryophyllaceae	*Cerastium tomentosum* L.	Woolly silver-gray leaves, white flowers.	2 - 7	1-7	Endemic	Southern Europe (It; Si)	Ch. suffr	VI - VIII
Aizoaceae	*Delosperma cooperi* (Hooker fil.) L. Bolus	Fine, cylindrical, deep - green **succulent** leaves, purple - pink flowers.	5 - 10	8-10	Cultivated	Southern Africa	Ch. succ	VI – IX
Aizoaceae	*Delosperma nubigenum* (Schlechter) L.Bolus	Fine, short, bright green **succulent** leaves, yellow flowers.	5 - 10	-	Cultivated	Leshoto	Ch. succ	-
Aizoaceae	*Delosperma* 'Kelaidis'	Fine, cylindrical, bright green **succulent** leaves, pink-salmon flowers	6 - 10	8-10	Cultivated	-	Ch. succ	VI - IX
Caryophyllaceae	*Dianthus deltoides* L. 'Leuchtfunk'	Dark green, lanceolate leaves, bright deep red flowers.	3 - 9	-	Cultivated	(Species :Western Europe to Eastern Asia)	H.caesp	V - IX

Leguminosae	*Dorycnium hirsutum* (L.) Ser.	Grey-green leaves, white flowers, ornamental brown fruits (seed pods).	7 - 9	-	Native	Mediterranean-Europe	Ch. suffr	V - VI
Cistaceae	*Helianthemum* 'Fire Dragon'	Grey-green leaves, light red flowers.	4 - 9	-	Cultivated	Genus native of Europe North and South America	Ch.suf fr	V
Hemerocallidacea e	*Hemerocallis* 'Stella de Oro'	Narrow-linear green leaves, yellow-gold flowers.	2 - 9	2-8	Cultivated	Genus native of Central Europe, China and Japan	Ge.	V-IX
Hypericaceae	*Hypericum calycinum* L.	Oblong, deep green leaves, yellow flowers	5 - 9	4-9	Naturalized	Med-Mount.	Ch.fru t	VII-VIII
Lamiaceae	*Hyssopus officinalis* subsp.*aristatus* (Godr.)Nyman	Narrow green leaves, violet flowers.	6 - 9	5-9	Native in most of the Italian territory	Orophyte Euroasiatic (Substeppic)	Ch suffr	VII - X
Polemoniaceae	*Phlox* 'Daniel's Cushion'	Slightly-waxed needle-like green leaves, purple-pink flowers	3 - 8	1-8	Cultivated	-	Ch suffr	IV - VI
Lamiaceae	*Satureja montana* L. subsp. *montana*	Linear-lanceolate, dark, grey-green leaves, white to pink flowers.	6 - 9	1-10	Native	Orophyte Western-Mediterranean	Ch suffr	VII-IV
Crassulaceae	*Sedum album* L.	Cylindrical, **succulent**, green leaves, white flowers.	3 - 8	1-8	Native	Mediterranean-Europe	Ch. succ.	V - VII
Crassulaceae	*Sedum kamtshaticum* Fisher and Mayer var. *floriferum* Praeg. > *Phedimus floriferus* (Praeger) 't Hart	Serrated, **succulent**, green leaves, yellow flowers.	-	1-8	Cultivated	Siberian to Central China	Ch. succ.	VIII-IX

Crassulaceae	*Sedum rupestre* L.	Blue-to grey- green **succulent** leaves, yellow flowers	3 – 9	3-7	Native	Western and Central Europe	Ch. succ	V - VII
Crassulaceae	*Sedum sexangulare*	Needle-like, tiny, **succulent**, green leaves, yellow flowers	2 - 9	1-8	Native	Central Europe	Ch. succ	V - VII
Crassulaceae	*Sedum spurium* Marchall and Bieberstein > Phedimus spurius (M.Bieb.) ‘tHart	Serrated, **succulent**, green leaves, red in winter, purple-pink flowers.	4 - 9	1-9	Naturalized	Western Asia and Caucasus	Ch. succ	VI - VII
Caryophyllaceae	*Silene schafta* Gmel. ex Hohen.	Lanceolate green leaves, pink flowers.	4 – 8	3-9	Cultivated	Western Asia	H. caesp	V - VII
Lamiaceae	*Stachys byzantina* C. Koch	Wrinkled, woolly, grey-green leaves, purple flowers.	4 - 9	1-9	Naturalized	Western Asia and Caucasus	H.scap	VI - VII
Lamiaceae	*Teucrium chamaedrys* L.	Deep green, serrated leaves, purplish-pink flowers.	5 – 9	4-12	Native	Mediterranean-Europe	Ch. suffr	V - VII
Lamiaceae	*Thymus pulegioides* L. subsp. *pannonicus* (All.) Kerguélen	Oval dark green leaves. Mauve flowers	-	-	Native	South Europe and Southern Siberia	Ch. suffr	IV - V
Scrophulariaceae	*Veronica prostrata* L.	Oblong-obtuse, serrate, green leaves, deep blue flowers.	5 – 8	5-8	Cultivated (subsp.prostrata is native)	Euroasiatic	H. caesp	III-V

USDA HARDINESS ZONES (range of average annual minimum temperatures): 1= <50°C; 2= -50°C to – 40°C; 3= -40 to -30 °C; 4 = −30 °C to −20 °C; 5= −10 °C to −20 °C; 6= −10 °C to 0 °C; 7= 0 °C to 10 °C; 8= 10 °C to 20 °C; 9= 20 °C to 30 °C; 10= 30 °C to 40 °C.

AHS (American Horticultural Society) Plant heat zones are expressed as average of number of days per year above 30°C: 1=<1; 2=1 to 7; 3= >7 to 14; 4= >14 to 30; 5= >30 to 45; 6= >45 to 60; 7= >60 to 90; 8= >90 to 120; 9= >120 to 150; 10= >150 to 180; 11= >180 to 210; 12= >210:

Life forms'legend: Ch.=Chamaephytes; Ch.frut = C. fruticose Ch.suffr = C. suffruticose; Ch.succ= C.succuent; H= Hemicryptophytes, H scap. = H. Scapose; H ros = H. rosulate; H.caesp.= H. Caespitose.

Botanical nomenclature was to reported according to the most recent information available from the following sources: Conti *et al.* (2005), Pignatti (1982), Stephenson (1994), the International Plant Name Index, the Index Synonymique de la flore de France, the Royal Horticultural Society, Italian Mycological and Naturalistic Telematic Association, USDA Germplasm Resources Information Network

Other Sources:Giles *et al.*, 1980; http://www.learn2grow.com.

2.2.4 RESULTS AND DISCUSSION

Pests and Diseases

During the experiment several taxa showed some pathological and entomological problems. *Delosperma nubigenum, Dianthus deltoides '*Leuchtfunk', *Sedum album* and *Sedum sexangulare* showed root rot symptoms caused by *Sclerotinia* spp. and *Fusarium* spp., while *Dorycnium hirsutum, Silene shafta* and *Veronica prostrata* were infested by aphis*, Hemerocallis '*Stella de Oro' by spider mites and *Stachys byzantina* by larva of *Lepidoptera*. Because of the negative impact of pests and pathogens on plant growth, the above-reported taxa were excluded from the trial. As well as *Coreopsis* 'Limerock Ruby' and *Delosperma* 'John Proffitt' were not included in the screening since plants showed stress symptoms even when, during the acclimation after the transplant, were under well watered conditions.

Plant Water Consumption and Growth relations

Water consumption of succulents and non-succulents in W2 irrigation schedule was significantly affected by plant taxa ($P=0.001$ and $P<0.001$ respectively). It is well known that the first cause of transpiration decline, as water stress develops, is the stomata closure (Hsiao, 1973, p.523; Gimenez *et al.*, 2005).

In non-succulents mean daily transpiration ranged between 71 ml/plant recorded in *Phlox* 'Daniel's Cushion' and 313 ml/plant recorded in *Thymus pulegioides* (Tab.2.2), while succulents mean daily evapo-transpiration was between 26-28 ml/plant (*Sedum spurium* and *rupestre*) and 54 ml/plant (*S. kamtshaticum*) (Tab.2.3).

Water use efficiency (WUE) was significantly different between taxa both in non-succulent ($P=0.001$) and in succulent ($P<0.01$) genotypes.

Within non-succulents *Aurinia saxatilis*, *Hyssopus officinalis, Teucrium chamaedrys* and *Thymus pulegioides* recorded the highest WUE values, while *Armeria maritima, Cerastium tomentosum, Helianthemum* 'Fire Dragon'*, Hypericum calycinum* and *Phlox* 'Daniel's Cushion' the lower. As regard to the mean number of days required to complete a drought cycle, *Phlox* 'Daniel's Cushion' and *Hypericum calycinum* had the longer cycles 8.5 and 8.3 days, on the other hand *Thymus pulegioides* cycle was the lowest (2 days).

Tab.2.2 Mean-daily transpiration value of non-succulent taxa in W2 treatment, number of days to complete a drought cycle and plant water use efficiency (WUE)

Non-succulents	Drought cycle (days)	Mean daily transpiration (ml/plant)	WUE (Kg water transpired/g shoot dry weight)
Armeria maritima 'Splendens'	6.3	110.59 b	2.45 a
Aurinia saxatilis	3	208.33 d	6.13 c
Cerastium tomentosum	6	95.00 b	2.22 a
Helianthemum 'Fire Dragon'	3.5	186.44 c	2.24 a
Hypericum calycinum	8.3	75.72 a	1.88 a
Hyssopus officinalis subsp.*aristatus*	2.5	268.85 e	5.99 c
Phlox 'Daniel's Cushion'	8.5	71.86 a	2.61 a
Satureja montana subsp. *montana*	3	214.8 d	4.12 b
Teucrium chamaedrys	3	200.14 cd	5.83 c
Thymus pulegioides subsp. *pannonicus*	2	313.67 f	5.30 c

Values not sharing a letter differ significantly at 0.05 probability level (Duncan's multiple range test).

Within succulents the higher WUE values were recorded for *Delosperma cooperi* and *Sedum rupestre*. Drought cycle duration reached 13 days for *S.rupestre* and 12 days for *S.spurium*, while *Delosperma* 'Kelaidis' had the shorter cycle (5.7 days).

Tab.2.3: Mean-daily evapo-transpiration value of succulent taxa in W2 treatment, number of days to complete a drought cycle and plant water use efficiency (WUE)

Succulents	Drought cycle (days)	Mean daily evapo-transpiration (ml/plant)	WUE (kg water evapotranspired/g dry biomass)
Delosperma cooperi	7.5	35.56 a	13.30 b
Delosperma 'Kelaidis'	5.7	48.65 b	7.60 a
Sedum kamtshaticum var. *floriferum*	6.7	54.56 b	6.48 a
Sedum rupestre	13	27.88 a	13.25 b
Sedum spurium	12	26.43 a	5.57 a

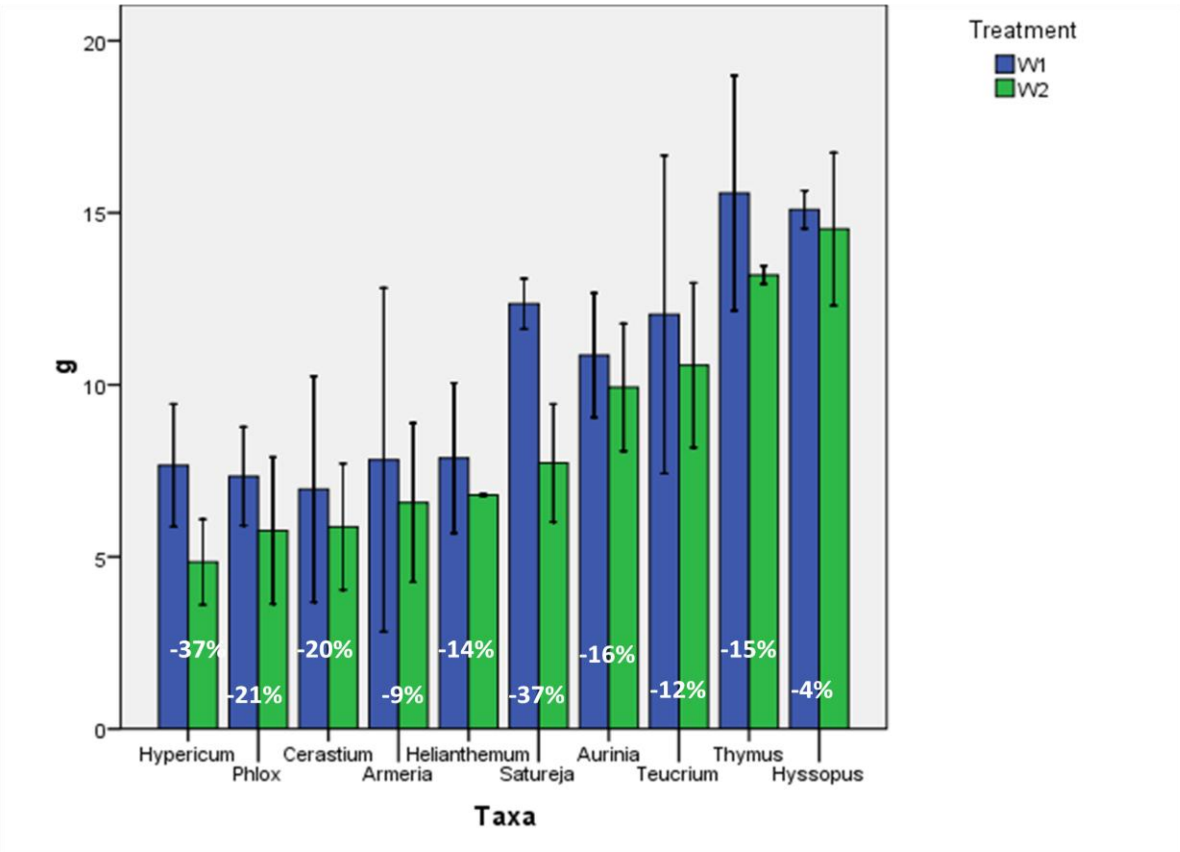
Values not sharing a letter differ significantly at 0.05 probability level (Duncan's multiple range test).

As expected the water use efficiency was in general higher in succulents, and non-succulent plants under stress conditions used more water than succulents. They lost generally earlier than the succulents the substrate moisture, even when transpiration versus evapo-transpiration values were compared.

Plant Growth

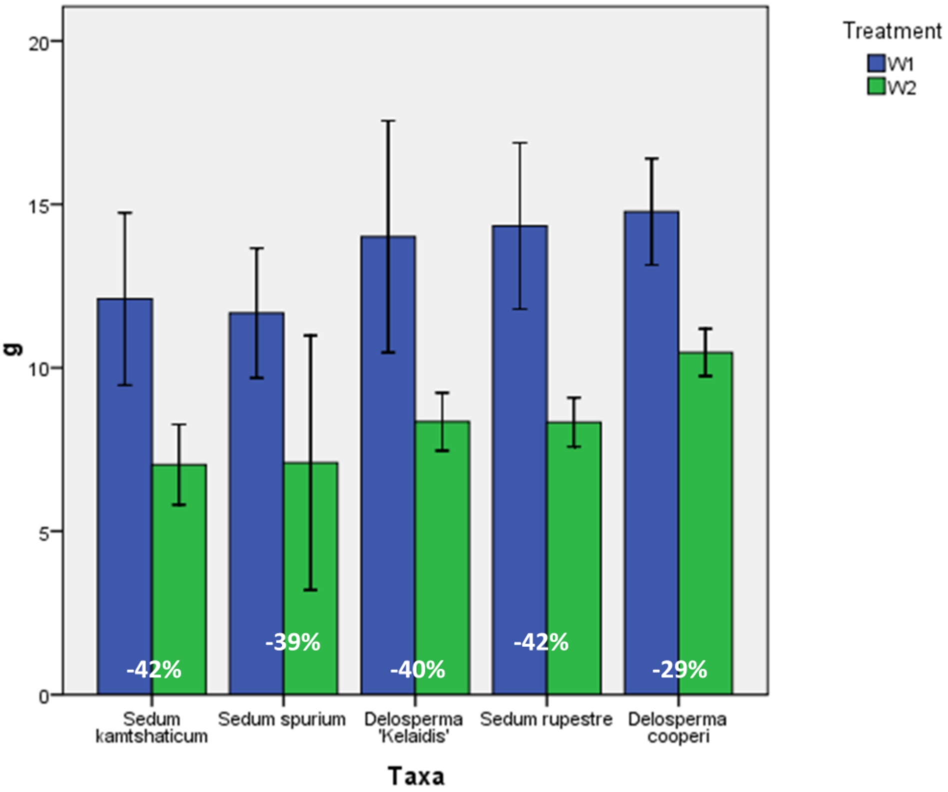
Shoot dry weight in non-succulent taxa was significantly affected by both taxa (P<0.001) and irrigation schedule (P<0.01) factors, but not by their interaction. When averaged over taxa, plant shoot dry weight varied between 10.3 g to 8.5 g in W1 and W2, respectively. When averaged over irrigation scheduling, results show different patterns in shoot dry biomass accumulation between taxa (Fig.2.2). *Hyssopus officinalis* (14.75g) and *Thymus pulegioides* (14.4g) had the greater values followed by *Teucrium chamaedrys* (11.3g), *Aurinia saxatilis* (10.4g) and *Satureja Montana* (10g), while *Helianthemum* 'Fire Dragon' (7.3g), *Armeria maritima* (7.19g), *Phlox* 'Daniel's Cushion' (6.55g), *Cerastium tomentosum* (6.41g) and *Hypericum calycinum* (6.25g) had the lowest values.

Figure 2.1: Shoot dry weight of non-succulents as affected by irrigation scheduling and percentages of biomass decrease between watering treatments W1 and W2. Error bars represent standard error.



In succulent taxa above-ground dry biomass was more significantly affected by watering treatment at P<0.001 level than non-succulents, while was less significantly affected by the factor taxa (P<0.05) and no influence was observed when the combined effect of taxa and irrigation schedules was concerned. The highest mean value of shoot dry matter was recorded in *Delosperma cooperi* (12.6g), whereas the lowest in *Sedum spurium* (9.4g) (Fig. 2.3). In W1 treatment the mean accumulation of biomass reached 13.4g whilst in W2 was 8.26g.

Figure 2.3: Shoot dry weight of succulents as affected by irrigation scheduling and percentages of biomass decrease between watering treatments W1 and W2. Error bars represent standard error.



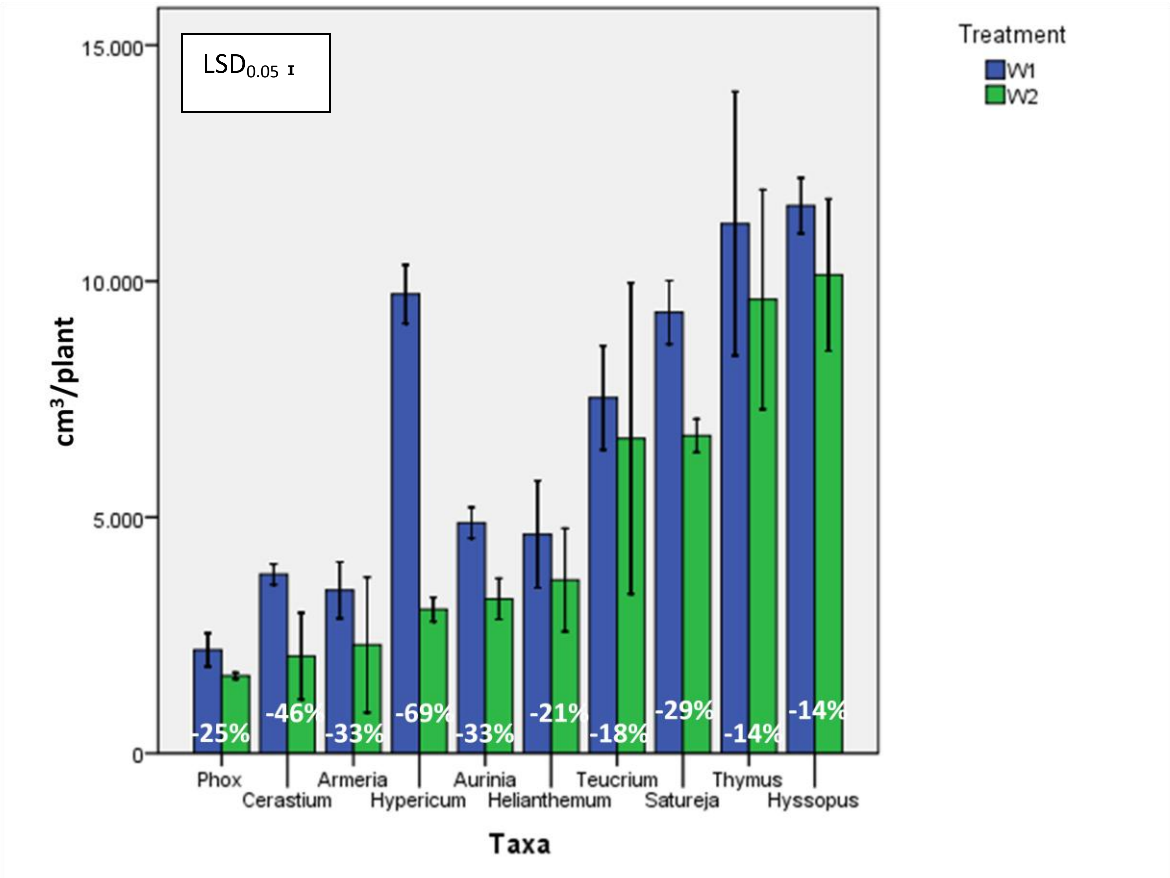
The data obtained shows evidence of plants tendency to limit shoot dry biomass accumulation when the water supply is sub-optimal (W2) especially in succulent species which were normally subjected to longer cycle of drought. Furthermore, within both plant groups the effect of plants diversity combined with drought was not significant, thus meaning that plants responded similarly to the stress imposed.

Among non-succulent group growth was influenced by both taxa and watering treatment factors at P< 0.001 level, and was also significant the effect when their interaction was concerned (P< 0.01). When averaged over taxa the mean growth index ranged between 6,836 cm^3/plant

(W1) to 4,912 cm^3/plant (W2), with a decrease of 28% caused by the drought effect. *Phlox* 'Daniel's Cushion' (1,914 cm^3/plant), *Armeria maritima* (2,876 cm^3/plant), and *Cerastium tomentosum* (2,924 cm^3/plant), recorded medially the lowest values of growth index, indicating the low growing (particularly *Armeria* and *Phlox*) and compact canopy structure which characterize these taxa. On the other hand *Hyssopus officinalis* (10,865 cm^3/plant) and *Thymus pulegioides* (10,415 cm^3/plant) had the greater growth index.

Hypericum calycinum and also *Armeria maritima* and *Cerastium tomentosum* seemed to react more strongly to sub-optimal watering levels (W2) than the other taxa, decreasing their growth by up to 68.7%, 40 % and 45.7% respectively (Fig.2.4).

Figure 2.4: Growth Index of non-succulent taxa as affected by irrigation scheduling and percentages of decrease between watering treatments W1 and W2. Error bars represent standard error.

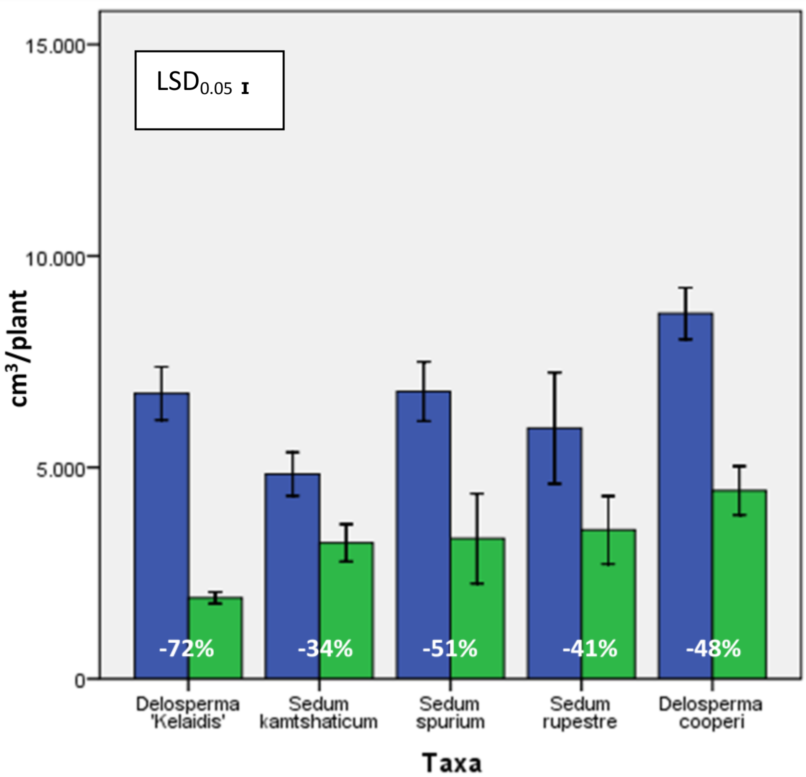


Although these three taxa all recorded low daily transpiration values their water use efficiency and growth were conversely the lowest. On the other hand *Hyssopus officinalis* and *Thymus pulegioides* growth seemed to be less affected by drought (percentage of decrease 13.7%).

The plant response to drought changes depending on species. It is arguable that short term effects of drought are less dramatic than long-term effects which involve physio-morphological changes like suberization of root surface or loss of fine roots (Pugnaire and Valladares, 2007). As plants approach to wilting point there is loss of stomatal conductance, transpiration and photosynthetic rate reduction. Hence as days of drought increase plant carbon assimilation, thus growth, tend to decline (Taiz and Zeiger, 2002). Plant respiratory response to drought differs, however at low stomatal conductance photosynthesis result to decline less than does transpiration, since decreased level of chloroplast and intercellular CO_2 concentrations are counteracted by higher atmospheric CO_2 concentration. This lead plant to increase their water use efficiency with decreasing stomatal conductance when stress is not prolonged (Lambers et al., 2008). Drought did not dramatically affect plant growth of *Thymus, Hyssopus,* and *Teucrium.* These taxa recorded higher water use efficiency values, and high transpiration rates, on the other hand they were exposed to drought for a number of days lower compared to the other taxa. As it will shown later, plant water consumption and growth index in non-succulents are positively related. The just mentioned taxa had in general canopies with higher volume and had greater biomass than the others. As a consequence their higher level of transpiration lead to reach earlier the value of 65% of the container capacity fixed for W2 watering schedule, thus they had shorter-drought cycles and the total length of study for these taxa was overall lower than 15 days and lower than 10 days when the days of watering are not counted. Hence their greater growth compared to the other species even when W2 treatment is concerned, can be explained by the fact that these plants experienced drought for relatively short times, and as it is also explained above at early stages of drought stress when stomata close the plant water use efficiency can increase (Taiz and Zeiger, 2002).

Concerning succulents, the irrigation had a great influence on growth index ($P<0.001$), also taxa influence was strong ($P<0.001$) and the two factors interaction was significant at $P<0.01$ level (Fig 2.4). When averaged over taxa the growth index values ranged between 6,590 cm^3/plant (W1) to 3,283 cm^3/plant (W2). In drought conditions *Delosperma* 'Kelaidis' exhibited the lowest growth (1,916 cm^3/plant) with a dramatic decline in growth between the two irrigation schedules (-72%), while *D.cooperi* had the highest growth (4,450 cm^3/plant). However, the lower susceptibility to drought was recorded for *Sedum kamtshaticum* (-34% from W1 to W2 treatment). It has to be noted that *S. Kamtshaticum* along with *D.*'Kelaidis' recorded the highest values of daily transpiration and also among succulents showed low water use efficiency. By contrast, while in *D.*'Kelaidis' growth response to drought stress in a relatively shorter-term was dramatic, in *S.kamtshaticum* it was not.

Figure 2.5: Growth Index of succulent taxa as affected by irrigation scheduling and percentages of decrease between watering treatments W1 and W2. Vertical bars represent standard error.



The above findings indicated that CAM species use water more efficiently than other terrestrial plants (Lüttge, 2004). When environmental conditions are limiting they continue to assimilate CO_2 by nocturnal uptake, and refix it during the day via the conventional photosynthetic carbon reduction cycle (Cushman, 2001). However, it is also true that plant growth is penalized by suboptimal environmental conditions even in plants well adapted to arid conditions (Tezara *et al.*, 2003, Gitels *et al.*, 2005).

Leaf Relative Water Content and Temperature

Leaf relative water content (RWC) was measured to control plant water status at every drought cycle's end.

RWC was significantly affected by taxa ($P<0.001$), irrigation scheduling ($P<0.001$) and their interaction ($P<0.001$) in non-succulent genotypes. When averaged over taxa RWC was 94% in W1 and 60% in W2, thus by the effect of drought leaf relative water content medially decreased by up to 34%.

Within W2 irrigation schedule the changes in relative water content between taxa were significantly different ($P<0.001$). *Hypericum calycinum* and also *Armeria maritima* showed the highest percentage of relative water content (Tab.2.4), their plant water status was less affected by drought compared to the other perennials. Conversely, *Aurinia saxatilis* (43%) was more affected by drought than the other taxa.

In succulent plants, the relative water content of leaves was also affected by both genotypes and watering treatments at $P<0.001$, but not by their combined effect. When W2 irrigation schedule was considered, there was a strong effect of taxa ($P<0.001$) on RWC. Genotypes that showed higher RWC values were *S.rupestre* (80.73%), *S.kamtshaticum* (84.9%) and *S.spurium* (85.85%), while *D.cooperi* and *D.*'Kelaidis' had the lowest (52.24% and 54.83%, respectively).

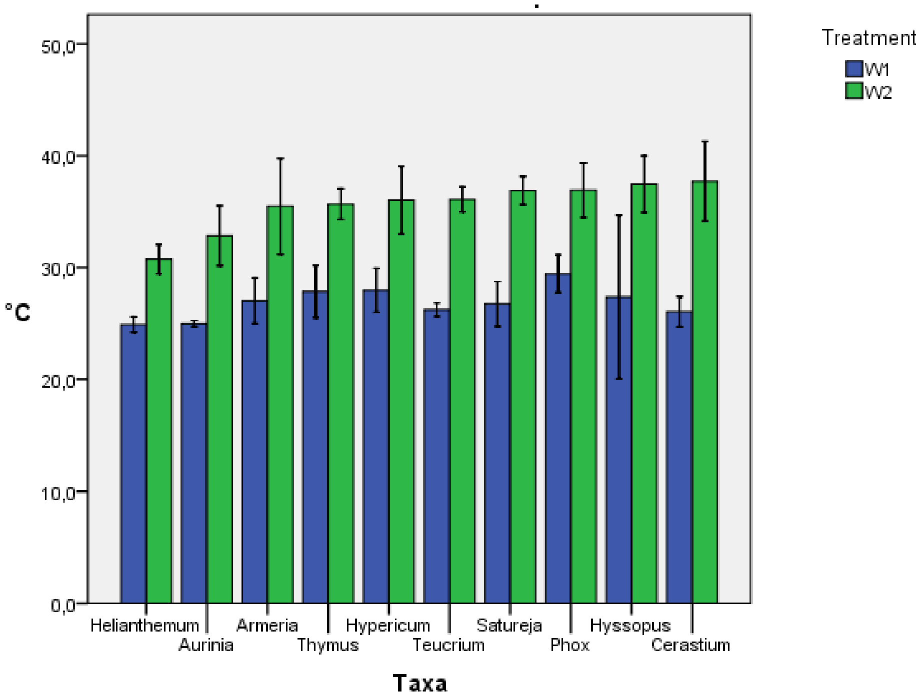
Table 2.4: Relative water content of leaves in non-succulent genotypes

Taxa	Irrigation treatment	RWC [%]
Armeria maritima 'Splendens'	W1	92.78
	W2	75.01 e
Aurinia saxatilis	W1	100
	W2	45.36 a
Cerastium tomentosum	W1	87.40
	W2	56.42 c
Helianthemum 'Fire Dragon'	W1	100
	W2	54.29 bc
Hypericum calycinum	W1	100
	W2	87.19 f
Hyssopus officinalis subsp.*aristatus*	W1	100
	W2	51.21 abc
Phlox 'Daniel's Cushion'	W1	86.69
	W2	66.49 d
Satureja montana subsp. *montana*	W1	100.00
	W2	63.80 d
Teucrium chamaedrys	W1	98.39
	W2	47.17 ab
Thymus pulegioides subsp. *pannonicus*	W1	95.15
	W2	53.04 bc
Significance	Taxa (T)	***
	Irrigation (I)	***
	T*I	***

Mean separation within rows by Duncan's multiple range test, P< 0.05

Leaf temperature of non-succulent taxa (Fig.2.6) was significantly affected by irrigation scheduling ($P < 0.001$) (Fig.2.7) and genotype ($P < 0.01$) but not by their interaction. When was averaged over taxa, the mean leaf temperature of non-succulent species increased from a minimum of 26.7 to a maximum of 35.7°C in W1 and W2 treatments, respectively. As expected leaf temperature increased with water stress. Since in drought conditions plants tend to limit water loss by closing their stomata, transpiration is absent or reduced. When transpiration is absent temperatures of leaves can rise up rapidly. Water evaporation from the leaf surface has also the role to cool down and mantain leaf's energy balance (Lambers *et al.*, 2008).

Figure 2.6: Leaf temperature of non-succulent taxa as affected by irrigation scheduling at P< 0.001 and taxa at P<0.01. Bars represent standard error.



When averaged over irrigation scheduling, the mean leaf temperature of non-succulent species was highest in *Phlox* 'Daniel's Cushion' (33.2°C) and lowest in *Helianthemum* 'Fire Dragon' (27.83°C) while the other taxa had intermediate values.

In all drought stress cycles, leaf temperature of succulent taxa was only affected by irrigation scheduling (*P*< 0.001) (Fig.2.8). When averaged over plant species, mean leaf temperature of succulents increased from 27.3 to 40.1°C in W1 and W2 treatments, respectively.

Looking at the leaf temperature detected on plants under both watering regimes, when taxa in W2 treatment reached the level of 65% of moisture content (end of a cycle and subsequent rewatering) the difference in leaf temperature between treatments reached a mean value of 8.7 in non-succulents and 13.4 °C in succulents. In the following tables (2.5 and 2.6) the mean leaf temperature as affected by irrigation scheduling in non-succulents and succulents distinguished by intervals of time between watering in W2 (drought cycle) and the air temperature recorded in the greenhouse are presented.

Table 2.5: Mean leaf temperature in non-succulents and air temperature over drought intervals and Δ temperature between watering treatments and air.

Drought Cycle	W1 [°C]	W2 [°C]	$\Delta T_{W2\text{-}W1}$	Air [°C]	$\Delta T_{W1\text{-}air}$	$\Delta T_{W2\text{-}air}$
1	26,66	35,70	9,04	34,55	-7,89	1,15
2	27,52	36,91	9,39	34,05	-6,52	2,87
3	26,42	34,01	7,60	33,70	-7,28	0,32

Table 2.6: Mean leaf temperature in succulents and air temperature over drought intervals and Δtemperature between watering treatments and air

Drought Cycle	W1 [°C]	W2 [°C]	$\Delta T_{W2\text{-}W1}$	Air [°C]	$\Delta T_{W1\text{-}air}$	$\Delta T_{W2\text{-}air}$
1	27,33	40,11	12,79	31,23	-3,91	8,88
2	27,33	43,56	16,24	31,95	-4,63	11,62
3	26,45	40,96	14,51	31,80	-5,35	9,16
4	24,85	35,05	10,21	27,28	-2,43	7,77

In W2 treatment leaf temperature was higher than the air, and succulents' reached higher level of plant canopy temperature than non- succulents (40 °C vs. 35.5°C). Moreover, differences with the temperature recorded in the air highlights that in W2 treatment leaf temperature of non-succulent taxa slightly differed from the air temperature (1.4°C), while in succulents the gap was higher (9.4 °C). Reversely, in W1 treatment the difference between leaf temperature and air temperature of non-succulent species was almost twice the one recorded in succulents (-7.2°C vs. -4°C).

Lüttge (2004) reported that the optimal temperature for CAM key enzymes is 35°C and that of decarboxylating enzymes is 53°C, thus the daytime temperatures recorded in succulents were favorable for their metabolism.

As supported by other findings (Hsiao, 1973; Singh and Singh, 1988; Golestani Araghi and Assad, 1998)the general increase in the leaf temperature in W2 treatment seems to be related to the plant water status as confirmed by negative correlation found between relative water content and leaf temperature ($R= -0.698$, $P<0.01$).

However, leaf temperature depends upon a series of factors and not only drought (Mellor, 1964; Lange, 1965). Light conditions are also closely related to this variable. Radiation load on and radiation balance of the leaf and heat convective exchanges with the air are other strongly influencing environmental factors of leaf temperature. In fact, in drought cycle 5 (late October) no differences between treatments in leaf temperature were recorded for some succulents, probably as a consequence of the low radiation conditions (data not shown).

Correlation analysis

Correlation analysis was performed between growth parameters, water consumption and water use efficiency.

Concerning succulents no significant correlation was found between the mean daily evapo-transpiration and the variables water use efficiency shoot dry biomass and growth index. The number of days to complete a drought cycle was negatively related to evapo-transpiration values ($R= - 0.82$, $P <0.05$), whilst positively related to RWC ($R=0.689$, $P<0.01$). Therefore as the dry shoot biomass increase significantly RWC decrease ($R=-0.663$, $P=0.01$). Finally, succulents' water use efficiency was not significantly correlated to any of the examined variables (Data not shown).

Concerning non-succulent perennials, as days required to complete a drought cycle decreased the shoot dry biomass ($R= -0.74$, $P<0.05$) and transpiration ($R= -0.94$, $P<0.01$) significantly increased (Tab.2.7). The amount of water transpired daily by plants was positively correlated to shoot dry biomass ($R= 0.85$, $P<0.01$) and to the growth Index ($R= 0.89$, $P<0.01$) and was negatively related to the leaf relative water content ($R= 0.66$, $P<0.05$).

Overall, these results suggested that while in succulent taxa water consumption seems mainly to depend on the plant drought-avoidance capacity, in non-succulents also shoot dry biomass and plant structure (Growth index) highly influenced the plant water use, either temporally than quantitatively. In fact, CAM species, as drought stress develops, continue to uptake and fix CO_2 (Kluge and Ting, 1978), while non CAM species stop more dramatically to accumulate biomass (as it is demonstrated by the lower water use efficiency values) until up reaching the permanent wilting point if water is not longer provided.

Tab.2.7 Statistic correlations between daily water consumption, time, shoot dry biomass, water use efficiency (WUE), relative water content (RWC) and growth index in non-succulent perennial plants.

	Drought cycle (days)	Mean daily transpiration	Shoot dry weight	WUE	RWC	Growth Index
Drought cycle (days)	1	-0.94**	-0.74*	-0,78**	0.78**	-0.76*
Mean daily transpiration (ml/plant)	-0.94**	1	0.85**	0,80**	-0.66*	0.89**
Shoot dry weight	-0.74*	0.85**	1	0,91**	-0.60	0.84**
WUE	-0,78**	0,80**	0,91**	1	-0.71*	0,71*
RWC	0.78**	-0.66*	-0.60	-0.71*	1	-0.42
Growth Index	-0.76*	0.89**	0.84**	0,71*	-0.42	1

**. Correlation is significant at the P<0.01 level and *.Correlation is significant at the P< 0.05 level

Chlorophylls and carotenoids content

Pigments are integrally related to the physiological function of leaves. Chlorophylls absorb light energy and transfer it to the electron transport system for photochemistry; carotenoids are accessory pigments which play also a role in the plant photoprotection phenomena (Taiz and Zeiger, 2002). Because of the importance of pigments for leaf function, variations in pigment content may provide valuable information concerning the physiological status of the plants. Chlorophylls and carotenoids tend to decline when plants are under stress or during leaf senescence (Gitelson *et al.*, 2003). In fact, in drought increases the formation of ROS (reactive oxygen species) which oxidize and damage also plant pigments (Egert and Tevini, 2002).

In non-succulents, chlorophyll and carotenoids contents were significantly affected by genotype, irrigation scheduling and their interaction (Tab. 2.8). Chlorophyll total and carotenoids content pattern among taxa and between irrigation scheduling can be seen in figure 2.9. Interestingly in *Phlox* ‘Daniel’s Cushion’ the contents of chlorophylls and carotenoids increased with drought stress while it declined in the other non-succulent species. Plants of this taxon generally exhibited slight chlorosis symptoms thus determining a lower content in chlorophyll and carotenoids when compared to the other species and particularly under optimal watering conditions (W1).

In non-succulent taxa, *Armeria maritima* had the highest level of chlorophyll and carotenoids content and also the lowest percentage of pigments decline from optimal to sub-optimal substrate moisture content (Fig.2.8). This result and the low RWC of *Armeria*'s leaves suggested that compared to the other taxa, plant water status and thus drought tolerance of *Armeria maritima* was greater.

In succulent genotypes, chlorophyll content was significantly affected only by irrigation scheduling ($P < 0.01$), while carotenoids content was affected by both irrigation and taxa effect ($P< 0.01$ and $P<0.05$, respectively). Both parameters were not significantly affected by the interaction between taxa and irrigation scheduling (data not shown). When averaged over taxa, the chlorophyll *a*, *b*, total and carotenoids contents increased with drought stress (2.33 vs. 5.37 mg/g DW for chlorophyll *a*; 0.94 vs. 1.99 mg/g DW for chlorophyll *b*; 3.27 vs. 7.36 mg/g DW for total chlorophyll; 2.03 vs. 5.18 mg/g DW for carotenoids content).

Table 2.8: Chlorophylls (Chl) and carotenoids content in non-succulent taxa.

Taxa	Irrigation treatment	Chl *a* [mg/g DW]	Chl *b* [mg/g DW]	Total Chl [mg/g DW]	Carotenoids [mg/g DW]
Armeria maritima 'Splendens'	W1	5.08	1.64	6.72	4.61
	W2	4.77	1.62	6.39 (5)	4.31 (6.4)
Aurinia saxatilis	W1	3.53	1.31	4.84	3.14
	W2	2.76	1.55	4.32 (10.8)	2.60 (17)
Cerastium tomentosum	W1	3.99	1.86	5.85	3.61
	W2	2.12	0.97	3.09 (47.1)	2.14 (40.6)
Helianthemum 'Fire Dragon'	W1	3.71	1.01	4.72	3.69
	W2	2.36	0.92	3.28 (30.6)	2.47 (33.1)
Hypericum calycinum	W1	2.43	0.75	3.19	2.30
	W2	2.31	0.71	3.03 (5.1)	2.41 (+4.5)
Hyssopus officinalis subsp.*aristatus*	W1	4.06	1.12	5.18	3.87
	W2	1.93	0.81	2.74 (47.1)	2.01 (48)
Phlox 'Daniel's Cushion'	W1	0.95	0.86	1.81	0.90
	W2	1.50	0.93	2.43 (+34)	1.81 (+102)
Satureja montana subsp. *montana*	W1	4.73	1.56	6.00	4.21
	W2	3.11	1.10	4.20 (30.7)	3.04 (27.9)
Teucrium chamaedrys	W1	4.51	1.44	5.95	4.30
	W2	2.50	1.00	3.50 (41.2)	2.72 (36.7)
Thymus pulegioides subsp. *pannonicus*	W1	4.60	1.35	5.95	4.54
	W2	2.37	0.96	3.32 (44.1)	2.82 (38.1)
Significance	Taxa (T)	***	***	***	***
	Irrigation (I)	***	***	***	***
	T*I	**	***	*	*
	$LSD_{0.05}$	0.15	0.10	0.28	0.28

*, **, *** significant at P≤ 0.05, 0.01, and 0.001, respectively. The percentage of reduction in water stress treatment (W2) with respect to control (W1) is reported in parenthesis.

Concerning the effect of the watering scheduling on succulents' chlorophyll and carotenoids content, the result observed is not in agreement with the literature (Alberte and Thornber, 1977; Stephen *et al*. 2008). In fact, data shows that the total chlorophyll and carotenoids contents appeared to increase in W2 treatment instead of decrease as effect of the drought stress. However, Herrera *et al.* (1991), studying the occurrence of inducible CAM in leaves of a portulacacea, observed that after 24 days of imposed drought total chlorophyll content per unit leaf dry weight was not different between stressed and watered control plants. It has also been demonstrated by several authors that CAM metabolism may have been originated not only as an adaptation to semi-arid environments but as a mechanism helping to reduce photoinhibition in plants (Griffiths *et al.* 1989; Herrera *et al.*, 1991). Moreover, Hsiao (1973) states that whether the water stress effect on chlorophyll content decrease is significant in etiolate tissues, in green leaves is not clear, and also that in some cases chloroplast isolated from severally stressed leaves contain as much or more chlorophyll as those from control. A relatively recent study conducted on *Allium schoenoprasum* confirm that in stressed plants pigments content does not change compared to well-watered plants (Egert and Tevini, 2002).

Furthermore, as mentioned before, many succulents were excluded from the trial due to root rot symptoms, even if remained taxa did not show pathological problems they could have suffered for excessive water supply. As it can be seen in the following picture (Fig.2.7), in W1 treatment the colour of *Delosperma cooperi* is extremely pale while in W2 is deep dark green.

Despite these considerations, these results need to be further investigated.

Figure 2.7: Visual effect of chlorophyll content increase from W1 to W2 treatment in *Delosperma cooperi* (succulent)

Overall considerations on taxa response to the imposed drought conditions

Considering the results obtained in this work an attempt is here made to summarize and rank taxa following those parameters taken in main consideration as descriptive of plant response in terms of drought tolerance and potential adaptability to the roof environment.

Concerning plant response to different watering regimes in non-succulents, plant tolerance to drought can be ranked on the basis of three main parameters: length of drought cycles, daily water consumption, percentage of growth reduction under W2 watering as measured by the growth index, leaf relative water content and chlorophyll content (Tab.2.9). Other parameters measured as shoot dry biomass or leaf temperature highlighted the effect of drought on plants, however plant behaviour was similar between taxa among non-succulent group.

Table 2.9 Ranking of non-succulent taxa constructed on the basis of plant response and tolerance to drought (W2 irrigation scheduling).

Non-succulents	Drought cycle (days)	Mean daily transpiration (ml/plant)	% of growth reduction in W2	RWC	% of total Chl reduction in W2
Phlox 'Daniel's Cushion'	8.5	71.86 a	25	66.49 d	+34
Armeria maritima 'Splendens'	6.3	110.59 b	33	75.01 e	5
Hypericum calycinum	8.3	75.72 a	69	87.19 f	5.1
Cerastium tomentosum	6	95.00 b	46	56.42 c	47.1
Helianthemum 'Fire Dragon'	3.5	186.44 c	21	54.29 bc	30.6
Teucrium chamaedrys	3	200.14 cd	18	47.17 ab	41.2
Satureja montana subsp. *montana*	3	214.8 d	29	63.80 d	30.7
Aurinia saxatilis	3	208.33 d	33	45.36 a	10.8
Hyssopus officinalis subsp.*aristatus*	2.5	268.85 e	14	51.21 abc	47.1
Thymus pulegioides subsp. *pannonicus*	2	313.67 f	14	53.04 bc	38.1

Phlox, Armeria and *Hypericum* maintained longer good level of plant water status as indicated by direct and physiological parameters measured and had lower daily water consumption and requirement, hence longer drought cycles which is highly desiderable for green roof plants. However *Hypericum calycinum* growth was significantly reduced and visible symptoms of stress, as leaves desiccation, were visible on plants. *Cerastium tomentosum* had also a limited growth response in W2 treatment, however drought cycle length, plant transpiration rate and the plant water conditions were acceptable. The other taxa recorded drought cycles too short and their plant water content was not optimal, therefore indicating their high water demand and requirement.

In succulents the parameters taken in main consideration in order to discriminate taxa response to limiting water conditions were: length of drought cycles, water use efficiency and growth index (Tab.2.10).

Table 2.10 Ranking of succulent taxa constructed on the basis of plant response and tolerance to drought (W2 irrigation scheduling).

Succulents	**Drought cycle (days)**	**WUE (kg water evapotranspired/g dry biomass)**	**% of growth reduction in W2**
Delosperma cooperi	7.5	13.30 b	48
Sedum rupestre	13	13.25 b	41
Sedum spurium	12	5.57 a	51
Sedum kamtshaticum* var. *floriferum	6.7	6.48 a	34
***Delosperma* 'Kelaidis'**	5.7	7.60 a	72

Overall *Sedum rupestre* and *Delosperma cooperi* seemed to tolerate and to respond better than the other succulents the W2 treatment requiring watering less frequently (longer drought cycles) and having higher water use efficiency and good growth under limiting conditions. On the other hand Delosperma 'Kelaidis' growth was significantly limited by drought and was subjected to shorter drought cycles compared to the other taxa.

2.2.5 CONCLUSIONS

It was concluded that there was variability among succulent and non-succulent species in terms of response to drought. However, as observed also by other authors (Monterusso *et al.*, 2005; Rowe *et al.*, 2005) native species do not necessarily perform as expected in stressful conditions.

The physiological parameters measured confirmed the presence and the effects of drought stress on plants with an increase in leaf temperature, a decline in leaf relative water content and chlorophyll and carotenoids content when plants were exposed to drought stress. As regards to leaf chlorophylls and carotenoids content it has to be specified that when this was generally reduced in non-succulent plants by drought stress, conversely in succulent plants was increased.

Succulents were more drought tolerant than non-succulent taxa in terms of greater water use efficiency shoot dry biomass accumulation, and lower capacity to loss water as proved by their high leaf relative water content in sub-optimal watering regime. Although succulent taxa seemed to maintain similarly plant water status (RWC) in drought conditions, they responded differently to drought when growth, mean daily evapo-transpiration and water use efficiency were concerned.

From the results it can be thus concluded that, among succulent taxa, *Delosperma cooperi* and *Sedum rupestre* were the most tolerant to drought, where *S.rupestre* was also generally subjected to longer drought cycles (days occurred between rewatering).

It was shown that the effect of drought stress on plant growth was different among the non-succulent taxa. Specifically, *Hypericum calycinum* (-68%) showed a dramatic growth declining when substrate moisture conditions were limiting. On the other hand this taxon and *Armeria maritima* recorded high values of leaf relative water content, therefore their plant water status seemed was greater than others species. However, it has to be said that *Hypericum calycinum* showed severe symptoms of stress (i.e. leaf necrosis and new leaf chlorosis). *Thymus pulegioides* subsp. *pannonicus* and *Hyssopus officinalis* subsp. *aristatus* had greater growth and water use efficiency compared to the other taxa. Even so, it is important to be aware that plant which showed to have low daily transpiration rates would be preferable for green roof planting, since watering cannot occur daily or at very short-time intervals. Despite not always having high level of growth in drought conditions, taxa that had low water consumption rates and higher level of leaf water content were: *Armeria maritima* 'Splendens', *Phlox* 'Daniels' Cushion', and the endemic species *Cerastium tomentosum*. This suggest that these plants, similarly to CAM species, have some physiological mechanism or morphological characteristics of adaptation to drought, which help them to tolerate longer drought exposition and limit water loss (i.e. leaf colour and pubescence in *Cerastium*). Moreover, *Armeria maritima* has been confirmed to be drought tolerant by recent experiments conducted in glasshouse conditions in UK (Nagase and Dunnett, 2010).

Results seems also to suggest that long-term drought-tolerant species, able to limit their growth while preserving plant functionality and vitality in limiting conditions, can be the most suitable for green roofs, and partially confirm that native species can be more tolerant to harsh conditions than non-native. Moreover, the information obtained in terms of drought tolerance of the tested species provide a better knowledge of these ornamental perennial groundcovers, part of which can be potentially used in green roof planting. However, this experiment considered plant response to drought in greenhouse environmental conditions, which can be different from the ones found on rooftops.

2.3 EXPERIMENT 2

2.3.1 Evaluation of potential perennials in a Mediterranean climate in roof top conditions

ABSTRACT

Because green roofs are frequently subjected to stress factors such as wind speed, heat, drought etc., they can represent an extreme environment for plant survival. Good results might be achieved utilizing native species in the planting design, since those are well adapted to the local environmental conditions and may play a considerable ecological role. The aim of the present work was to evaluate whether or not a range of native and non-native perennial species would survive in green roof conditions in a Mediterranean climate. During the trial, the mean height and spread of each individual was recorded, together with flowering performance and % vegetation cover. In addition the numbers of all spontaneous colonised species were recorded. . Results pointed out interesting differences in plant performance among the selected taxa. Overall sedums confirmed to be the best planting option for extensive green roofs in a Mediterranean environment. Nevertheless, there are other potential perennials which can be also considered for the planting design for 8 cm as in 15 cm deep systems. Finally, local genotypes and native species seemed to respond positively, with relatively fast and good establishment on the roof.

Keywords: green roofs, perennials, native species, Mediterranean climate, sedums

2.3.2 INTRODUCTION

As an ecological design landscape strategy (Calkins, 2005) green roofs offer many advantages, especially on an urban scale. In cities they provide new green areas and reduce the volume of water runoff, heat urban island effect and air and water pollution while also increasing biodiversity. With regard to buildings, green roofs contribute to lower energy consumption and potential economic benefits (Dunnett and Kingsbury, 2008).

The widely-known advantages of green roofs, along with the European policy supporting urban environmental sustainability, have encouraged the expansion of the roof greening strategy in several countries. Despite the growing interest, and research on green roofs, this greening technology in Italy is relatively new and the scientific findings are deficient (Fioretti *et al.*, 2010). Although green roofs have been proven to be efficient as a passive cooling building technology, especially in warmer climates (Theodosiou, 2003), the higher construction cost and the scarcity of quantitative data supporting the effectiveness of green roofs have discouraged the expansion of green roofs in the Italian construction industry.

Studies carried out on plant survival and selection for green roofs have mostly been performed in northern regions or in other continents such as America (Durhman *et al.*, 2004; Emilsson and Rolf, 2005). Besides which the plant selection (particularly for extensive systems) has often been focused on the Crassulacean genus *Sedum* (Durhman *et al.*, 2004; Emilsson and Rolf, 2005). In fact, in extensive systems the planting choice falls almost on succulents' genera such as *Sedum* spp., *Phedymus* spp. and *Hylotelephium* spp. (Emilsson, 2008). Particularly, the success of these genera on the roof is due to the fact that sedums are a colonizing species generally associated with poor and coarsely textured soils and often characterized by a peculiar photosynthetic metabolism (CAM), which helps plant to tolerate drought stress. However, a green roof plan must consider also other species in order to diversify the selection and improve the ecological and the functional value of the construction (Dunnett and Kingsbury, 2008; Dunnett *et al.*, 2008b, Oberndorfer *et al.*, 2007).

The lack of information on green roof performance, green roof plant selection and survival in Mediterranean areas and in Italy suggests a need to further investigate these fields.

As it is logical regarding the lighter green roof systems - due mainly to the shallow substrate depth and the harsh rooftop conditions - plants must tolerate drought, particularly in hot and dry climates (Koehler, 2003; White and Snodgrass, 2003; Monterusso *et al.*, 2005). On the other hand, as Simmons *et al.* (2007) suggested, it may be possible to look at the extreme environment represented by the roof not as a problem to solve trying to find the most adaptable species, but instead could also be regarded as the perfect artificial environment which recreate some biological and ecological conditions lost at the ground level. From both these stand points, the use of native plants on extensive green roofs is expected to be successful, since these are presumably well-adapted to the local environment and are found in naturally harsh habitats.

Within the selection criteria which moved to test certain species rather than others were represented by fast and good establishment, substrate coverage, and low mortality, important requirements which plants have to fulfil to be selected for green roof planting. Factors which directly or indirectly influence these characteristics have been identified and investigated over the time of which substrate depth, substrate composition, watering regime and establishment method are the most important. Due to these considerations, researcher have tested plant taxa in roof conditions looking at these characteristics (Koehler, 2003; Durhman *et al.*, 2004; Emilsson and Rolf, 2005; Monterusso *et al.*, 2005; Nagase and Thuring, 2006; Getter and Rowe, 2008; Martin and Hinckley, 2007) and as Monterusso *et al.* 2005 clearly explained the justification has to be found mostly in economical reasons. A fast growth is important both for economic and practical reasons. Plants which are able to cover the substrate faster and have higher survivability are cost effective, as the number of plants required is lower (i.e. plant density and replacement for dying); as well as a good ground cover reduce risks of spontaneous colonization and may also ensure the system greater performance in terms of stormwater retention (Nagase and Dunnett, 2008) and thermal mitigation (Nyuk Hien *et al.*, 2007). Plant selection for this study was mostly orientated to native species typical of the Mediterranean flora to test their suitability to roof top conditions. Moreover, it also tested plant material collected from the wild. Both native and non-native taxa, commercial and non-commercial material was pre-selected based on the criteria described in the introduction of chapter 2 and, furthermore, on the results obtained from the experiment 1 described in the previous chapter 2.2. As this experiment was partially funded by Harpo-Seic green roofs division, plots layout and dimensions, substrate depth and some of the species used (Phanerophytes on 15 cm substrate depth) were agreed according to the company requirements and to the space available.

Research hypotheses were that native plant and local plant material would perform better than non-native and commercial material, and that taxa proved to be drought-tolerant in the previous screening would confirm their suitability for green roofs application when grown in rooftop conditions. The main task of this work was to collect data on plant growth in roof condition under a Mediterranean climate that will provide building professionals with more information about green roof plant selection and plant performance on green roofs in central Italy.

To verify the current hypotheses a range of native and non-native taxa were grown in green roofs plots, where two different substrate depth were concerned (8cm and 15cm). Plants tested were divided in groups according to their life form and the substrate depth in which they were planted and were compared for growth, ground coverage and survival. MATERIALS AND METHODS

Identification of plant species

The criteria adopted to select plants are described in the introduction of chapter 2 and the steps adopted to identify the plant species used in this study are here summarized as follows: 1) initial investigation among the most frequently used groundcovers on green roofs in northern Italy and Europe- where there is actually an established market for green roofs – which narrowed the selection to about 30 perennials taxa; 2) 26 of these were screened in a greenhouse for drought tolerance at Tuscia University during 2008 (Experiment 1, Subchapter 2.2); 3) in the same year wild species were collected from several habitats with conditions similar to those found on the extensive green roof systems (shallow and rocky substrates, sunny exposure, drought, etc.) and along with the 30 commercial ones were placed in a field collection to observe their agronomical and aesthetic features (i.e. winter aspect, flowering time and abundance, *etc*.); 4) finally from the results obtained 19 taxa were selected to test their growth and survival on rooftop conditions. Additionally, to fulfil the research agreement with Harpo-Seic two native phanerophytes species were chosen within the Mediterranean maquis flora: *Cotinus coggygria* and *Prunus mahleb*.

To simplify the discussion the groundcover perennial taxa tested in this study were divided into three groups: Sedums, Perennials, and Subshrubs. The deciduous Phanerophytes used will be named as Shrubs.

Experimental Site

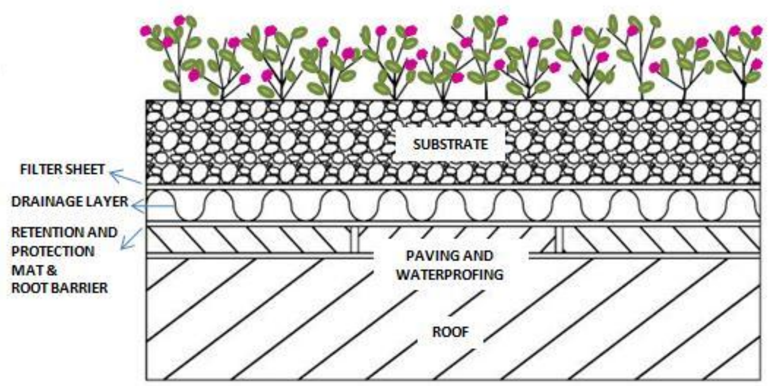
The experiment was set up in June 2009 on a flat roof of a two-story building at Tuscia University (at latitude of N42˚ 26’, at longitude E12˚ 40’, and altitude of 310 m), Viterbo, Italy. The roof is about 438 m^2. Plots were placed in two lines along the longer side of the building.

Daily weather observations were made at Tuscia University’s Experimental Farm “Nello Lupori”- at about 2 km distance from the roof site- by using a scientific automatic weather station with measurements logged to a CRX-10 data logger (Campbell Scientific).

During almost two years of study the climatic pattern was typically of the Mediterranean, characterized by dry and hot summer and wet winter.

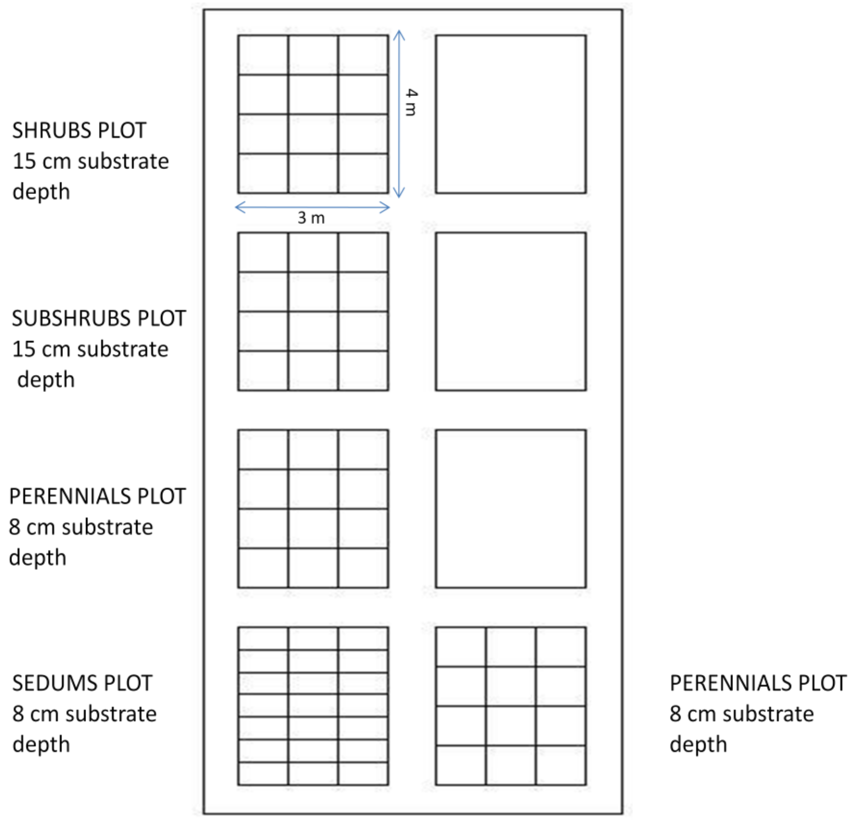
The plots consisted of a multiple-layer green roof system (Harpo-Seic, Italy) placed into a timber-made structure 4mx3m. As it is shown in figure 2.9 From the bottom to the top each one contained : root barrier, retention and protection layer, plastic drainage, geotextile filter, and growth medium composed of 92% v/v of mineral soil (crushed bricks, expanded clay and pumice) mixed with organic matter (peat) according to Italian standards UNI 11235:2007.

Figure 2.9: Cross section of the green roof system structure adopted for the trial



The substrate thickness varied between plots. In total there were 5 plots (fig.2.10): two semi-extensive (15 cm substrate depth) of which one was planted with subshrubs and one with shrubs; three extensive (8 cm substrate depth) of which two contained groundcover perennials and one contained stonecrops (sedums).

Figure 2.10: Diagram of the experimental roof and plots layout



Plant materials

In total 18 plant taxa were planted: 8 perennials of which one endemic and one native (Tab.2.11); 4 native *Sedum* species (Tab.2.12); 4 subshrubs of which 3 were native (Tab.2.13), and 2 native shrubs (Tab.2.14). The experiment was designed according to a single-factor completely randomized layout where each plant group was considered separated to the others because of plant diversity and also substrate depth diversity and the number of replication for each subplot was 3 except for shrubs of which subplots were replicate 6 times. Excepting the Crassulacean-planted plot, the vegetated plots were each divided into 12 squares (subplot of 1m x 1m). Each taxon was randomly assigned and planted into 3 mono-species quadrats and each plot was planted with a total of four species. The two shrubs species chosen were assigned to a total of 6 subplots (quadrats). Seven genotypes of *Sedum*, including four local ecotypes and three commercial genotypes, each of which was planted in 3 randomly assigned mono-species rectangles, 1 m x 0.57 m. Number of individuals per subplot (planting density) is described below.

Sedums and perennial plants, provided in pots 7x7x7cm, were planted at a density of 20 plants/m^2, excepting *Helianthemum* 'Fire dragon' and *Eragrostis spectabilis* which were planted at a densities of 16 and 12 plants/m^2, respectively. Subshrubs were provided in pots 14 cm Φ and planted at a density of 8 plants per m^2. Finally, shrubs were planted from 9x9x9cm pots at the density of 3 plants/m^2. The planting density varied from species to species according to the densities normally utilized by various green roof companies and to their growth capacity.

Table 2.11: Characteristics of Perennials tested on 8cm substrate depth

FAMILY	BOTANICAL NAME	FOLIAGE AND FLOWER CHARACTERISTICS	HARDINESS (USDA)	HEAT (AHS ZONE)	ITALIAN STATUS	CHOROLOGICAL TYPE	LIFE FORM	FLOWERING TIME
Plumbaginaceae	*Armeria maritima* Willd. 'Splendens'	Green narrow - linear leaves pink flowers.	3 - 9	1 - 9	Cultivated	-	H.ros	V - VII
Caryophyllaceae	*Cerastium tomentosum* L.	Woolly silver - gray leaves, white flowers.	2 - 7	–1 - 7	Endemic	Southern Europe (It; Si)	Ch. suffr	VI - VIII
Aizoaceae	*Delosperma cooperi* (Hooker fil.) L. Bolus	Fine, cylindrical, deep - green leaves purple - pink flowers.	5 - 10	–8 - 10	Cultivated	Southern Africa	Ch. succ	VI - IX
Poaceae	*Eragrostis spectabilis* (Pursh) Steud.	Light - green leaves, red - purple to tan in autumn and winter.	4 - 9	–3 - 10	Cultivated	Northern America	H.scap	VIII-IX
Cistaceae	*Helianthemum* 'Fire Dragon'	Grey - green leaves, light - red flowers.	4 - 9	-	Cultivated	Genus native of Europe North and South America	Ch.suffr	V
Polemoniaceae	*Phlox* 'Daniel's cushion'	Slightly-waxed needle-like green leaves, purple-pink flowers	3 - 8	1 - 8	Cultivated	-	Ch suffr	IV - VI
Lamiaceae	*Thymus longicaulis* C.Presl. (wild collected)	Narrowly elliptic green leaves, rose-purple flowers.	5 - 10	-	Native	Western Asia and Southern Europe	Ch suffr.	IV - VIII
Lamiaceae	*Thymus serpyllum* L.	Small oval-shaped green leaves, pink, pale mauve flowers.	4 - 9	–1 - 9	Cultivated	Northern Europe	Ch suffr	IV - VIII

*

Table 2.12: Characteristics of Sedums tested on 8cm substrate depth

FAMILY	BOTANICAL NAME	FOLIAGE AND FLOWER CHARACTERISTICS	HARDINESS (USDA)	HEAT (AHS ZONE)	ITALIAN STATUS	CHOROLOGICAL TYPE	LIFE FORM	FLOWERI NG TIME
Crassulaceae	*Sedum acre* L. (wild collected)	Needle-like, tiny, bright-green succulent leaves, yellow flowers.	2 - 9	1 - 8	Native	Europe and Caucasus	Ch. succ.	V - VII
Crassulaceae	*Sedum album* L.	Cylindrical green leaves, white flowers.	3 - 8	1 - 8	Native	Mediterranean-Europe	Ch. succ.	V - VII
Crassulaceae	*Sedum album* L. (wild collected)	Cylindrical green leaves, white flowers.	-	-	Native	Mediterranean-Europe	Ch. succ.	V - VII
Crassulaceae	*Sedum rupestre* L.	Blue-grey- green leaves, yellow flowers	3 – 9	3 - 7	Native	Western and Central Europe	Ch. succ	V - VII
Crassulaceae	*Sedum rupestre* L. subsp. *rupestre* (wild collected)	Grey-green leaves, yellow flowers	-	-	Native	Western and Central European	Ch. succ	V - VII
Crassulaceae	*Sedum sexangulare*	Needle-like, tiny, green leaves, yellow flowers	2 - 9	1 - 8	Native	Central Europe	Ch. succ	V - VII
Crassulaceae	*Sedum sexangulare* L. (wild collected)	Needle-like, tiny, green leaves, yellow flowers	-	-	Native	Central Europe	Ch. succ	V - VII

*

Table 2.13: Characteristics of Subshrubs tested on 15cm substrate depth

FAMILY	BOTANICAL NAME	FOLIAGE AND FLOWER CHARACTERISTICS	HARDINESS (USDA)	HEAT (AHS ZONE)	ITALIAN STATUS	CHOROLOGICAL TYPE	LIFE FORM	FLOWERING TIME
Asteraceae	*Helichrysum italicum* (Roth.)G. Don subsp. *italicum* (wild collected)	Grey - silver narrow leaves, yellow flowers.	7 - 11	7 - 10	Native	Southern Europe	Ch. suffr	V - IX
Asteraceae	*Helichrysum italicum* subsp.*microphyllum* (Willd.) Nyman	Silver woolly narrow leaves, yellow flowers.	7 - 11	7 - 10	Endemic	Western Mediterranean	Ch. suffr	V-IX
Lamiaceae	*Hyssopus officinalis* subsp.*aristatus* (Godr.)Nyman	Narrow green leaves, violet flowers.	6 - 9	5 - 9	Diffuse (probably native)	Orophyte Euroasiatic (Substeppic)	Ch. suffr	VII - X
Lamiaceae	*Teucrium* x *lucidrys*	Deep green, slightly waxed, serrated leaves, purplish-pink flowers.	-	-	Cultivated	-	Ch. suffr	V - VII

*

Table 2.14: Characteristics of Shrubs tested on 15cm substrate depth

FAMILY	BOTANICAL NAME	FOLIAGE AND FLOWER CHARACTERISTICS	HARDINESS (USDA)	HEAT (AHS ZONE)	ITALIAN STATUS	CHOROLOGICAL TYPE	LIFE FORM	FLOWERING TIME
Anacardiaceae	*Cotinus coggygria* Scop.	Suboval leaves	4 - 9	3 - 9	Native	Eastern Europe, Central Asia, China	DPh.	V – VI
Rosaceae	*Prunus mahleb* L.	W.	-	-	Native	Southern Europe	DPh.	IV - V

*****TABLES' LEGEND and SOURCES:*

USDA HARDINESS ZONES (range of average annual minimum temperatures): 1= <50°C; 2= -50°C to – 40°C; 3= -40 to -30 °C; 4 = −30 °C to −20 °C; 5= −10 °C to −20 °C; 6= −10 °C to 0 °C; 7= 0 °C to 10 °C; 8= 10 °C to 20 °C; 9= 20 °C to 30 °C; 10= 30 °C to 40 °C.

AHS (American Horticultural Society) Plant heat zones are expressed as average of number of days per year above 30°C: 1=<1; 2=1 to 7; 3= >7 to 14; 4= >14 to 30; 5= >30 to 45; 6= >45 to 60; 7= >60 to 90; 8= >90 to 120; 9= >120 to 150; 10= >150 to 180; 11= >180 to 210; 12= >210:

Life forms' legend: Ch. =Chamaephytes; Ch.frut = C. fruticose Ch.suffr = C. suffruticose; Ch.succ= C.succulent; H= Hemicryptophytes, H scap. = H. Scapose; H ros = H. rosulate; H.caesp. = H. Caespitose.

Botanical nomenclature was to reported according to the most recent information available from the following sources: Conti *et al.* (2005), Pignatti (1982), Stephenson (1994), the International Plant Name Index, the Index Synonymique de la flore de France, the Royal Horticultural Society, Italian Mycological and Naturalistic Telematic Association, USDA Germplasm Resources Information Network

Other Sources: Giles *et al.*, 1980; http://www.learn2grow.com.

Climatic conditions and irrigation management

During the establishment period the plots were watered when the substrate moisture content decreased to approximately 20% on gravimetric dry weight. The substrate moisture content was regularly checked with 2 sensors (Irrometer Company, Inc., California) for each plot. Sensors were calibrated through the pot gravimetric method, thus each amount of substrate moisture loss was related to the level of soil moisture content measured with sensors and to the estimated values of the substrate field capacity (28,5% of dry weight) and the wilting point (11,4% of dry weight) estimated through laboratory tests. When the probes reached the 20% of moisture content the content of water available for plants was reduced approximately by 50%.

During the second year of experimentation regular irrigation was interrupted and substituted by only emergency water supply. During summer 2010 plants required watering due to very dry conditions and all plots were watered two times at interval of 50 days (beginning of July and mid-August), excepting the sedum one which was watered only once in June.

Concerning the local climatic conditions during summer 2009, from July to August, the average of mean daily maximum temperature was 33.8 °C, total rainfall was 25 mm distributed during the period and never reaching values higher than 11 mm each 10 days, while the crop reference total evapo-transpiration during the same period was 302.9 mm (Fig.2.11). In winter 2009-2010 the total amount of rainfall recorded was 493.6 mm, while evapo-transpiration was 129.7 mm and minimum temperature average was -1.75 °C. In 2010 from July to August the average of maximum temperature was 33.5 °C, the total rainfall was 90 mm, of which 80 mm were recorded only during the last decade of July, while the total evapo-transpiration over the two months was totally 442.5 mm (Fig.2.12).

Figure 2.11: Average of maximum temperature and difference between rainfall and evapo-transpiration as recorded in Viterbo during 2009

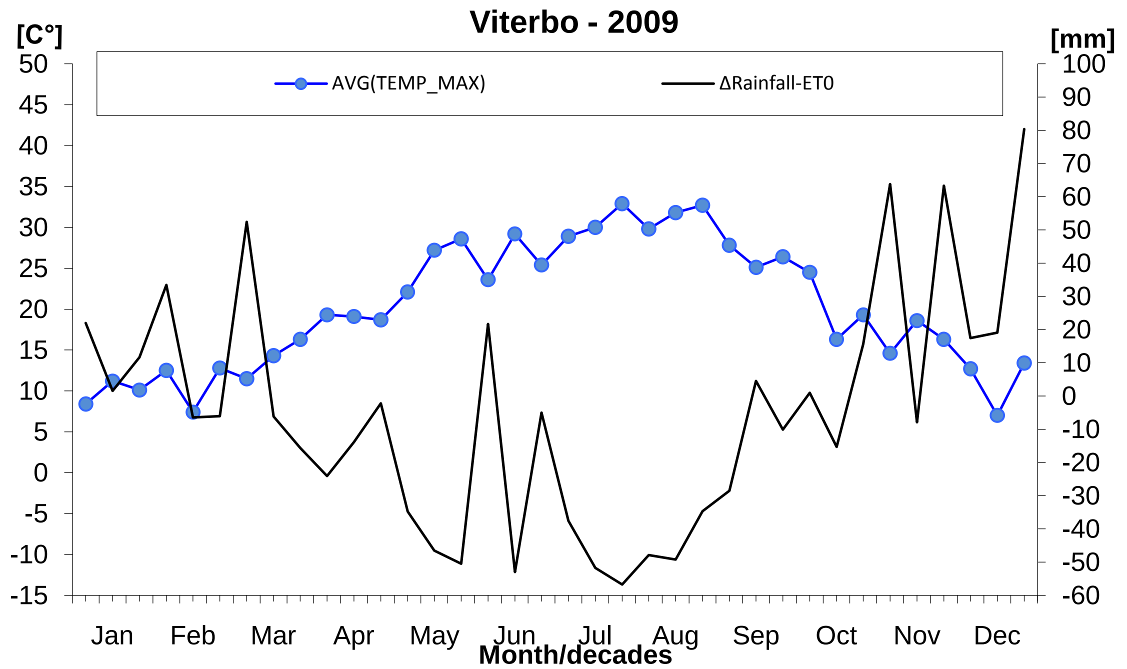
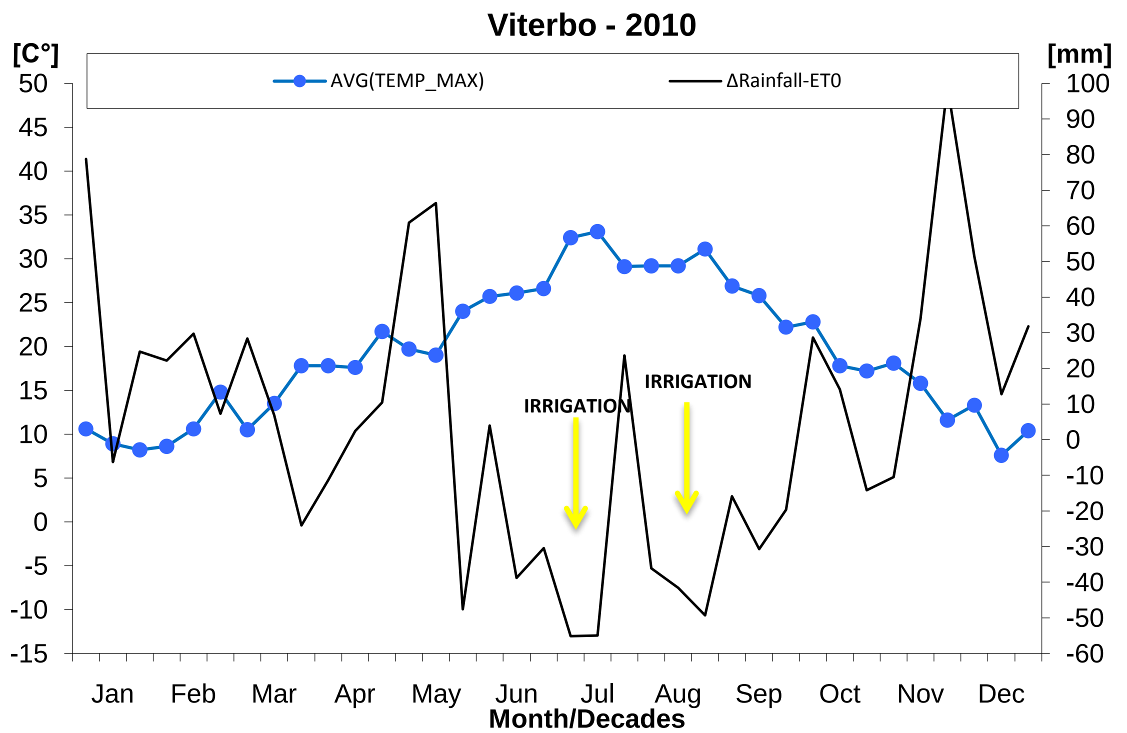


Figure 2.12: Average of maximum temperature and difference between rainfall and evapo-transpiration as recorded in Viterbo during 2010



DATA COLLECTION AND MEASUREMENTS

Plant growth

Plant height and plant diameters were measured on three plants per square (3 samples for each replicate), on a total of 9 plants per species. Height (H) was determined as the distance from the surface of the growth medium to the top of the plant, width (diameter, W) as the average of two measurements (one perpendicular to the other). Plant growth index (GI) was calculated as:

$$GI = \pi \left(\frac{W}{2} \right)^2 H$$

The percentage of ground green cover was visually estimated every month. To minimize the subjectivity of the method, the planting plots were photographed with a digital camera located 2m from the roof surface. Each square has been photographed individually with a centimetre ruler as reference and then, the area of green coverage was measured by computer processing. Every square was divided into 20cm x 20cm squares helping to calculate the % of coverage of each taxon. It was not possible to take this measurement on shrubs because of the excessive height of plants.

The percentage survival of each taxon was assessed in summer 2010, while the number of spontaneous colonizing species was recorded regularly from June 2009 to June 2010. Seedlings of the self-sowing taxa were also recorded. After the recording these plant were removed from the plots so as not to influence plants growth and coverage.

Flowering was assessed seasonally through the following scale: flowering was considered to have started when 30% of plants had at least one flower in bloom; flowering when 50% of plants had at least one flower in bloom; full flowering when more than 70% of plants had more than one flower blooming, finally flowering was considered finished when more than 70% of plants had no flowers blooming remained. The number of flowers per shoot was counted in 2010 on S*edum* species on 10 shoots per species.

Statistical analyses

All data was statistically analyzed as one-way ANOVA using GLM (General Linear Model) procedures of SPSS software package (SPSS 16 for Windows, 2001). Concerning sedums a comparison of commercial material vs. ecotypes was performed with independent samples T-test analysis.

Data expressed as percentages were appropriately transformed with arcsine square root formula, and back-transformed for display. Duncan's multiple range test was performed at P = 0.05.

2.3.3 RESULTS AND DISCUSSION

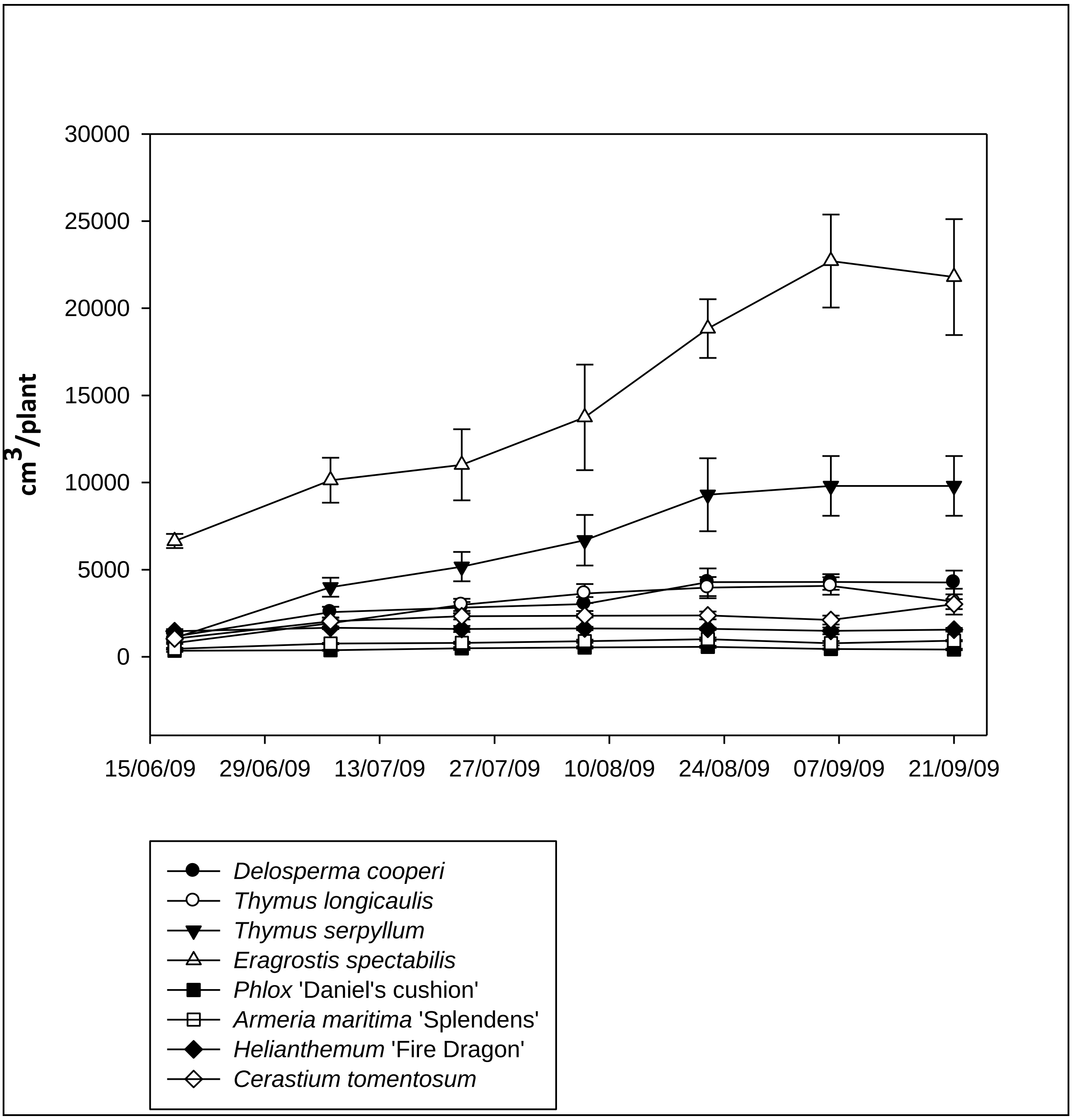
Plant Growth

Figure 2.13 shows the various growth patterns over the time and the different plant structures between perennial genotypes. *Eragrostis spectabilis* had the highest values of growth index. This taxon can be defined as groundcover but is also characterized by tall canopy that can reach a height of 0.6 m (Darke, 1999). *Thymus serpyllum*, even though is a groundcover, had a taller canopy compared to the other perennial groundcovers (excepting *Eragrostis*) and consequently had a higher growth index.

The most significant data shown concerns *Armeria maritima* 'Splendens', *Helianthemum* 'Fire dragon' and *Phlox* 'Daniel's cushion'. In fact, the growth of these taxa was limited and static compared to the others, while a greater growth response was expected considering the regular watering initially provided. Probably, these taxa tolerated less the transplant shock and simply required a longer time to establish than the other taxa, and also their growth in width (spread) capacity is lower (http://www.learn2grow.com).

By the end of September 2009 it was not possible to determine the growth index on the following species, since it was no longer possible to distinguish individual plants from each other: *Cerastium tomentosum*, *Delosperma cooperi*, *Thymus serpyllum* and *Thymus longicaulis* (wild form).

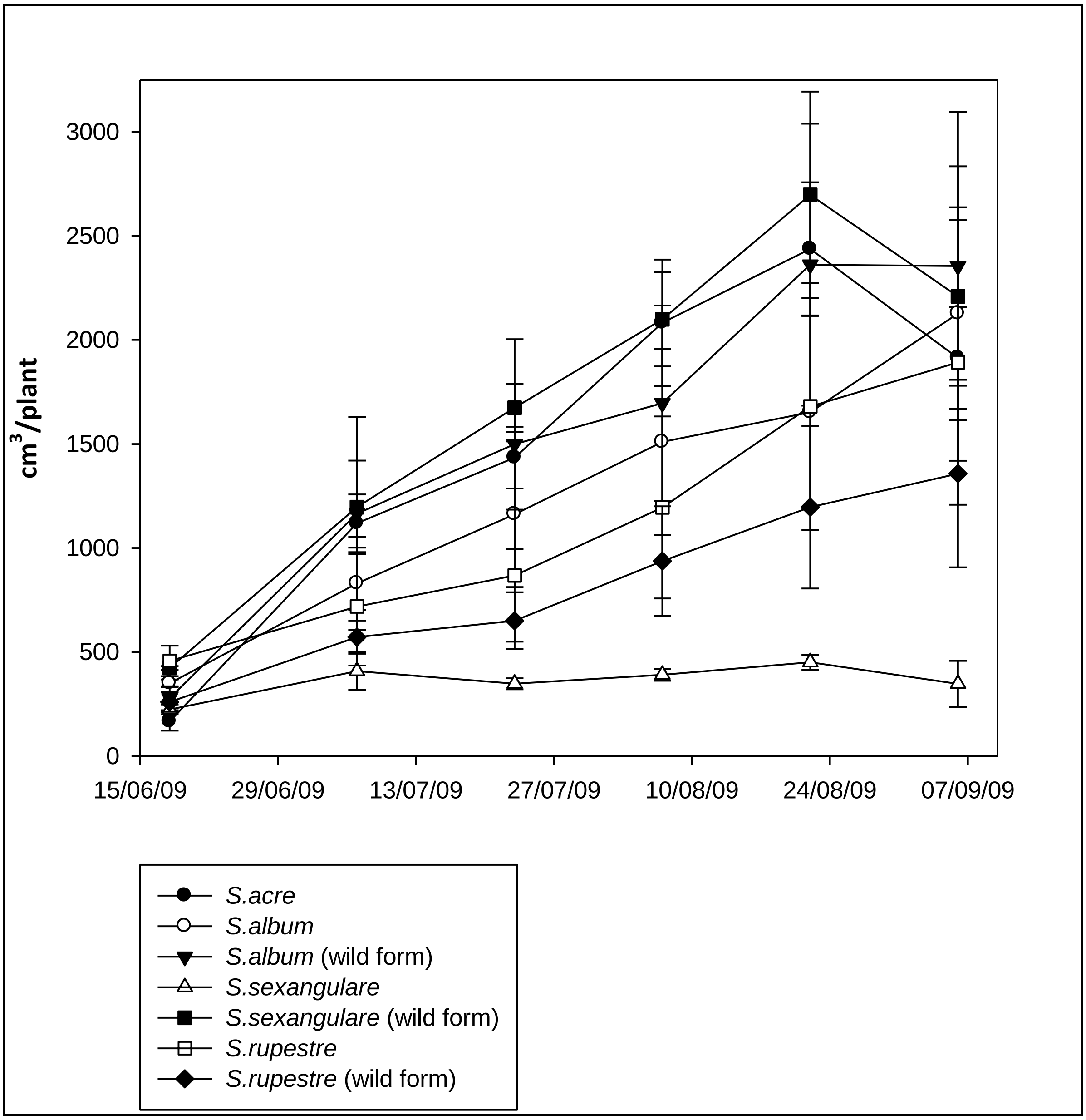
Fig.2.13 Growth Index pattern of perennials genotypes from transplanting (June) to September 2009.



Although during the experiment sedum growth did not show significant differences between genotypes, by the end of the growing season it was highly evident ($P<0.05$) that *S.sexangulare* (commercial forms) had a limited growth (Fig.2.14) compared to the other species. This taxon recorded the lower growth index value (408 cm^3/plant). On the other hand, *S.rupestre* (commercial) reached a growth index (3,402 cm^3/plant) higher than the others and than its local ecotype. Despite this, by early January both *S.rupestre* genotypes were comparable. This last data showed the marked potential of *S.rupestre* wild form, even if later in the season.

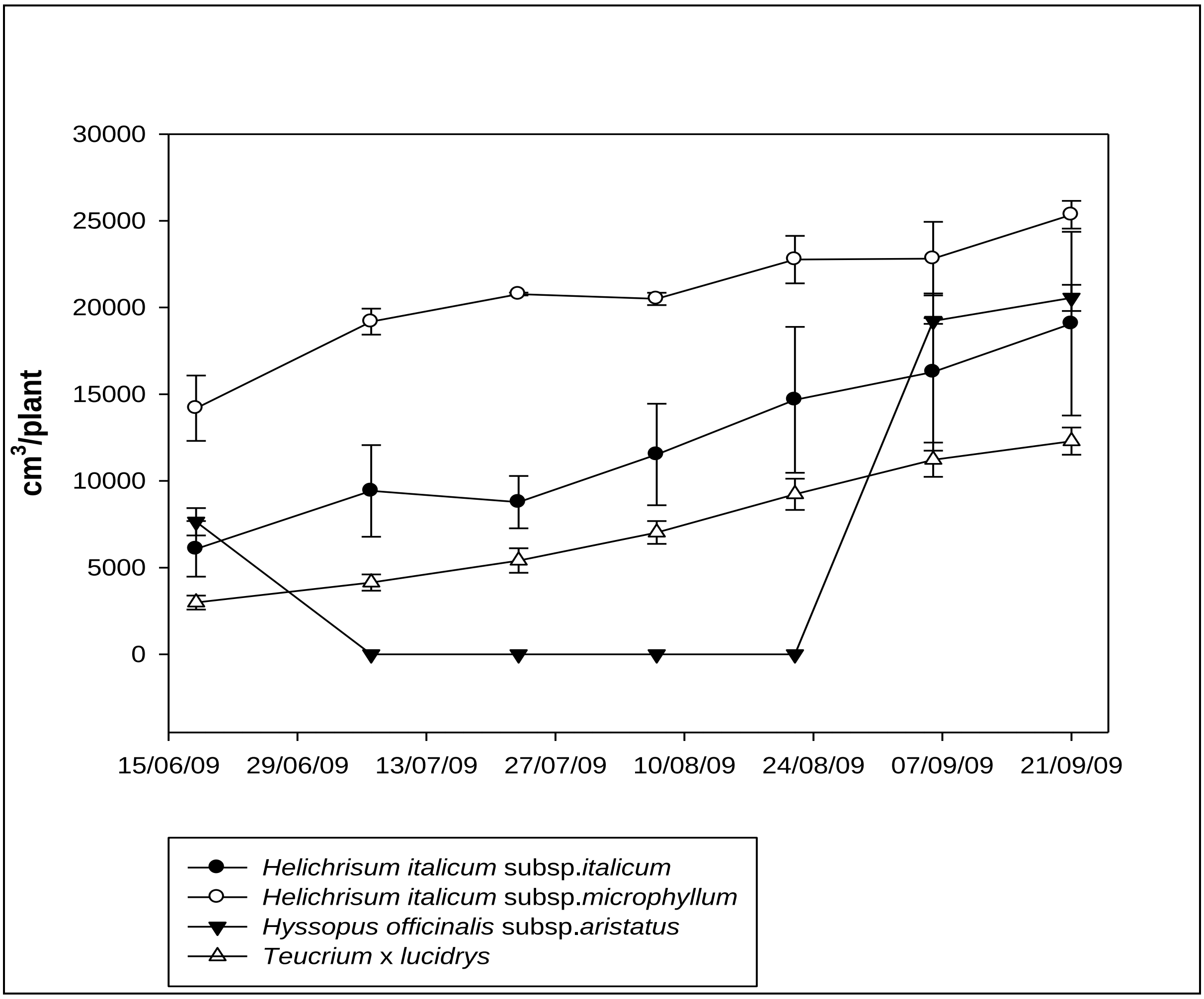
After September 2009, it was not possible to determine the growth index on the following taxa *S.acre* (wild form), *S.album* (wild form) and *S.sexangulare* (wild form), since it was no longer possible to distinguish individual plants from each other, thus comparisons within sedums were not longer possible.

Fig.2.14 Growth Index pattern of Sedums species from transplanting (June) to September 2009.



In subshrubs group, growth differed significantly between taxa (P>0.05) and mean estimated values of growth index ranged from 8,888 cm^3/plant recorded for *Teucrium* x *lucidrys* to 22,500 cm^3/plant reached by *Helichrysum italicum* subsp. *microphyllum*. The high difference observed at the beginning, particularly between the above-mentioned two taxa, was statistically maintained until the end of the growing season (autumn) (Fig.2.15). Moreover, although *Hyssopus officinalis* subsp. *aristatus* had initially presented symptoms of transplant shock, by the end of August all plants had recovered and, notably, the taxon rapidly reached by autumn a level of growth (20,56 cm^3/plant) comparable to *Helichrysum italicum* subsp. *italicum* (19,068 cm^3/plant) and to *Helichrysum italicum* subsp. *microphyllum* (25,384 cm^3/plant).

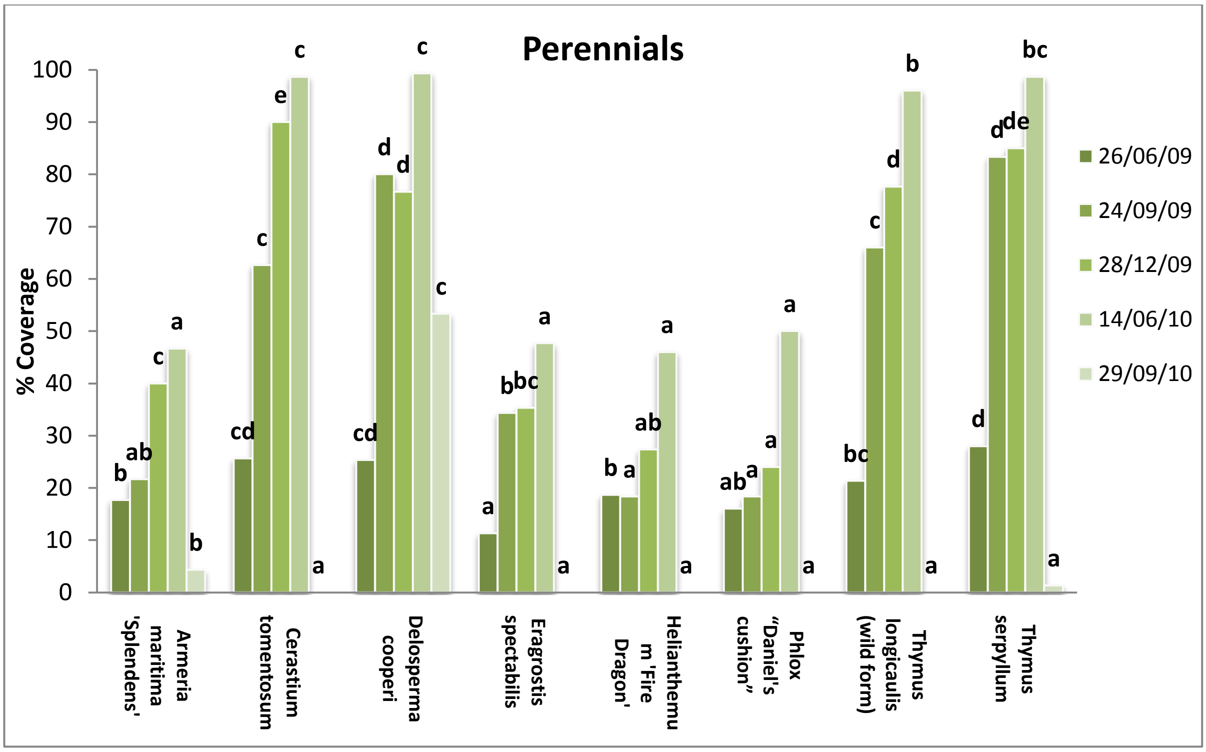
Fig.2.15: Growth index pattern of subshrubs from transplanting (June) to September 2009.



Concerning shrubs, no significant differences were found between the tested species. Nevertheless, plants showed a good growth during the trial and responded positively to the limiting depth, the limiting water supply and roof environmental conditions. However, during summer 2010, *Prunus mahleb* plants completely lost their leaves due to the drought stress occurred in July, while *Cotinus coggygria* continued to flourish. These species are known to be thermophilous shrubs typical of the maquis flora, which are also found in other areas in the country characterized by high soil drainage (Nardini *et al.*, 2003). These different responses given by the two species to the drought experienced in summer seems to reflect results found by Nardini *et al.* (2003). Indeed, *Cotinus coggygria* probably managed water stress more efficiently (through a low root's hydraulic resistance) and was consequently less vulnerable to drought than *Prunus mahleb*.

In the Perennials group, *Eragrostis spectabilis,* despite planted at a lower density compared to the others, thus initially recording a lower percentage of ground coverage (11,3%), had, by winter, recorded a higher coverage value than both *Phlox* 'Daniel's cushion' and *Helianthemum* 'Fire dragon'. In winter (28/12/09) these two species had the lowest levels of substrate cover, 24% and 27% respectively. *Cerastium tomentosum* and *Thymus serpyllum* showed a high capacity for growth, covering respectively 90% and 85% of the ground by winter. In addition*, Delosperma cooperi* and the native species *Thymus longicaulis* also showed themselves to be good groundcovers with a speedy growth rate in the short term (Fig.2.16). However, during the second summer season drought conditions on the roof were dramatic, and only *Delosperma cooperi*, *Armeria maritima* and *Thymus serpyllum* survived, maintaining respectively 53%, 5% and 1.3% of ground coverage.

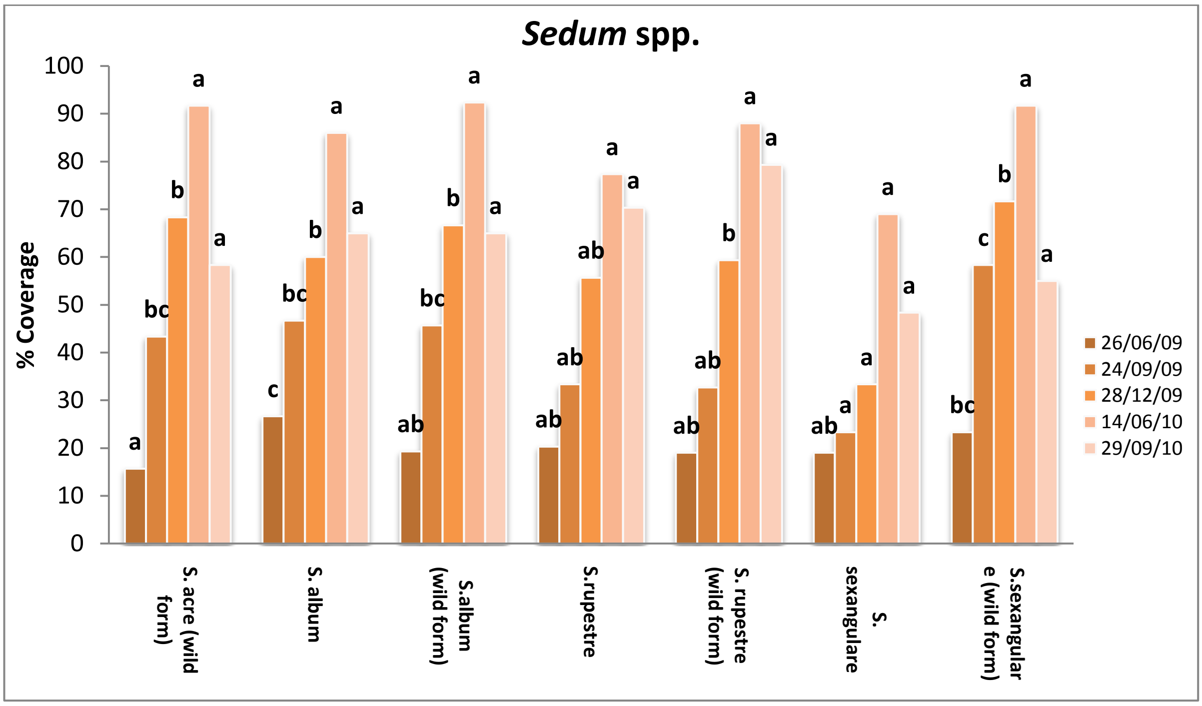
Fig.2.16 Percentage of substrate coverage within a 1 m^2 area of perennials from transplanting to September 2010. Means separation within each group of time series (days at which measures were recorded) by Duncan's multiple range test, P< 0.05.



Bars with the same superscript do not differ significantly from each other.

Sedum spp. exhibited a good growth and coverage rate reaching reasonable levels by autumn 2010. The development of sedums cover changed over the time, differences between species could be noted until up autumn 2010 (29th September) when reached similar percentages. By the end of the trial, these levels of coverage varied from 55% recorded for *S.sexangulare* (ecotype) to a maximum of 79% recorded for *S.rupestre* (ecotype), excepting *S.sexangulare* (commercial) with a lower value of 48% (Fig.2.17). All the native species performed as expected, satisfying the characteristic of good and fast groundcovers in the short-term. The less suitable species seems to be the *Sedum sexangulare* commercial which had a lower growth rate and smaller percentage of coverage compared to the other sedums. Nevertheless, all sedums survived to the summer drought conditions during the second year of study, while showed a reduction of cover as a result of the drought effect combined with the post-flowering shoots dying. In fact, as observed by other authors (Schuber and Kluge, 1981), some *Sedum* species tend to lost shoots vitality after blooming, while new stems grow from the root system. Finally, compared to the perennials, sedums covered relatively faster and more densely the substrate. This result, is not surprising at all, since in previous studies (Kirckher, 2004), performances of the same sedums taxa were similarly successful to the ones obtained in this study.

Fig.2.17 Percentage of substrate coverage within a 1 m^2 area of sedums from transplanting to September 2010. Means separation within each group of time series (days at which measures were recorded) by Duncan's multiple range test, $P < 0.05$.



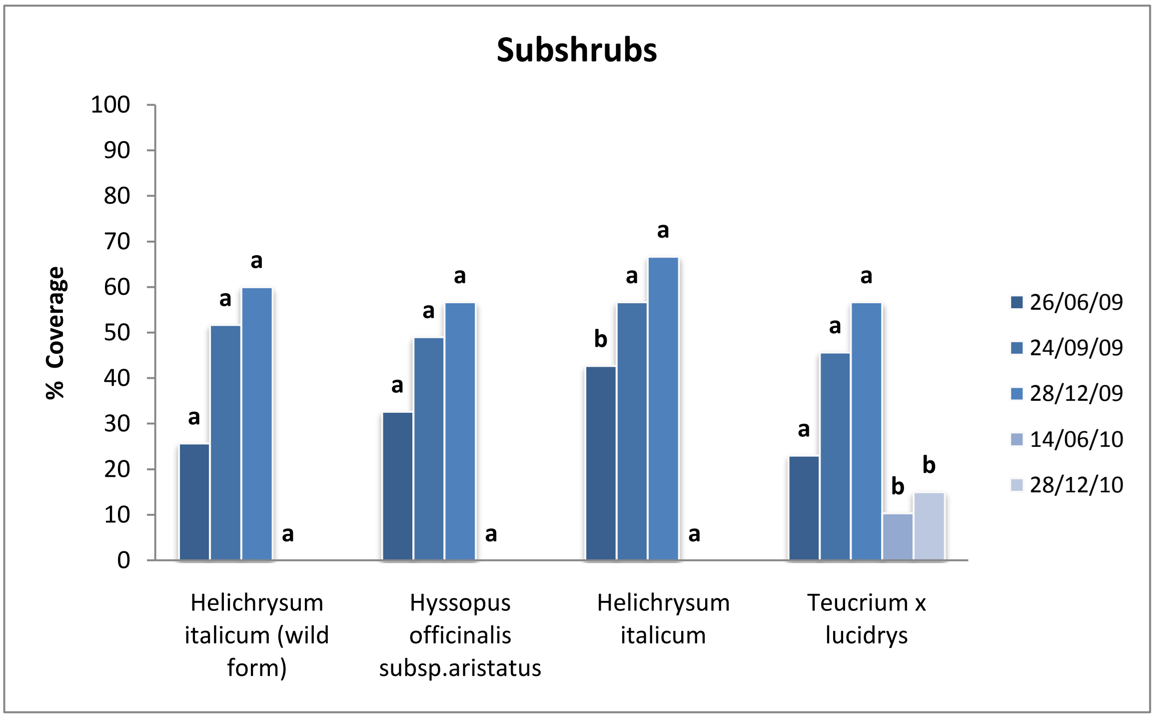
Bars with the same superscript do not differ significantly from each other.

Concerning subshrubs, even if the coverage values had initially (at the time of transplant) differed significantly within the species, by autumn all species were comparable to each other (Fig.2.18). *Teucrium* x *lucidrys* and *Helichrysum italicum* subsp. *italicum* (ecotype) demonstrated a potential to be valuable species for green roofs, showing in the short term a higher growth rate than the other subshrubs.

Although *Hyssopus officinalis* subsp. *aristatus* had initially presented symptoms of transplant shock, thus was at a disadvantage compared to the other plants, by the end of August all plants had recovered and by autumn the percentage of coverage was comparable to the others taxa.

During winter subshrubs plants stopped their growth because of the low temperatures, consequently values of cover were unchanged. From spring was no longer possible to record the percentage of ground coverage since plants died from rot root. This problem occurred for two reasons, firstly, the season was unusually heavy raining and secondly the plot had problem of water retention, which was not possible to detect visually. Nevertheless, few plants of *Teucrium* x *lucidrys* survived to both winter moisture excess and summer drought, showing a good resistance to adverse conditions.

Fig.2.18 Percentage of substrate coverage within a 1 m^2 area of subshrubs from transplanting to September 2010. Means separation within each group of time series (days at which measures were recorded) by Duncan's multiple range test, P< 0.05.



Bars with the same superscript do not differ significantly from each other. Flowering Dynamics

Plants used in this experiment were chosen also for their flowering characteristics in order to give a long interesting flowering season from spring through to autumn.

During the first year after transplant several taxa flowered. Because of plants were regularly watered flowering was longer and more abundant in the first year (Tab.2.15) than in the second year (Tab.2.16). Furthermore, subshrubs flowering is shown only for 2009 since by 2010 most of plants had died. Shrubs did not flower during the first or second year and flowering times of these species were hence not recorded.

Armeria maritima, as well as *Delosperma cooperi* and *Phlox* 'Daniel's cushion', were already in flower before transplanting. However, while *Phlox* flowering duration was relatively short, *Armeria* and *Delosperma* also flowered after transplanting. *Delosperma cooperi* had a great flowering performance, it flowered for a long season from June to November. Similarly, *Thymus serpyllum* was in flower for more than two months during summer (end of June to early September). *Armeria maritima* had between July and August a flowering declining probably due to after-transplanting acclimatizing phase.

Among subshrubs both *Helichrysum* genotypes flowered, and the commercial lasted flowers longer than the local ecotype. *Hyssopus officinalis* flowered later in the season (September to October), *Teucrium* x *lucidrys* did not flower.

During the second year, 2010, flowering dynamics were more interesting because plants had already established on the roof.

Among perennials *Eragrostis spectabilis* was the only taxon which did not reach the flowering phase because plants died before the flowering time. It is possible to see how some taxa like *Thymus* spp. had limited flowering times respect to those reported in the literature (Pignatti, 1982; Sell and Murrel, 2009). For *T.serpyllum* the strong drought between June and July caused the plants to withering July instead of August, as well as for *T.longicaulis* the roof conditions may had affected plant blooming duration, which was shorter than expected. *Delosperma cooperi* despite the severe conditions experienced, which reduced plants survival, had two flowerings. The first flowering was from May to July while plants started to re-flower in late September, when the environmental conditions were probably be less limiting than in the previous moths.

All Sedums regularly flowered in time (Pignatti, 1982). However, ecotypes exhibited more spectacular and abundant flowering than the commercials. Flowers were visually bigger and number of shoots higher. It was not possible to count the number of flowering shoots per plant because of their extreme fragility and, in some cases (*S.rupestre* wild form, *S.album* wild form and *S.sexangulare* wild form), their high density, but by measuring the number of flowers per flowering shoot, *S.rupestre,* commercial form recorded a significantly lower value than the wild form, while *S.album* and *sexangulare* genotypes were not different in respect to this variable.

Excepting *S.acre* which was not included in the analysis, since its shoots have normally one or at maximum two flowers, there was a significant difference in the number of flower per shoot between species ($P<0.001$). *Sedum sexangulare* genotypes and *S.rupestre* commercial form recorded the lowest values which ranged from 22 (*Sedum sexangulare* commercial) to 33.5 (*Sedum rupestre*) number of flower per shoot. Whereas *Sedum rupestre* wild form and *S.album* genotypes had more flowers per shoot for a maximum value of 68 flowers recorded in *Sedum album* wild form and a relatively lower recorded for *S.rupestre* wild form (60 flowers/shoot).

It was also observed that the height of shoots was positively related to the number of flowers ($R= 0.513$, $P<0.01$). The height of flowering shoots was highly different between species. *S.sexangulare* commercial recorded the lowest value (5.2 cm), followed by its ecotype (8.3 cm), by *S.album* (12.6 cm) and its ecotype (16 cm), while the highest values were recorded for *S.rupestre* genotypes (commercial 22.5 cm; ecotype 32.2 cm).

Table 2.15 Flowering times on from June (transplanting) to December 2009

Taxa	June	July	August	September	October	November	December
Delosperma cooperi	•• •••	••• ••• •••	••• ••• •••	••• ••• ••	•• • •	• • x	
Thymus longicaulis							
Thymus serpyllum	•	•• •• •••	••• •• •	• x			
Eragrostis spectabilis				• • •			
Phlox **'Daniel's Cushion'**							
Armeria maritima **'Splendens'**	•• •	•	•	• •• ••	• x		
Helianthemum **'Fire Dragon'**							
Cerastium tomentosum							
Sedum acre (wild form)							
Sedum album					• • ••	•• ••• ••	• x
Sedum album (wild form)					• •	•• •• •••	•• x
Sedum rupestre							
Sedum rupestre (wild form)							
Sedum sexangulare							
Sedum sexangulare (wild form)							
Helichrysum italicum subsp.microphyllum	•	•• ••• ••	• x				
Helichrysum italicum **subsp.*italicum*** ***(wild form)***	•	• •• ••	••• •• x				
Hyssopus officinalis **subsp.*aristatus***				• •• ••	•• • x		
Teucrium x lucydris		•	•				

Legend of symbols: •= >30% of plants with at least one flower in bloom; ••= >50% of plants with at least one flower in bloom; •••= >70% of plants with more than one flower in bloom; x= >70% of plants with no flower in bloom.

Table 2.16 Flowering times of groundcover perennials on 8 cm substrate depth during 2010 (3 values are given monthly from the measurements within 10 days of the months)

Taxa	**January**	**February**	**March**	**April**	**May**	**June**	**July**	**August**	**September**	**October**	**November**	**December**
D. cooperi					•• •••	••• ••• ••	• x		• •••	••• ••• ••	• x	
T. longicaulis				• •• •••	••• •• x							
T. serpyllum				•	••• ••• •••	••• •• ••	x					
E. spectabilis												
***P.* 'Daniel's Cushion'**			• ••	••• ••• •••	••• •• x							
***A. maritima* 'Splendens'**				• •• •••	••• ••• •••	•• •• •	x					
***H.* 'Fire Dragon'**				•	••• ••• ••	x						
C. tomentosum				•	••• ••• ••	x						
S. acre (wild form)						•	••• • x					
S. album					• •	••• ••• •••	••• • x					
S. album (wild form)					• ••	••• ••• •••	••• •• •	x				
S. rupestre				•	•• ••• •••	••• ••• ••	•• • •	x				
S. rupestre (wild form)					• •• •••	••• ••• •••	• x					
S. sexangulare					•	•• ••• •••	••• • x					
S. sexangulare (wild form)					••	••• ••• •••	••• • x					

Legend of symbols: •= >30% of plants with at least one flower in bloom; ••= >50% of plants with at least one flower in bloom; •••= >70% of plants with more than one flower in bloom; x= >70% of plants with no flower in bloom.

Colonizing species

The most represented colonizing species found on the roof belonged to the Asteraceae and Poaceae families (Figure 2.19). Asteraceae family followed by the Poaceae are generally the most represented in term of number of species within the Italian flora (Pignatti, 1982). The species found on the roof were mostly annual herbaceous with seeds dispersal by wind (Tab.2.17).

Table 2.17 Characteristics and names of the most represented colonizing species found on the experimental plots during the first year of study.

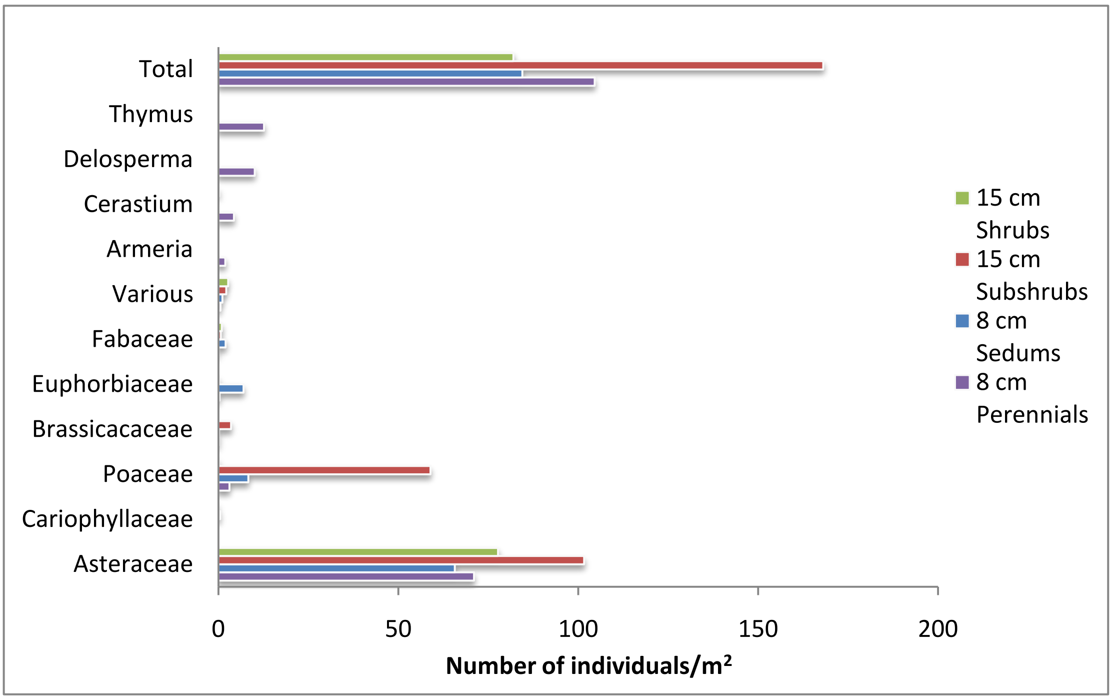
Family and specie	Type	Habitat
Amaranthaceae		
Amaranthus retroflexus L.	Annual herb	Naturalized, disturbed areas
Asteraceae		
Conyza canadensis (L.) Cronq.	Annual herb	Naturalized, meadows, uncultivated fields
Sonchus asper (L.) Hill.	Annual herb	Native, uncultivated fields
Sonchus oleraceus	Annual herb	Native, uncultivated fields
Lactuca serriola L.	Annual/biennal herb	Native, uncultivated fields
Senecio vulgaris L.	Annual/biennal herb	Native, uncultivated fields, meadows,
Portulacaceae		
Portulaca oleracea L.	Annual herb	Cosmopolitan, cultivated fields, meadows
Poaceae		
Poa annua L.	Annual herb	Cosmopolitan, cultivated fields, pasture
Setaria viridis (L.) Beauv.	Annual herb	Subcosmopolitan, abandoned fields
Digitaria sanguinalis (L.) Scop.	Annual herb	Cosmopolitan, arable weeds, uncultivated
Brassicacaceae		
Cardamine hirsuta L.	Annual herb	Native, disturbed sites, cultivated fields
Myrtaceae		
Eucalyptus globulus Labill.	Tree	Exotic, cultivated
Euphorbiaceae		
Euphorbia prostrata Aiton	Annual herb	Naturalized, disturbed sites
Chenopodiaceae		
Chenopodium opulifolium	Annual herb	Native, cultivated fields
Leguminosae		
Trifolium repens L.	Perennial herb	Native, meadows, disturbed sites

Seedlings of the self-sowing taxa *Armeria maritima*, *Cerastium tomentosum*, *Delosperma cooperi* and *Thymus* spp. were found and recorded. While *Sedums* spp. were not included since was often not certain the origin of the new plants found, as *Sedum* shoots detach and re-root easily and can be thus confused with seedlings.

Comparing the total number of adventitious plants found on the tested plots, the 15 cm planted with subshrubs seemed the more subject to colonization by volunteers. This was probably due to the higher level of moisture retention occurred in this plot besides the partial ground coverage of plants. Subshrubs reached by winter 2009 a maximum percentage of ground cover of about 60%, which was subsequently reduced due to plants death from rot root. On the other hand plant coverage in the other plot was comparable or even higher than that of the subshrubs, which with the new growing season increased at the expense of volunteers' development and growth. Moreover, the higher level of moisture retention occurred in subshrubs plot had further encouraged volunteers expansion and colonization.

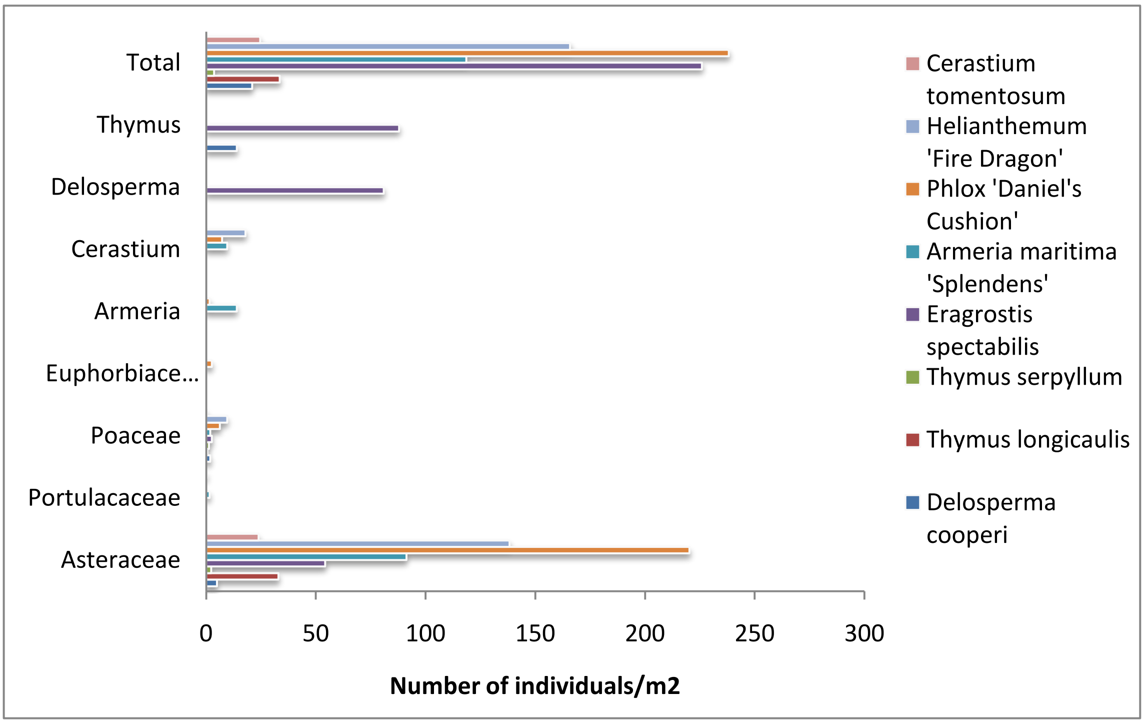
Concerning the 8 cm plots, without considering self sown seedlings, there is not substantial difference between the two planting types (Perennials vs. Sedums). However, it has to be considered that in general all Sedums required less maintenance during 2010, when ground coverage was almost completed while during the establishment cover was not high. On the other hand, within the perennials group, several taxa rapidly reached high percentage of ground cover before autumn 2009, while others remained at low level of ground coverage, giving space to volunteers to grow.

Figure 2.19: Total abundance of colonizing species found on the vegetated experimental plots.



In fact, as it is shown in the following chart (Fig.2.20) it is evident how among perennials the low growing taxa, *Phlox* 'Daniel's cushion', *Helianthemum* 'Fire Dragon' and *Armeria maritima*, showed higher susceptibility to be colonized by volunteer plants.

Figure 2.20: Total abundance of colonizing species found on perennials plots



Among planted taxa *Armeria maritima*, *Cerastium tomentosum, Delosperma cooperi* and *Thymus* spp. showed to be good self-seeders and generated new plants. *Thymus* spp. and *Delosperma* seedlings were the most abundant, even if *Thymus* spp. comprises two taxa the number of structural plants was twice that of other taxa.

Interestingly, the seedlings of *Delosperma cooperi* and *Thymus* spp. were found to grow mostly on the neighbourhood subplots of *Eragrostis spectabilis*, suggesting that dead plants of this taxon provided a favourable environment for new seedlings growth and development. In fact, seedling of these taxa were found at the base of *Eragrostis* plants, were moisture was probably kept longer by the shading effect of the dead canopy and probably also by the fibrous root system of the grass plants. Concerning the other two self-sowing taxa *Cerastium tomentosum* seemed to have higher colonising potential than *Armeria maritima,* which recorded a lower number of seedlings and also it showed to grow only nearby mother plants since seeds weight and size is higher than *Cerastium* (http://b-and-t-world-seeds.com). Nonetheless, *Armeria* in a 5 years long term study carried out in UK shown to increase its abundance in shallower substrate depth (10 cm) more than in deeper (20 cm) (Dunnett *et al.*, 2008a)

2.3.4 CONCLUSIONS

In terms of plant growth, establishment, regeneration and flowering performance results from this study confirm that the investigation of native plants as green roof resources should be pursued and promoted both from ecological and commercial standpoints. Even if is not necessarily true for some native species to be more successful than non-native species on green roofs, these findings demonstrated that for some Mediterranean native species it is true. Moreover it can be said that overall wild-collected material gave good results, performing equally or even better than commercial material. However, findings also suggest to further investigate drought tolerant non-native species with potential for green roof planting.

In a Mediterranean environment succulent species seems to be the best planting option which can guarantee more than others perennials a relatively fast and constant green cover on low-input lightweight green roofs. Among sedums local genotypes responded positively to the harsh roof conditions and seemed to perform overall better than commercial genotypes. While within commercial genotypes *S.sexangulare* had undeniably the lowest growth and coverage capacity.

The selection and the planting design for perennials can be seen from a different perspective than usual. Perennials can be used in these harsh circumstances as annuals, thus drought-tolerant and good self-sowing species like *Armeria maritima* 'Splendens'*, Cerastium tomentosum, Delosperma cooperi* and *Thymus spp.* could be recommended when depth and level of maintenance are limited. Particularly, it can be noticed how native species such as *Cerastium tomentosum* and *Thymus longicaulis* confirmed their predicted suitability for green roofs, while some non-native taxa such as Phlox 'Daniels' Cushion' and Helianthemum 'Fire dragon' can be discouraged for a green roof purpose in a Mediterranean environment. As concluded by Koehler (2003) from 20 years of experience and observation of green roofs' vegetation survival and dynamics, plant species for extensive systems should have the characteristics of dry adaptation/tolerance, high seed production and finally a short life cycle or a life cycle with plant stand-by when environmental conditions are not ideal for growth (i.e. hemicryptophyte growth form like *Armeria*). Therefore in order to ensure plant diversity and ground coverage through the time, for plant establishing it has to be considered the low growing habit of species like *Armeria maritima*, and the plant density or seeds percentage in these cases should be probably higher to ensure better results. Also, the use of grass species should be further investigated and probably promoted considering the positive effect had by *Eragrostis spectabilis* on *Thymus* and *Delosperma* re-generation. Additionally, grasses having ascendant growing form more than mat-forming, may give to the extensive vegetation element a more interesting structure.

Moreover, *Delosperma cooperi* exhibited higher potential compared to the other perennials taxa, but it is also true that this species has the advantage to be succulent and CAM as sedums do. Nevertheless, respect to all taxa (succulents and non-succulents) its long flowering times can also visually ensure to the roof more interesting aesthetical dynamics.

It can be thus concluded that in these latitudes for sustainable extensive green roofs, which mainly rely on precipitation events, succulents species fulfil the requirement to tolerate harsh conditions. However, substrates deeper than 8 cm could provide greater water holding capacity as well as faster plant establishment and substrate coverage of a wider planting selection (Monterusso *et al.*, 2005; Durhman *et al.*, 2007).

Concerning subshrubs, as far as the first 9 months of study, all taxa gave good results in term of growth and coverage attitude. Particularly, *Hyssopus officinalis* subsp. *aristatus* (native)*,* which showed a notably restoration after transplant, and *Teucrium* x *lucidrys,* which survived to all severe stress conditions, may be recommended for semi-extensive realization in a Mediterranean climate. However, a longer term studies needs to be performed to confirm these preliminary results.

Finally, shrubs on 15 cm substrate depth surprisingly survived. Nevertheless, between the two native species selected *Cotinus coggygria* seemed to tolerate more the stress experienced on the rooftop, giving better results than *Prunus mahleb*.

3 EXTENSIVE AND SIMPLE-INTENSIVE GREEN ROOFS' THERMAL EFFECT IN A MEDITERRANEAN CLIMATE

3.1.1 INTRODUCTION

The city surfaces have different albedo compared to green areas. For a city as a whole, the albedo can be as low as 10-15% (the albedo for fresh snow is greater than 80%) which means that a lot of the incoming solar energy is absorbed by the city. Additionally, most of the building materials used in the construction of cities are characterised by a high heat capacity and high heat conductivity. These factors contribute to the so-called Heat Urban Island Effect, by which temperature in cities results higher than in the greener surrounded areas (http://www.epa.gov/heatislands/about/index.html).

The original, natural land cover may be preserved in lawns or parks or even green roofs, by which local government can mitigate the negative environmental effects caused by impermeable-hard surfaces, which instead dominate in cities.

The greatest benefit which green roofs can immediately provide in a Mediterranean climate is the summer cooling effect and the relative energy saving (Nichau *et al.*, 2001; Santamouris *et al.*, 2007). Nonetheless, it may also reduce the buildings heating load compared to conventional roofs (Bass *et al.*, 2006).

The cooling effect of green roofs is not only due to the shading provided by greenery. Some author stress more the effect of the water evaporation from soil and the plant transpiration than the shading effect (Ferrante and Mihalakakou, 2001; Lazzarini *et al.*, 2005).

Even though many studies have assessed the thermal benefits of green roofs, the climatic conditions were often different to those of the Mediterranean and data specifically referred to the Italian climate are scarce (Liu and Baskaran, 2005; Fioretti *et al.*, 2010; Teemusk and Mander, 2010). Despite the wide literature available on green roofs thermal properties and benefits, many research studies carried out seems to simplify the role of the vegetation, and the effect of its morpho-physiological characteristics has been probably underestimated (Castleton *et al.*, 2010; Fioretti *et al.*, 2010; Ludholm *et al.*, 2010).

Vegetation composition and structure is important in determining the roof performance, including thermal performance. Several studies have paid attention to the vegetation composition and how the plant choice can affect temperature recorded on the surface as well as under the studied green roof systems (Sidwell *et al.*, 2008; Lundholm *et al.*, 2010). In addition another factor which has been investigated in thermal green roof properties is the substrate depth (Sidwell *et al.*, 2008)

The study previously-described led to consider the monitoring of the different green roof system characterized by different vegetation types and collect data to address future thermal green roofs experiment in Mediterranean climate condition.

Starting from the above considerations, the aim of the research activity was to monitor the temperatures of two different kinds of green roof systems - extensive and semi-extensive, both with and without vegetation. As this experiment was partially funded by Harpo-Seic green roofs division, plots layout and dimensions, substrate depth and some of the species used (Shrubs on 15 cm substrate depth) were agreed according to the company requirements and to the roof space available.

The main objective was to compare the temperatures under the root membrane and on the soil surface of two green roof systems (8cm vs. 15cm) and in addition to compare these against the temperature found on a conventional roof. Another objective was to analyse how temperature fluctuations can be influenced by the presence or absence of vegetation.

3.1.2 MATERIALS AND METHODS

Study site

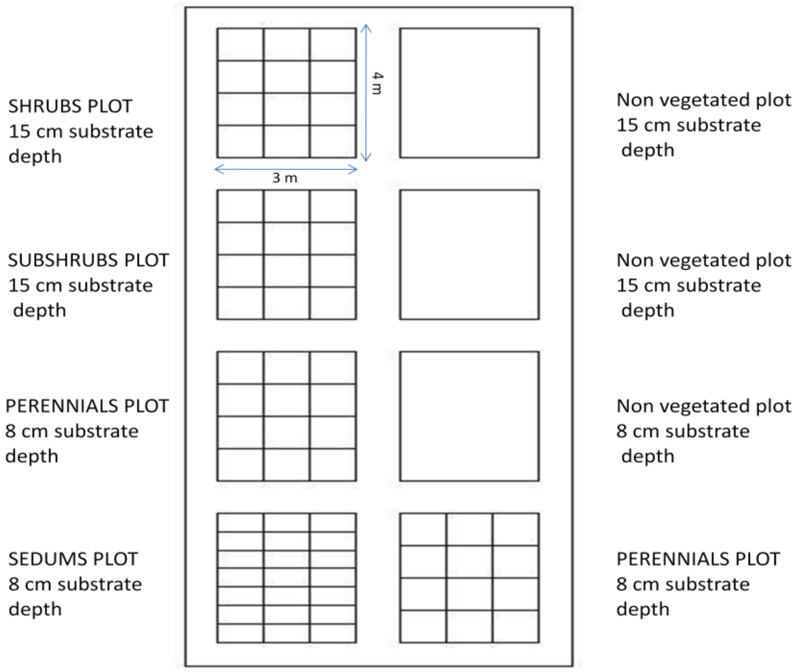
The experiment was set up in June 2009 on a flat roof of a two-story Tuscia University building (at latitude of N42˚ 26’, at longitude E12˚ 40’, and altitude of 310 m), Viterbo, Italy. The roof was about 438 m^2. Plots were placed in two lines along the longer side of the building. Seven experimental plots with two different substrate depths (8 cm and 15 cm) both in the presence or absence of a vegetation layer were thus set up.

Plots characteristics and layout

The plots and substrate characteristics have been previously described in chapter 2.2.

The substrate thickness varied between plots. In total there were 7 plots (Fig.3.1): four semi-extensive (15 cm substrate depth), of which one was planted with subshrubs and one with phanerophyte species and two were left without vegetation; three extensive (8 cm substrate depth) of which two contained groundcover perennials, while one was left bare.

Figure 3.1: Experimental roof and plot layout



In total 14 plant taxa were planted, 4 subshrubs [*Helichrysum italicum* subsp. *italicum*, *Helichrysum italicum* subsp. *microphyllum* (commercial), *Hyssopus officinalis* subsp. *aristatus*, *Teucrium* x *lucidrys*]; 8 perennials [*Armeria maritima '*Splendens', *Cerastium tomentosum*, *Delosperma cooperi*, *Eragrostis spectabilis*, *Helianthemum* 'Fire dragon', *Phlox* 'Daniel's Cushion', *Thymus serpyllum*, *Thymus longicaulis*]; 2 shrubs [*Cotinus coggygria* and *Prunus mahleb*]

Plots were placed in pairs along two lines and three rows, treatments with vegetation were on the same line, while type of vegetation and depth on rows. The second line was constituted by non-planted plots excepting for one of the perennials plot which was placed on the same line and creating a fourth row, for space availability reasons.

Temperature, Moisture and Meteorological Measurement

The temperature was measured every 15 minutes using FT-102T, a temperature data logger equipped with two thermistors produced by Econorma (Italy). Measurements formally started on

9th August 2009. Temperatures were recorded for every plot at the following locations: 1) on the paved surface 2) on the bare soil surface ; 3) on the soil surface under the vegetation; 4) under the protection membrane; 5) in the air 80 cm above the roof.

Building internal temperature was not recorded since could be influenced by occupancy and by the heating/cooling devices used.

During the establishment period the plots were watered when the substrate moisture content decreased to approximately 20%. The substrate moisture content was regularly checked with 2 sensors (Irrometer Company, Inc., California) for each plot. Sensors were calibrated through pot gravimetric method, thus each amount of substrate moisture loss was related to the level of water soil conductivity measured with probes. The level of 20% moisture was decided after conducted a laboratory test which estimated the substrate field capacity and the wilting point.

During the second year of study regular irrigation was interrupted and substituted by only emergency water supply. During summer plants required watering due to very dry conditions, all plots were watered three times at mean intervals of days.

Daily weather observations were made at Tuscia University's Experimental Farm "Nello Lupori" by using a scientific automatic weather station with measurements logged to a CRX-10 data logger (Campbell Scientific).

Concerning the local climatic conditions during summer 2009 from July to August the average of mean daily maximum temperature was 33.8 °C, total rainfall was 25 mm distributed during the period and never reaching values higher than 11 mm per decade, and the crop reference total evapo-transpiration during the same period was 302.9 mm. In winter 2009-2010 the total amount of rainfall recorded was 493.6 mm, while evapo-transpiration was 129.7 mm and minimum temperature average was -1.75 °C. In 2010 from July to August the average of maximum temperature was 33.5 °C, the total rainfall was 90 mm, of which 80 mm were recorded only during the last decade of July, while the total evapo-transpiration over the two months was totally 442.5 mm (Fig.2.11). Data are shown in the Chapter 2, paragraph 2.1.6.

Statistical analysis

Surface temperature were not included in the analysis since were subjected to the effects of many variables and thus they can be misleading.

T-test independent sample was performed with SPSS 16.1.

3.1.3 RESULTS AND DISCUSSION

The main significant result obtained from summer temperature data is that green roofs (extensive and simple intensive, 8 cm and 15 cm substrate depth) highly reduced the maximum temperature by up 30 °C when compared to the conventional paved roof. While the roof reached at midday values over 60°C, underneath the green roof plots temperature was always lower than 35°C (Fig 3.1). This result seems to confirm data found in central Italy by Fioretti *et al.* (2010).

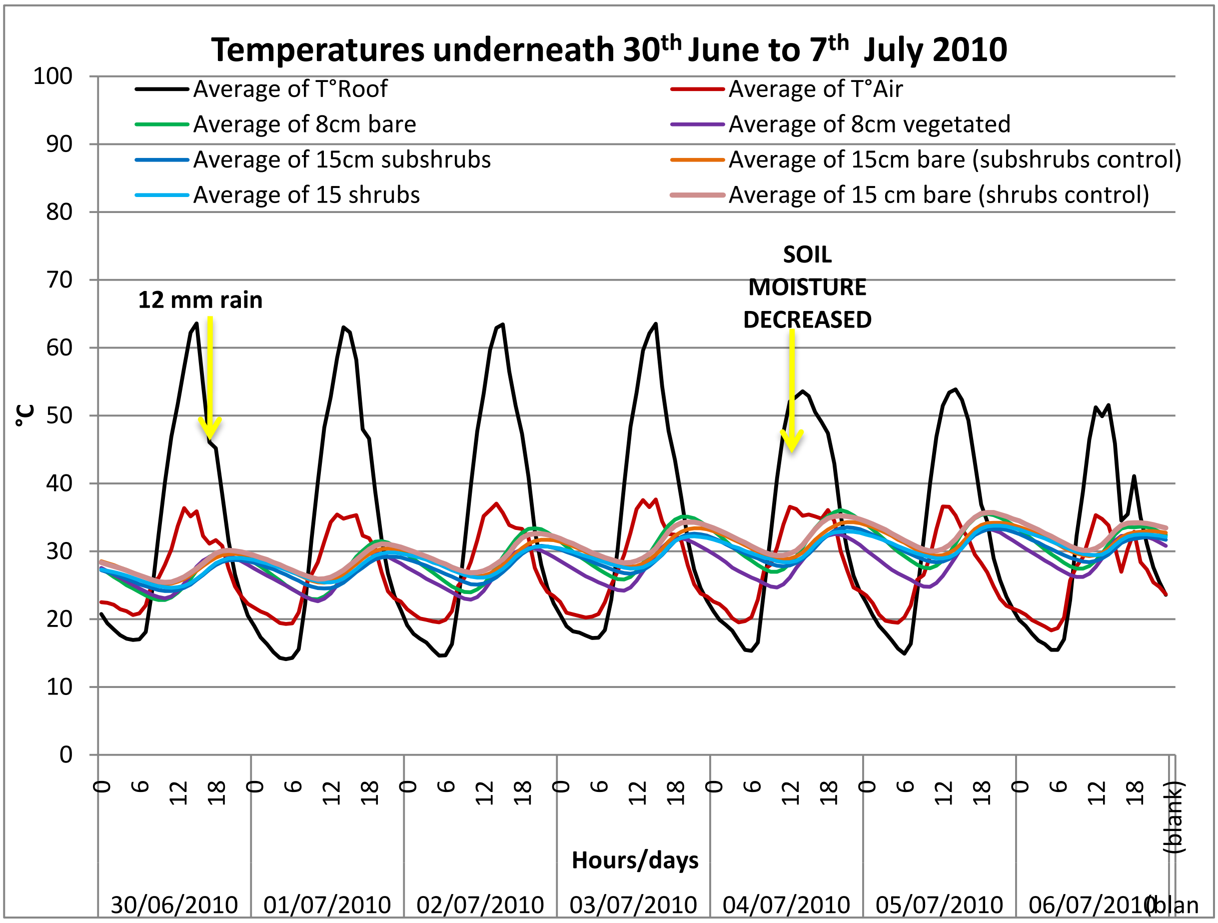
No significant differences were found between all systems tested, meaning that their mitigation effect was similar in summer times, when minimum and also when mean and maximum daily temperatures were concerned. Nonetheless, maximum temperatures recorded underneath vegetated plots during summer were significantly lower by 1.3°C than bare soil plots. Also, the contrast between vegetated and non-vegetated plots' underneath temperature was significantly different when only shallow substrate depth was considered (8cm).

Figure 3.1 illustrates the temperature patterns of different roofing systems over a typical summer week. Observing the temperature patterns on these days it is evident that the presence of a green roof system significantly ($P<0.001$) reduced the extreme temperature fluctuations normally registered on the conventional roof surface, as has been observed also by other authors (Liu and Baskaran, 2003; Teemusk and Mander, 2010). In addition, the deeper green roof system reduces the range of temperature fluctuation even further than the shallower one.

Comparing the peak temperature recorded on the conventional roof surface to the temperature found under the green roof systems at the same time of the day, the difference may reach values by up to 30°C. Moreover, the hour at which peak temperature is reached is later with the green roof system than on the conventional roof. During summer (June to August) this peak is delayed by 5 hours, while peak of the paved roof surface was medially recorded around 2 pm underneath the systems the daily peak temperature was reached at 7pm. This result seems to be in agreement with those found by Liu and Minor (2005).

Moreover, temperatures underneath the plots were almost equal, especially when substrate was wet, whilst as the substrate moisture decreased the temperature underneath the green roof systems increased. Lazzarini *et al.*, (2005), noted that when substrate is wet the transferred heat flux underneath the system result null and a slight outgoing flux seems to be produced. As the substrate lose water the transferred heat underneath the system increases, as seems to be confirmed by results. With regard to this fact Nyuk Yen (2007) in a greenroof experiment under tropical climate observed that the substrate temperature can exceed the roof temperature when the substrate is very dry. Thermal properties of the soils, it is known, are affected by several factors such as soil texture, composition, density and water content, and the temperature and heat movement in soils is also function of time and depth (Abu-Hamdeh, 2003).

Figure 3.1 Temperature pattern underneath the green roof systems, of the air and of the paved roof surface during a typical summer week (30/06/2010 to 7/07/2010)



Surprisingly, differences between plots were not statistically significant and also the 8 cm deep perennials-planted plot seemed to cool more than expected. This fact can be perhaps explained by the following considerations. Firstly, the subshrubs-planted plot had only the combined effect of evaporation and partial plant canopy shade, since almost all plants were not vital. Additionally, shrubs/trees can control water loss, in other words transpiration, more than herbaceous or subshrubs plants. It has been observed that trees can close their stomata during all day when soil moisture conditions are limiting (Yocum, 1953). Furthermore, phanerophyte have a lower number of stomata on their leaves because stomata presence is limited to the leaf lower page (Yocum, 1953). Secondly, while perennials almost covered the plot (particularly the area beneath which was placed the probe) the canopy high and density of the phanerophytes probably allowed more solar radiation to reach the soil surface. All this considerations may lead to think that while in the 8 cm substrate there was a stronger effect of transpiration and shade from plants, on the shrubs planted plot this effect was much lower.

Regardless of several studies have emphasized evapo-transpiration as the main function of green roofs in reducing heat flux through the roof (Ferrante and Mihalakakou, 2001; Lazzarini *et al.*, 2005), recent studies also demonstrated that plant shade (Fioretti *et al.*, 2010) and albedo (reflectivity) can significantly help in reducing the incident solar irradiance, thus the temperature and heat gain (Lundholm *et al.*, 2010).

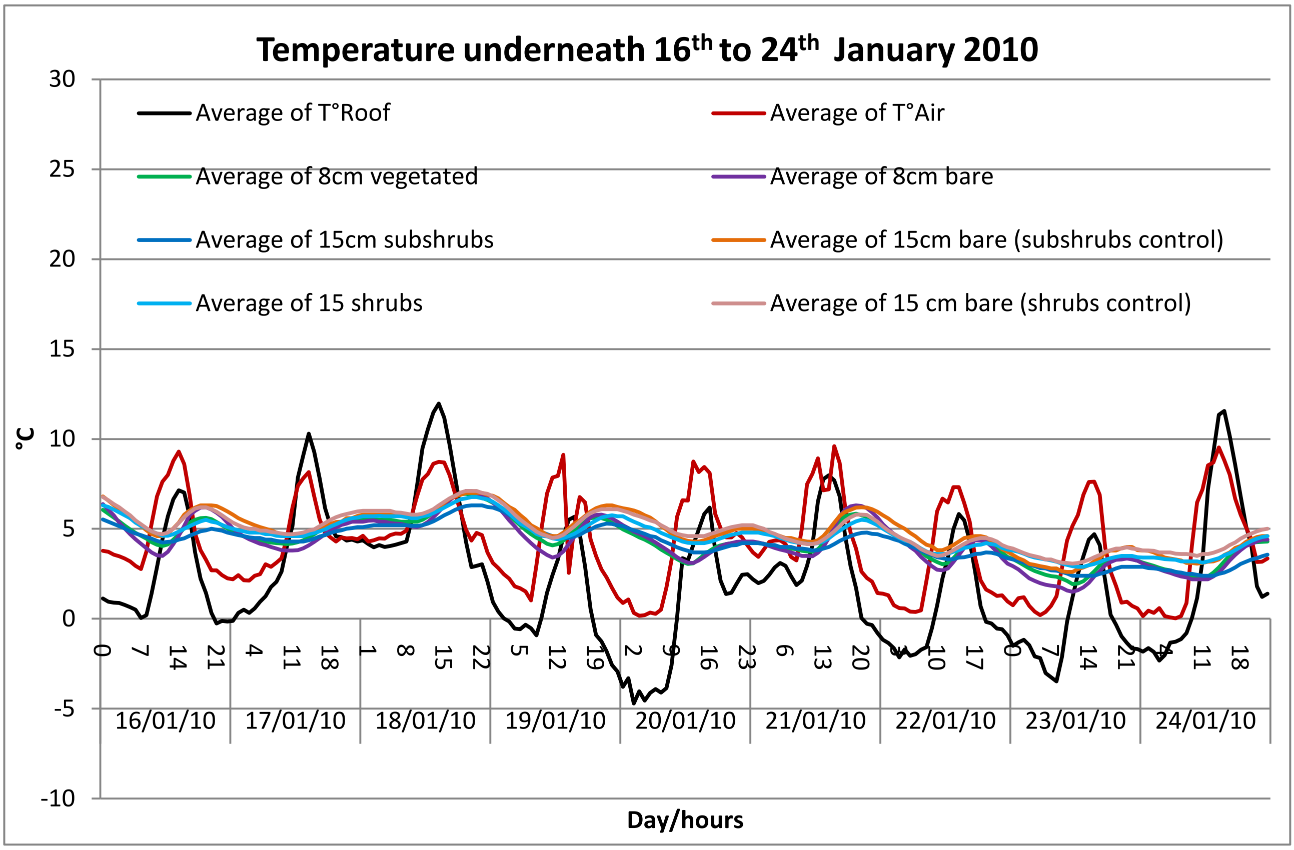
Perennials such as *Cerastium tomentosum, Helianthemum* 'Fire Dragon' and *Eragrostis spectabilis* tested in this study, may have high albedo due to the silver-grey and pale green colour of their leaves (Turner and Kramer, 1980). Therefore also this factor might had played a role in the temperature declining observed in the 8 cm plot compared to the 15 cm one.

Moreover, Sidwell *et al.* (2008) in their work arrived to the conclusion that by increasing of the system depth do not necessarily increase the thermal benefit, while changing plant species (even if within a same genus) can make more difference in the temperature change.

Some authors consider green roof's efficiency during the winter season as insignificant in warm climates (Santamouris *et al.*, 2007) or even less efficient in reducing the outgoing heat flux from the building respect to a conventional roof (Lazzarini *et al.* 2005). However for warm climates, such as the Mediterranean, the mitigation effect of green roofs on the roof surface and consequently on the bituminous membrane should not be discounted (Liu and Baskaran, 2005; Teemusk and Mander, 2010).

In fact, the thermal data recorded clearly evidences that the presence of a green roof system can significantly mitigate the extreme temperature fluctuations during winter as well ($P<0.001$). When the temperature on the green roof system never decreased to 0°C the conventional roof reached a temperature below this (Fig.3.2). During winter the differences between the minimum daily temperature recorded underneath the green roofs systems and on the paved roof surface can reach 7°C.

Figure 3.2 Pattern of temperatures underneath the green roof systems, of the air and of the paved roof surface during a typical winter week (16/01/2010 to 24/01/2010)



During winter the 15 cm and 8 cm deep systems did not differ significantly. Instead, it is noticeably that the absence of vegetation seems to make a significant difference in temperature fluctuations, minimum temperatures recorded underneath the bare systems were higher ($P<0.05$) than the ones recorded underneath the vegetated plots (4.2°C vs. 3.9°C). However it has to be distinguished that in shallower systems the vegetation seemed to probably reduce the outgoing heat transfer ($P<0.001$; 3.9 vs. 3.85), while in 15 cm substrate depth the vegetation presence seemed to do not have the same effect ($P<0.01$; 4 vs. 4.4°C). This reverse attitude may be related to the fact that while in 8 cm plot the perennials canopy was closer to the ground and more dense than in 15 cm plots (subshrubs and shrubs), a lower wind movement could have been recorded. In subshrubs plant canopy tend to cover the ground but stems are not so close to it, while the shade effect of canopy along with the wind action can help to reduce the heat gain and to facilitate more the heat loss compared to the respective bare plot. Also, the shrub planted plot seemed to follow the same pattern of bare ones, since phanerophyte deciduous plant lost their canopy in this season and substrate was almost uncovered. Thus in 15 cm vegetated plots, shading combined with the air movement on the roof surface could had played a role in the heat gain/release through the system.

After these considerations, *ad hoc* investigations are required in order to confirm the reported results.

3.1.4 CONCLUSIONS

It may be concluded that the thermal data has confirmed that green roof is an effective tool to reduce extreme temperature fluctuation on the roof surface during the whole year. Particularly in the most critical period, such as summer in the Mediterranean environmental conditions, the temperature underneath the green roof can differ from the one of the roof surface as much as 30°C.

Also, the increase of depth do not seems to affect as expected temperature underneath the systems. Nevertheless, results obtained suggest that the vegetative layer can make a significant difference in the extreme temperature fluctuations mitigation compared to the non vegetated systems in winter as in summer. Moreover, plant choice and selection looking at specific morpho-physiological features, which are often also related to drought-tolerance characteristics (i.e. green-silver foliage, leaves pubescence), result to be an important aspect to be considered also in the perspective of improving green roofs performance, since plant has been proven to give an important contribution by providing additional thermal benefits.

Finally, both extensive and semi-extensive green roofs systems reduce significantly extreme temperature fluctuation on the roof surface in Mediterranean climate conditions; however the investigation needs a longer time period and more accurate details to better quantify the differences between and within green roof systems and vegetation types.

4 STUDIES ON PLANT SELECTION, PERFORMANCE AND DYNAMICS ON GREEN ROOFS IN A MARITIME CLIMATE

This part of the thesis describes a series of experiments which took place at Sheffield University, Landscape Department (Sheffield, United Kingdom), during the year 2010 and under the supervision of Dr.Nigel Dunnett and Dr.Zoe Dunsiger.

To follow, the two plant screenings presented are part of ongoing projects in which I was allowed to take part and a study concerning a vegetation survey conducted on a case-study roof also located in Sheffield is also presented.

Although maritime climate is not properly comparable with the Mediterranean, the systematic approach which characterizes the research methodology of Dr.Dunnett and his assistants, along with their long experience on roof greening during the last decade, especially on ecological and planting aspects, were the key elements which had been considered to select the exchange research site for this PhD.

As a temperate climate the maritime climate is characterized by the absence or a very slight difference in extreme temperature and precipitations. Furthermore, passages between the two main season winter and summer are usually not dramatic, where winter is typically mild and summer cool. In fact, the maritime climate is strongly influenced by the ocean's airflows which help to mitigate climate through seawater heating capacity which takes longer than soil to warm up in summer and release slower the heat gained arising winter air temperature over zero (http://www.metoffice.gov.uk).

In contrast to the Mediterranean, British climate experience a much smaller temperature and precipitation range, consequently plant growth in roof condition could benefit of a better distributed rainfall throughout the year and of a less extreme environmental condition also on the roof.

Additionally, Great Britain, during the last decade, has been theatre of much progress and important steps made in the green roof field. Nowadays it can be defined as one of the most active countries within Europe on the roof greening theme.

Finally, because the prediction of climate changes gives researchers a new input to consider for their investigations, in the following studies, many potential genera for green roof application, originally from or well distributed in the Mediterranean area, were examined. Supplementary data was taken to look at both long term study of plant vegetation on roofs and short term study on plant germination in roof condition in order to widening the plant-growth in roof conditions.

4.1 STUDIES ON POTENTIAL GENERA FOR GREEN ROOF: EFFECT OF PLANT PROVENANCE AND ENVIRONMENTAL CONDITIONS IN A MARITIME CLIMATE

4.1.1 Abstract

Roof greening has recently become an important tool included in many local government strategies adopted to support sustainable urban ecosystems and improve urban dwellers' quality of life.

Even though there are various benefits derived from having vegetated roofs, the associated and significant environmental stresses - including drought, heat, wind speed and shallow substrate depth - which represent a limit for planners and a challenge for plant survival, must not be underestimated.

The present study has focused on plant selection. *Thymus* and *Sedum* exhibit characteristics potentially conducive to flourishing in a green roof environment were investigated. The majority of species belonging to these two genera grow in rocky and well-drained environments.

Several species of *Sedum* and *Thymus* were selected for this study. Among the selection a subset consisting of mostly Mediterranean native species, collected from different geographical areas, was included in the experiment to refine the study.

To highlight the desirable traits of planted species, their ornamental features and their growth responses to roof conditions in the UK climate, were recorded. Additionally, some Thymus species were also investigated in glasshouse conditions to see whether or not roof conditions are more limiting to plant growth compared to the optimal-standard environmental conditions found in the glasshouse.

Results concerning plant growth, ground coverage capacity, relative growth rate and dry biomass accumulation are presented and discussed in the present work.

In maritime conditions sedums seem to give the better growth performance in the roof environment. Particularly, *S.album* (ecotype from the lowest altitude) was the most successful. However, within the *Thymus* spp. some taxa distinguished themselves for their high growth and

establishment capacity. Results seemed to confirm the hypothesis that plant provenance has an influence on plant establishment and performance.

Key words: *Thymus*, sedums, green roofs, plant selection, native taxa, plant provenance

4.1.2 INTRODUCTION

Thymus is one of the eight most important genera in terms of number of species, within the Lamiaceae family. It comprises more than 300 species and subspecies, and more than 300 cultivars (Flora Europaea Database; http://www.thymus.co.uk/). *Sedum* (Crassulaceae family) *sensu lato* could contain as many as a thousand species and subspecies (Stephenson, 1994).

The focus and interest on *Thymus* and *Sedum* genera is not only due to their species richness and wide diversity; but also to their broad distribution and their native habitats, which are mostly sun-exposed poor shallow soils and well-drained or rocky soils (Tutin at al., 1964; http://www.thymus.co.uk/). In fact, because of the nature of substrates required for roof greening purposes, designed to be very freely draining, plants naturally adapted to these growing conditions are predicted to perform well or better than other highly demanding ornamental species.

Additionally, although cosmopolitan, these genera are better represented in the regions neighbouring the Mediterranean Sea and are found to grow naturally in all European countries (Stephenson, 1994; Hazzit *et al.*, 2006, Flora Europaea Database).

Many green roof studies had as their object *Sedum* growth and performance. However most of the studies published in English have been performed in other climates and continents, and very few in Europe. Concerning *Thymus*, despite some species being commonly used in green roof planting, no specific studies have been performed on this genus before

In this study the plant material was selected by examining its original habitat. European, and Mediterranean native species have been chosen from commercial sources and additionally non-commercial material has been wild-collected. Similar criteria were followed for the screening as are described in chapter 2 with main focus on *Thymus* and *Sedum* genera. The use of native taxa in urban design or in landscaping is an argument which periodically returns to be discussed. Nowadays, many authors recognize the potential and the ecological role which native plant communities play, especially in urban or anthropized environments (Monterusso *et al.* 2005;

Tinsley *et al.* 2006; Dunnett and Kingsbury, 2008). Native plants show good success in landscape design, they are adapted to the existing climate, resulting in a lower input requirement (fertilizer, pesticides and water); they have several positive ecological effects such as providing a natural habitat for native flora and fauna, and finally they provide regional character and identity. However, the use of native species is not to be considered exclusive or vital since using both native and non-native species seems to become increasingly important and especially in highly demanding environments such as green roofs they may have positive advantages (Dunnett and Kingsbury, 2008).

Concerning the green roof application, the potential of native species has also been investigated. The first known experiment focusing on native taxa for green roofs was the one conducted by Monterusso *et al.* (2005). Thereafter, other authors enhanced the use of native plants for roof greening with their studies. They emphasize the contribution of the native flora to the ecological function of the green roofs, and that these dry plant native species or even communities potentially exhibit all the characteristics to withstand the typical stresses on low-input extensive green roofs (MacDonagh *at al*., 2006; Martin and Hinckley, 2007; Sutton, 2008).

Tinsley *et al.* (2006) observed that few studies in landscaping do compare the establishment of native versus non native species. And when available these studies not usually test commercial versus non-commercial plant material for their performance.

Besides the greater effectiveness of using native material for extensive roof greening, we are convinced that plant differences in terms of growth/performance are also dictated by their provenance, where the most important changing factors are represented by the altitude and the growing conditions. With increasing altitude, irradiance and wind blow increase, while humidity decline. At higher altitudes plants have reduced leaf surfaces to limit transpiration. Furthermore, as altitude increase the dry season water stress decline as the number of species (Pugnaire and Vallardes, 2007; Lambert et al., 2008). Both *Thymus* and *Sedum* species, as explained before, grow over a wide environmental gradient all over Europe. Gurevitch *et al.* (1986) observed that in *Sedum wrightii* there is high variability in the effectiveness of CAM expression due to both genetic and environmental factors. Results from this study confirmed the hypothesis that the altitude of origin of the plants affected their response to drought stress and thus their capacity to uptake CO_2 during night time, resulting in a different growth capacity under stressful conditions.

It was noted that plant species including *Thymus* and *Sedum* species were growing in the local area around the test roof in Italy (Central Italy). They had a prostrate growth habit and grew in limited soil depth at a range of altitudes. It was speculated that these varieties of species grown widely in horticulture, were growing in conditions similar to those which are experienced in a rooftop environment. An experiment was set up to test the hypothesis that their growth form was determined by the environmental conditions they experienced at a local level, rather than an intrinsic difference in form at the genotype level.

They were removed from their physical environment in Italy, and transferred to a roof environment in a maritime climate, and a controlled environment in a glasshouse, where watering was optimal, and removal of water from the leaf surface was reduced by preventing desiccation by wind.

The experiment sought to compare the natural high stress environment at a range of altitudes, the high stress environment of a roof, and a low stress environment in a glasshouse.

The goal of this study was to assess the suitability of *Thymus* and *Sedums* wild types, species grown in cultivation and cultivars from different geographical areas and climates (mainly Mediterranean and maritime) for green roof planting in UK.

4.1.3 MATERIALS AND METHODS

Site description and experimental design

Two experiments were conducted at two different sites: a plant screening set up on the top of an 8 storey building (Education Dept. Building) at Sheffield University and a glasshouse screening performed at Sheffield City Council Norton Nursery in Sheffield, (53°38’N, 1°48’ W), between July and November 2010. Mean annual rainfall is 824 mm, quite uniformly distributed throughout the year (http://www.metoffice.gov.uk/climate/uk/).

Plugs were planted according to a completely randomized design. Randomization was performed using a random number generator (Research Randomizer). Five replicates, where one repetition was composed of three plants, for a total of fifteen individuals for each genotype were assessed in *Thymus* screening, while in *Sedum* three replicates (with three plants each) for each genotype were assessed.

Growth conditions

For both the plant screenings, trays of internal dimensions L73,5xW42,5xH16 cm were used. The growing medium consisted of a mix made up of 55% Zincolit® crushed brick, 30% pumice, 15% organic matter, a blend of 5% composted bark and 10% coir. Beneath the substrate a filter sheet SF and a drainage mat DVB-12 were placed to avoid water storage underneath the substrate and to provide an air gap. The substrate was 10 cm deep.

A total number of 15 plants, corresponding to 5 genotypes, were allowed to grow in each tray. Each tray was virtually divided in 3 rows along the shorter side and 5 columns across the longer side. For each genotype and tray 3 plants for each species were planted in one line (column) at a spacing of about 10,5 cm between plants and from the tray borders, while the distance between columns and from the tray borders was 12,5 cm.

Plant material

Plant material was propagated in the glasshouse of the Sheffield Botanical Garden in May 2010. Cuttings of 4,5-5,5 cm for thyme and 3-4 cm for sedums, were placed in 40 cell plug trays (cell dimensions were: 4 cm wide across the top, 3cm wide at the bottom and 5 cm deep) filled with the growing medium described above.

Thirteen species and cultivars of Thyme were chosen according to the following criteria: habit, origin and distribution, hardiness, soil requirements and the ornamental characteristics (foliage, flowers' colour) as are described in the table 4.1.

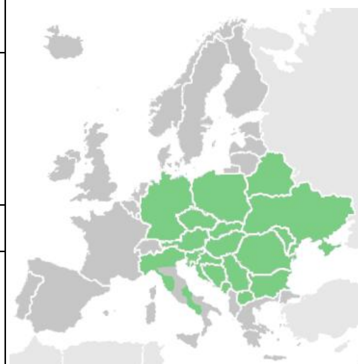
Thyme's screening on the roof included a total of 16 genotypes: *Thymus doefleri* 'Bressingham'; *T. herba-barona* 'Lemon scented'; *T. longicaulis*; *T. longicaulis* (wild collected at 730 m in Marche region, Italy); *T. longicaulis* (wild collected at 536 m in Marche region, Italy); *T. longicaulis* (wild collected at 600 m in Lazio region, Italy); *T. pannonicus*; *T. pseudolanuginosus; T. pulegioides*; *T. serpyllum*; *T. serpyllum* 'September'; *T. vulgaris*; *T. vulgaris* 'Compactus'; *T.* 'Pink ripple'; *T.* 'Purple beauty' (Coccineus Group); *T. '*Rasta'.

Amongst these the selection for the greenhouse screening was narrowed to only the species or its cultivar, when the pure species was not available, and was: *Thymus doefleri* 'Bressingham'; *T. herba-barona* 'Lemon scented'; *T. longicaulis*; *T. longicaulis* (wild collected at 730 m in Marche region, Italy); *T. longicaulis* (wild collected at 536 m altitude in Marche region, Italy); *T. longicaulis* (wild collected at 600 m in Lazio region, Italy); *T. pannonicus*; *T. pseudolanuginosus; T. serpyllum*; *T. Vulgaris*.

Concerning the sedum trial the species tested were a mix of European species, selected for hardiness, origin, and aesthetical value. Part of the plant material was wild collected from different geographical areas (Central Italy). Totally 8 genotypes were collected and tested. The list of sedums used, commercial and non-commercial material, and their provenance is reported in table 4.2.

Table 4.1: List of the *Thymus* genotypes selected and their morpho-physiological and pedo-climatic features.

Specie	Origin-Chorological type	Leaves and Flowers characteristics	General characteristics		Distribution map	Picture
***Thymus doefleri* All.'Bressingham'**	Cultivar, earliest known record 1956 in UK.	Elliptic, white-hairy, mid-green leaves. Purple-pink flowers, splashed dark crimson	**Altitude**	-		
			Habits/plant shape	Mat forming with prostrate stems.		
			Height	15 cm		
			Flowering season	May to June (It)		
			USDA Zone	3 to 9		
			Sun exposure and Soil requirements	Full sun; Well-drained soil; pH: 6.6-7.5 (neutral) to 8.6-9.0 (strongly alkaline)		
***Thymus herba-barona* Loisel. 'Lemon Scented'**	Cultivar wild-collected in Corsica	Small oval lemon-scented leaves. Pink flowers.	**Altitude**	-		
			Habits/plant shape	Mat forming with long prostrate stems		
			Height	10-15 cm		
			Flowering season	-		
			USDA Zone	6 to 8		
			Sun exposure and Soil requirements	Full sun; Well-drained soil; pH 6.6 - 7.5 (neutral) to 7.9 -8.5 (alkaline)		

Thymus longicaulis Presl.	Euro-Mediterranean	Oval dark-green leaves, glabrus, narrowly elliptic to linear spatulate. Rose-purple flowers.	**Altitude**	0-1600 m	 *Source:http://luirig.altervista.org/schedeit/index.htm*	
			Habits/plant shape	Flat, mat forming with long rooting prostrate stems, hardy dwarf shrub.		
			Height	3-6 cm		
			Flowering season	April to August (It).		
			USDA Zone	5 to 10		
			Sun exposure and Soil requirements	Full sun; Well-drained soil; pH: 6.6-7.5 (neutral)to 7.6-7.8 (mildly alkaline)		
Thymus pannonicus All. Hungarian Thyme [= T. pulegioides L. subsp. pannonicus (All.) Kerguelen]	South west and Central Europe, south Siberia region	Oval dark-green leaves. Mauve flowers.	**Altitude**	0-800 m (rarely 1800 m)	 *Modified from: http://luirig.altervista.org/schedeit/index.htm*	
			Habits/plant shape	Hardy, low growing shrub with long and prostrate stems.		
			Height	10-25 cm		
			Flowering season	May to August (It)		
			USDA Zone	-		
			Sun exposure and Soil requirements	Full sun; Well-drained soil.		

***Thymus* 'Pink Ripple'**	Earliest known record 1983 in Scotland.	Small pale-green hairy leaves. Pink flowers.	**Altitude**	-		
			Habits/plant shape	Mat forming with creeping stems		
			Height	10-15 cm		
			Flowering season	Mid summer		
			USDA Zone	5 to 9		
			Sun exposure and Soil requirements	Full sun; Well-drained soil; pH: 6.6 - 7.5 (neutral) to 7.9 -8.5 (alkaline)		
Thymus pseudolanuginosus Ronniger	It is of unknown origin.	Grey-green woolly leaves. Mauve-pink flowers.	**Altitude**	-		
			Habits/plant shape	Mat forming prostrate shrub with creeping-hairy stems.		
			Height	2,5 -5 cm		
			Flowering season	June to August (It)		
			USDA Zone	4 to 9		
			Sun exposure and Soil requirements	Full sun; Well-drained soil; pH 6.6 - 7.5 (neutral) to 7.9 -8.5 (alkaline)		

Thymus pulegioides L. Large thyme	Eurasiatic	Vary from small to large, either round or oval, green, and gold or variegated, lemon scented. Flowers from pale pink, through purple-pink to purple or white.	**Altitude**	0-2200 m	 *Source:http://luirig.altervista.org/schedeit/index.htm*	
			Habits/plant shape	Variable in growth habit, from dwarf shrub to large loose bush with long stems		
			Height	20-30 cm		
			Flowering season	July to August(UK) April to August (It)		
			USDA Zone	4 to 9		
			Sun exposure and Soil requirements	Full sun; Well-drained soil; pH 6.6 - 7.5 (neutral) to 7.9 -8.5 (alkaline)		
***Thymus* 'Purple Beauty'** (Coccineus Group)	Cultivar from the Netherland.	Small dark green leaves. Dark crimson flowers with darker centre	**Altitude**	-		
			Habits/plant shape	Bushy mat forming with creeping stems		
			Height	2-5 (7) cm		
			Flowering season	May to June		
			USDA Zone	8 to 9		
			Sun exposure and Soil requirements	Full sun; Well-drained soil; pH 6.6 - 7.5 (neutral) to 7.9 -8.5 (alkaline)		

Thymus serpyllum L. Breckland Thyme	Europe northwards from north east France, north Austria and North Ukraine.	Small oval-shaped green leaves, sometimes hairy. Pink, pale mauve, purple-pink to dark crimson flowers.	**Altitude**	0-2200 m	 *Source:http://luirig.altervista.org/schedeit/index.htm*	
			Habits/plant shape	Mat forming with creeping stems.		
			Height	15 cm		
			Flowering season	July to August (UK). April to August (It)		
			USDA Zone	5 to 10		
			Sun exposure and Soil requirements	Full sun; Well-drained soil; pH: 6.1-6.5 (mildly acidic) to 7.6-7.8 (mildly alkaline)		
Thymus serpyllum L. **'September'**	Cultivar from UK.	Small dark-green leaves. Purple-pink flowers, splashed claret.	**Altitude**	-		
			Habits/plant shape	Mat forming with long reddish prostrate stems		
			Height	-		
			Flowering season	(Late flowering)		
			USDA Zone	-		
			Sun exposure and Soil requirements	Full sun; Well-drained soil.		

***Thymus* 'Rasta'**	Cultivar from Germany.	Small overlapping leaves variegated cream and buff. Purple-pink flowers.	**Altitude**: - **Habits/plant shape**: Mat forming **Height**: 2-3 cm **Flowering season**: From June to July **USDA Zone**: - **Sun exposure and Soil requirements**: Full sun; Well-drained soil. -		
Thymus vulgaris L. Garden thyme	Native of the west Mediterranean region extending to south-east Italy – Western Steno-Mediterranean.	Grey-green or green; linear to narrowly oblong-elliptic in outline, 3-8x0.5-2.5 mm. White to pale pink flowers.	**Altitude**: 0-800 m **Habits/plant shape**: Small bushy shrub without procumbent stems rooting at nodes **Height**: 10 to 30 (-50) cm **Flowering season**: June to September (UK). May to June (It) **USDA Zone**: 4 to 8 **Sun exposure and Soil requirements**: Full sun; Well-drained soil; pH: 6.1-6.5 (mildly acidic) to 7.6-7.8 (mildly alkaline)	 *Source:http://luirig.altervista.org/schedeit/index.htm*	

***Thymus vulgaris* L. 'Compactus'**	Cultivar from Netherland.	Dark-green leaves. Mauve-pink flowers.	**Altitude**	-		
			Habits/plant shape	Bushy shrub, more compact in habit than the species		
			Height	15 to 30 cm		
			Flowering season	From June to July		
			USDA Zone	-		
			Sun exposure and Soil requirements	Full sun; Well-drained soil.		

(It)= Italy, (UK) = United Kingdom. USDA HARDINESS ZONES (range of average annual minimum temperatures): 4 = from −30 °C to −20 °C; 5= from −10 °C to −20 °C; 6=from −10 °C to 0 °C; 7= from 0 °C to 10 °C; 8= from 10 °C to 20 °C; 9= from 20 °C to 30 °C; 10= from 30 °C to 40 °C.

For the nomenclature the following sources have been consulted: Conti *et al.* (2005), Pignatti (1982), the International Plant Name Index, the Index Synonymique de la flore de France, the Royal Horticultural Society, Italian Mycological and Naturalistic Telematic Association, USDA Germplasm Resources Information Network. For other information: Pignatti (1968), Sell P. and Murrel G.(2009), USDA Germplasm Resources Information Network, http://davesgarden.com/, http://www.esveld.nl/htmldia/t/thppbe.htm, http://www.thymus.co.uk/, http://zipcodezoo.com/.

Table 4.2: List of *Sedum* chosen for this study with habitat and provenance details

Specie	Habitat	Provenance
Sedum album L.	European-Mediterranean	Commercial
Sedum album L.	European-Mediterranean	Lazio, Civita Castellana, VT (150 m)
Sedum album L.	European-Mediterranean	Marche, Leccia, Serra Sant'Abbondio, PU (536 m)
Sedum album L.	European-Mediterranean	Marche, Fonte Avellana, PU (730 m)
Sedum sexangulare L.	Eastern Mediterranean	Commercial
Sedum sexangulare L.	Eastern Mediterranean	Marche, Leccia, Serra Sant'Abbondio, PU (536 m)
Sedum sexangulare L.	Eastern Mediterranean	Marche, Fonte Avellana, PU (730 m)
Sedum sexangulare L.	Eastern Mediterranean	Abruzzo, Scanno, AQ (1200 m)

Sources: Pignatti (1982); Stephenson (1994)

Measurements

All measurements were taken on plants grown in roofs as in glasshouse conditions.

To obtain from each taxa under study the original biomass the shoot dry weight of an initial sample was obtained oven drying for 72h at 80°C and measured with a precision scale balance 10 cuttings for each species at the time of propagation (30 May 2010). Equally, at the end of the experiment (November 2010), all plants were harvested to determine the dry weight of shoots (oven-drying for 72 h at 80°C).

To compare performance of species the Relative Growth Rate, known also as efficiency index, was determined on dry shoot weight according to Hunt (1990):

$$RGR = (\log_e W_2 - \log_e W_1)/(t_2 - t_1)$$

Plant height and plant diameters were measured every 15 days on all plants. Height (H) was determined as the distance from the surface of the medium to the top of the plant, while width (diameter, W) as the average of two measurements (one perpendicular to the other). According to Sloan *et al.* (2003) plant growth index (GI) was calculated as:

$$GI = \pi\left(\frac{W}{2}\right)^2 H$$

Percentage of plant coverage was determined visually.

Statistical analyses

All data was statistically analyzed using SPSS 16.0 software package for Microsoft Windows Vista.

ANOVA analysis of variance was performed using a General Linear Model both Univariate and Repeated Measures. Combined analysis of variance was performed using species as fixed variable. The result of ANOVAs was subjected to the Duncan's new multiple range test at P = 0.05 on each of the significant variables measured. T-test independent sample was performed assuming means not equal to compare plant growth under different conditions.

4.1.4 RESULTS AND DISCUSSION

THYMUS SCREENING

During three months of observation, plant growth between taxa significantly differed (P<0.001) between roof conditions and glasshouse conditions. The graphs to follow (Fig.4.1 and Fig 4.2), show taxa development in different environmental conditions. As expected *Thymus* spp. in glasshouse conditions seemed to perform better than in green roofs conditions, showing overall at the end of the experiment higher growth (755.26 cm^3 roof vs. 2624.65 cm^3 glasshouse). Particularly, the taxa affected by the growth environmental conditions which showed a strong growth decrease on the roof were: *T. longicaulis* (600m) (-713%), *T.vulgaris* (-403%), *T. pannonicus* (-265%), *T. longicaulis* (536m) (-256%) and *T. longicaulis* (commercial) (-119%).

It is noticeable that all genotypes had different values of growth index when transplanted, since all plants were grown form cuttings of the same dimensions all the differences highlighted are due to the plants growth form. *Thymus* 'Pink Ripple' registered during the trial a substantial decrease in growth, plants had desiccation of some shoots.

Figure 4.1: Growth Index pattern of *Thymus* spp. in roof and glasshouse conditions from July to October 2010.

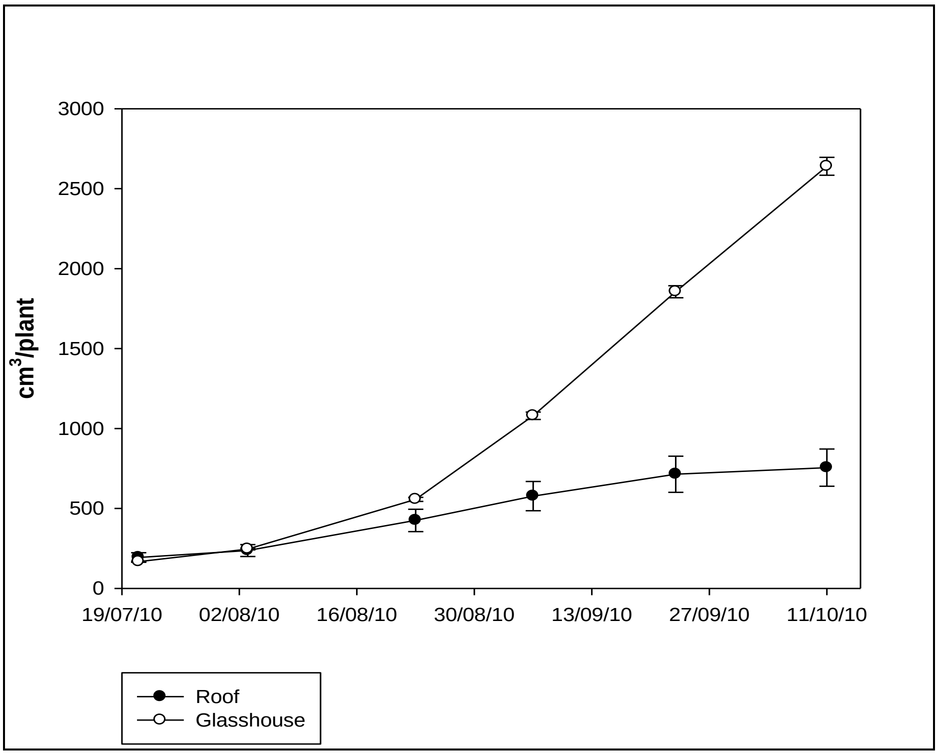
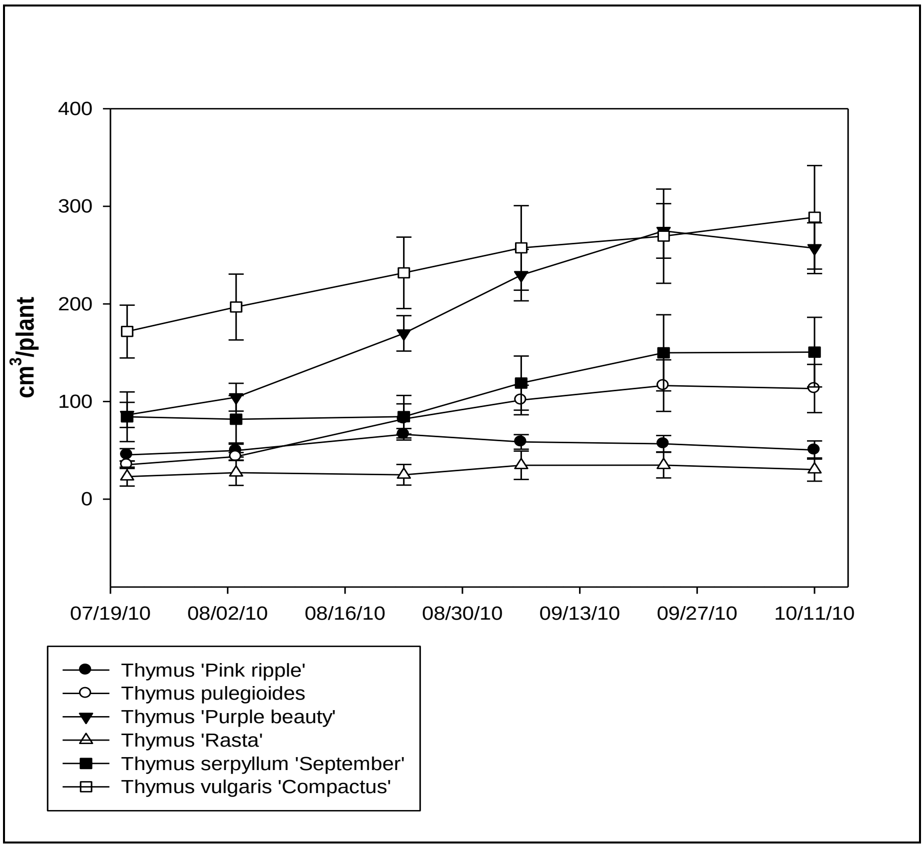


Fig 4.2 Growth Index pattern of *Thymus* spp. grown only in roof conditions from July to October 2010.



In roof conditions, *Thymus longicaulis* (commercial form) was absolutely the most vigorous, its growth index was 2,402 cm^3/plant by October. On the other hand, the varieties 'Rasta' and 'Pink ripple' and the species *T.pulegioides* recorded the lowest growth values (30.2, 50.3 and 113.3 cm^3/plant) as well as the lower biomass accumulation and relative growth rate (Tab. 4.3).

Table 4.3: Growth Index (11/10/2010), Shoot dry biomass and Relative Growth Rate (RGR) of *Thymus* spp. tested in roof conditions. Means separated by Duncan's multiple range test within columns.

Taxa	Mean Growth Index	Std. Error +/-	RGR
***Thymus* 'Pink ripple'**	50,27 a	9,67	0,0057 a
***Thymus* 'Purple beauty'**	257,10 ab	45,63	0,0169 c
***Thymus* 'Rasta'**	30,22 a	378,04	0,0080 a
***Thymus doefleri* 'Bressingham'**	158,43 ab	34,69	0,0182 cd
***Thymus herba-barona* 'Lemon scented'**	264,55 ab	69,56	0,0266 g
Thymus longicaulis	2401,97 d	34,89	0,0266 g
***Thymus longicaulis* (536m)**	626,08 b	382,56	0,0229 f
***Thymus longicaulis* (600m)**	465,01 ab	9,27	0,0221 ef
***Thymus longicaulis* (730m)**	428,94 ab	75,18	0,0198 de
Thymus pannonicus	1631,60 c	24,70	0,0266 g
Thymus pseudolanuginosus	186,63 ab	26,00	0,0225 ef
Thymus pulegioides	113,26 a	11,88	0,0112 b
Thymus serpyllum	128,23 ab	37,79	0,0233 f
***Thymus serpyllum* 'September'**	150,58 ab	35,62	0,0185 cd
Thymus vulgaris	1261,19 c	53,06	0,0273 g
***Thymus vulgaris* 'Compactus'**	288,70 ab	199,58	0,0236 f
Significance	***		***

*** Significant at P<0.001. Mean values sharing different letters differ at P<0.05.

Overall, cultivars showed lower growth habit compared to the related species and to the other species as well, in fact they were selected for their compact habit, hence the differences found are due to plants' habit. Regardless of that, most of them showed good biomass accumulation and relative growth rate. Particularly, *T.herba-barona* had a high relative growth rate which was comparable to the one recorded for the most vigorous taxa *T.vulgaris, T. longicaulis* (commercial) and *T. pannonicus*, while *T.*'Pink ripple' had the lowest biomass accumulation and relative growth rate. Furthermore, *T. longicaulis* (commercial), *T.vulgaris, T.*'Purple beauty' and *T. pannonicus* had also higher gain in shoot dry biomass.

Concerning *T.longicaulis* wild genotypes, the ones from lower altitudes, particularly the ecotype from 536 m, showed a better response to the roof conditions compared to the others, and also a good growth capacity when compared to the other taxa.

Among the plants grown in glasshouse conditions the most vigorous species were: *T.vulgaris, T.pannonicus* and *T.longicaulis* (commercial) (Tab. 4.4). In this growth environment the difference between *T.longicaulis* and local ecotypes and commercial genotype was less strong; particularly the ecotype from 600 m altitude had a higher growth than on the roof, which was comparable to the commercial *T.longicaulis,* as well as growth responses within the wild-form group were less significant to the ones found on the roof.

Table 4.4 : Growth Index (11/10/10) Shoot dry biomass and Relative Growth Rate (RGR) of *Thymus* spp. tested in glasshouse conditions. Means distinguished by Duncan's multiple range test within columns.

Taxa	Mean Growth Index	Std. Error +/-	RGR
Thymus doefleri 'Bressingham'	218.96 a	62,91	0,0120 a
Thymus herba-barona 'Lemon scented'	189.18 a	63,39	0,0144 a
Thymus longicaulis	5279.38 de	967,24	0,0252 cde
Thymus longicaulis (536m)	2231.96 bc	620,43	0,0233 bcd
Thymus longicaulis (600m)	3780.36 cd	339,03	0,0264 de
Thymus longicaulis (730m)	1849.97 ab	403,49	0,0219 bc
Thymus pannonicus	5965.76 e	1257,96	0,0249 cde
Thymus pseudolanuginosus	51.54 a	7,63	0,0147 a
Thymus serpyllum	337.88 a	79,18	0,0208 b
Thymus vulgaris	6352.54 e	841,57	0,0281 e
Significance	***		***

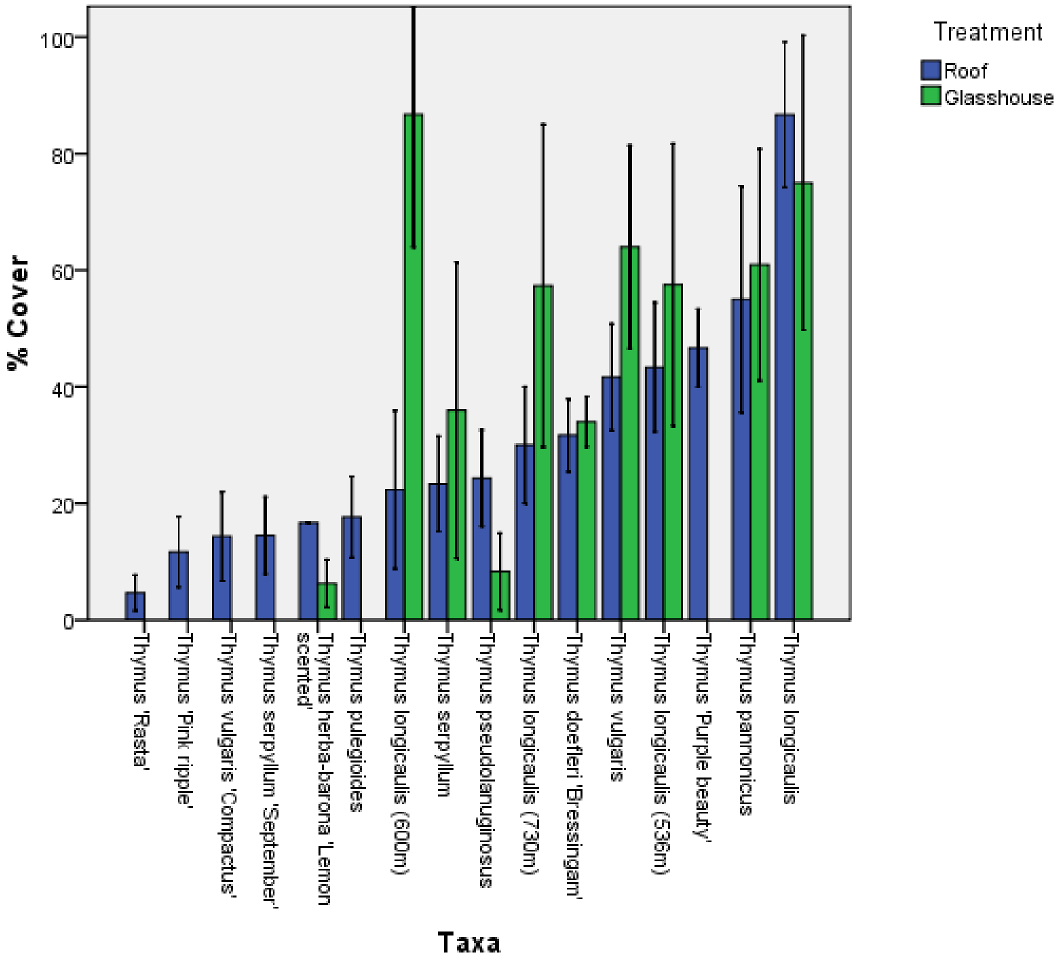
*** Significant at P<0.001. Mean values sharing different letters differ at P<0.05.

The relative growth rate in green roof conditions was significantly greater (P<0.001) than those in the glasshouse (0.24 vs. 0.21), while shoot dry biomass was not affected by the environmental conditions. Taxa and growth conditions had a significant effect on the relative growth rate as on the shoot dry biomass accumulation (P<0.001). Particularly, *Thymus doefleri* 'Bressingham', *T. herba-barona* 'Lemon scented', *Thymus longicaulis* (600m) and *Thymus pseudolanuginosus* showed to be strongly influenced by the different growth environments. These species showed an increase in their efficiency index (RGR) and their above-ground dry biomass in those conditions which were expected to be more limiting, the roof ones.

Nonetheless, it has to be noted that the majority of plants grown in glasshouse showed pathological symptoms, this was probably due to the lack of ventilation and the favourable conditions to cryptogam pathogens flourishing found in glasshouse environment. In general, plants were attacked by caterpillars (Lepidoptera larvae), which in some cases caused severe loss of leaves, and showed desiccation and rot symptoms. Despite this, the most vigorous taxa reached sufficiently large plant dimensions that the damage did not affect growth significantly, whereas the lower growing taxa could not recover sufficiently. Finally, it is worth pointing out that *T.vulgaris* did not show any pathological symptoms as did *T.longicaulis* (600m).

Independently from the growth conditions, ground cover was significantly different between taxa (P<0.001), and when environmental conditions were compared, taxa (P<0.001), growth condition (P<0.01), and their interaction (P<0.001) effects were significant. Plants grown in the glasshouse showed medially a higher coverage capacity than those on the roof (55% vs. 30%). Particularly, *T. herba-barona* 'Lemon scented', *T. longicaulis* (600m) and *T. pseudolanuginosus* were, also in this case, the taxa most affected by the growth conditions. This is shown in the following bar chart (Fig.4.3). While in *T.longicaulis* (600 m) the glasshouse environment was revealed to be optimal for growth (86% recorded in glasshouse conditions vs. 23% in roof conditions), for the other two taxa it was not, since they conversely showed higher performance in the roof conditions (6% vs. 17% for *T. herba-barona* and 8% vs. 24% for *T. pseudolanuginosus*).

Figure 4.3: Mean percentage of cover of *Thymus* taxa grown in two different environmental conditions (Roof and Glasshouse)



Concerning cover percentage recorded on the roof, the variety 'Rasta' had the lowest coverage (4.7%), while *T.longicaulis* (commercial) reached the highest level (87%) followed by *T.pannonicus* with 55%, 'Purple beauty' (47%) and T.longicaulis (536m) (43%).

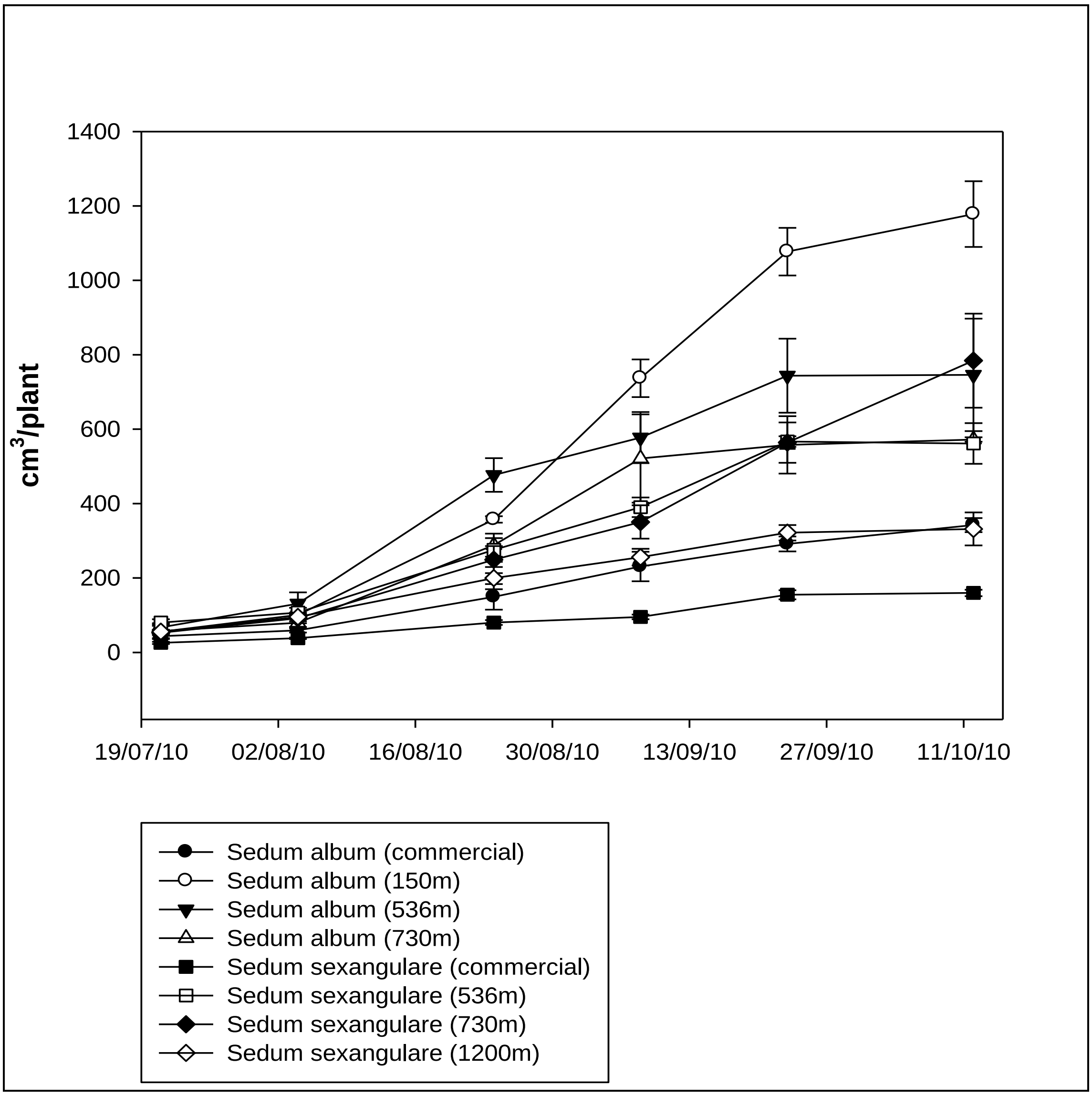
In glasshouse conditions *T. herba-barona* 'Lemon scented' and *T. pseudolanuginosus* had the lowest growth (6.2% and 8.3%), while *T.longicaulis* (600m) had the highest growth (87%).

T.longicaulis collected in Lazio region from 600 m altitude showed a reverse trend compared to the others genotypes with other provenances. It was observed that this taxon and *Thymus vulgaris* did not show parasitic symptoms as the other taxa grown in glasshouse conditions. It may be explained by the fact that as aromatic plants *Thymus* spp. contain essential oils which possess many beneficial characteristics and can be distinguished also on the basis of their phytochemical properties (Hazzit et al., 2006). Moreover, it has been demonstrated that chemical composition of the essential oils may vary from species to species and within species with provenance (Hazzit *et al*., 2006). Because of their antibacterial and insecticidal properties *Thymus*' essential oils are widely-used as green healthy-friendly medical compound and pesticides (Hazzit *et al.*, 2006; Koul *et al*., 2008). Thus probably in the glasshouse *T.longicaulis* ecotype from 600m altitude as also *T.vulgaris* could express completely their growth potential probably through their chemical characteristics which protected plants from biotic stress.

SEDUMS SCREENING

Plant growth in sedums changed significantly over time (Fig.4.4). Sedum sexangulare made little progress (55 cm3/plant to 160 cm3/plant), while S.album collected from 150m altitude reached by the end of the experiment the highest growth index (55 cm3/plant to 1,178 cm3/plant).

Figure 4.4: Sedums growth index pattern from transplant (July 2010) to October 2010.



All sedums performed well, showing a rapid and higher growth capacity compared to the *Thymus* taxa as a whole, and reaching by October higher coverage percentages (52% vs. 45%) (Tab.4.5). At the end of the experiment the taxa which showed overall lowest growth performance were: *Sedum album* and *sexangulare* of commercial provenance and the *S.sexangulare* from higher altitude (1200m). On the other hand, *S. album* from 150 m of altitude had the greatest ability to grow and rapidly cover the substrate in the roof conditions.

Table 4.5: Growth Index (11/10/2010) Shoot dry biomass and percentage of cover of sedums. Means distinguished by Duncan's multiple range test within columns.

Taxa	Mean Growth Index	Std. Error +/-	Mean Shoot dry biomass	Std. Error +/-	Cover (%)	Std. Error +/-
Sedum album **(commercial)**	342,25 ab	18,82	2,26 ab	0,56	44,45 ab	2,78
Sedum album **(150m)**	1178,22 d	5,84	5,98 c	0,11	72,78 cd	18,89
Sedum album **(536m)**	746,15 c	88,31	5,52 c	0,44	83,33 d	0,00
Sedum album **(730mt)**	572,31 bc	151,17	3,86 b	0,27	65,00 bcd	7,64
Sedum sexangulare **(commercial)**	160,02 a	4,77	1,97 a	0,02	30,55 a	1,52
Sedum sexangulare **(536m)**	561,44 bc	29,90	3,52 ab	0,15	66,67 bcd	0,00
Sedum sexangulare **(730m)**	784,22 c	69,21	3,38 ab	0,24	72,22 bcd	4,03
Sedum sexangulare **(1200m)**	331,91 ab	24,21	2,07 a	0,21	50,00 abc	4,57
Significance	***		***		**	

***, ** Significant at P<0.001 and P<0.01. Mean values sharing different letters differ at P<0.05.

It is arguable that provenance and altitude may have an influence on plant physiology and phenology. P'yankov and Kondrachuk (2003) observed that in different groups of alpine plants (xerophytes, subalpine and mesophyte), the plants' leaf structure change depending on the altitude and on the group as well. Thus characteristics which affect photosynthesis differ over a range of altitude due to a progressive plant adaptation to the mountain conditions. Similarly, another study conducted on different populations of CAM *Sedum wrightii* found that the plant resistance and responses to imposed drought changed significantly within this species relatively to the plant provenance (Gurevitch *et al.*, 1986).

These preliminary findings, especially regarding sedums, seems to support what has been found also by Gurevitch et al. (1986), where the populations from lower latitudes seemed to tolerate the roof conditions more than the plants from higher altitudes, with higher growth rates, dry biomass gain, and coverage. Despite it could be expected that population from higher altitudes tolerate more the roof environment than the ones from lower altitudes, however it has to be taken in consideration that at higher altitude dry season water stress decrease (rainfall is higher) and temperatures tend to be lower than in lowlands (Lambers *et al.,* 2008). Hence plant tolerance to drought, which in summer is also associated with heat and temperature rising, and morpho-physiological characters associated to it could be probably found more at lower altitudes. Van de Water *et al.* (2002) conducted an experiment testing several C_3 taxa for the ratio of ^{13}C to ^{12}C fixed during photosynthesis, the authors pointed out that this ratio, which is strictly associated to stomatal resistance and thus plant capacity to reduce water loss, decrease with elevation.

However these results have to be further investigated in a longer term perspective and also a wider range of species and genera have to be screened to confirm such findings.

4.1.5 CONCLUSION

It can be concluded that in maritime conditions sedums seem to give the better growth performance in the roof environment. Particularly, *S.album* (Italian ecotype from 150m altitude) was the most successful.

However, within the *Thymus* spp. some taxa distinguished themselves for their high growth and establishment capacity, which was comparable with the greater ones recorded for *Sedum* species. Particularly, the *T.longicaulis, T.pannonicus* and *T.vulgaris* with commercial provenance could be highly recommended.

It was shown that among the *Thymus* genus the native taxa tested were not necessarily more flourishing than the other non-native taxa in roof conditions as in the theoretically optimal glasshouse conditions. Furthermore the glasshouse environment is not always more suited to plant growth potential expression.

Results confirmed the hypothesis that plant provenance has an influence on plant establishment and performance and shown that material from lower altitude seems to be more appropriate for green roof planting. Therefore the results suggest that wild collected material of low altitude provenance seems to fulfil tolerance and growth requirements for green roofs better than the higher altitudes and in some cases even appreciably more than the commercial material. Due to these considerations, a more detailed investigation of the relationship between altitude and green roof hardiness or moisture stress tolerance would be one area for further work. Furthermore, through the glasshouse experiment was demonstrated that also the local material when grown in optimal conditions exhibit its growth potential, which does not normally occur in natural as in the roof condition, since plant adaptation to limiting condition lead plants to change in form (i.e. prostration) and growth.

Of course, more research is needed in order to support and further investigate the results of this short-term study. In addition, commercial market and plant production has to be sensitive to this aspect, which appears to be non neglectable for this kind of application.

4.2 SUBSTRATE DEPTH EFFECT ON PLANT GERMINATION IN GREEN ROOF CONDITIONS

ABSTRACT

Compared to a natural ground-based ecosystem green roofs have a limited "soil profile", limited resources, and limited water storage capacity. The main cause is their disconnection from the ground and deeper soil horizons. Those limitations, besides the exposure to extreme temperature fluctuations, wind and drought, represent a pivotal factor for plant establishment and growth.

Data available on plant establishment on green roofs is relatively scarce and has often referred only to the genus *Sedum*. Characterization of a wider range of green roof plants is thus required.

Although sowing is one of the preferred methods adopted in establishing green roofs, few studies have been conducted to investigate germination response of green roof species in a green roof environment.

The aim of the project was to provide systematic data on the growth performance of species which are typically grown on green roofs. A green roof experiment was set up to test whether the germination of the 42 species selected, was effected by the substrate depth.

Plant were sown on modules with three different substrate depths (5,10 and 15 cm) and placed on the rooftop of a three-story building.

Here preliminary data on germination is presented. Results show the significant influence of the substrate depth on seed germination, and also the variability of response to this factor across the species.

Key words: Green roofs, substrate depth, germination, perennials

4.2.1 INTRODUCTION

Plant selection has been defined as an extremely important factor for green roofs' success (Getter and Rowe, 2006). However, experiences from several research studies suggest that not only the species choice is important, but also design construction characteristics as substrate composition and depth strongly influence the green roof performance (Getter and Rowe, 2006;

Dunnett et al., 2008b; Simmons and Lady Bird Johnson Wildflower Center, 2010). Many findings suggest to do not create green roofs with substrate depth too much shallow and to possibly vary the depth so that different micro-habitats are created on the roof. These microhabitat may help a wider range of organisms, including plants, to survive (Brenneisen, 2006; UNI, 2007; Dunnett and Kingsbury, 2008).

As described by several authors there are different ways to establish vegetation on green roofs: seed sowing (wet or dry); application of cuttings; planting of plugs or pot-grown plants or root ball plants; laying of pre-cultivated vegetation mats; and, finally, spontaneous colonization (Dunnett and Kingsbury, 2008; FLL, 2008). Depending on the design and the plant selection, it is possible to combine any of these above-mentioned methods.

The world-wide dominating sedum mats are commonly and easily installed via seeding, cuttings or even via pre-cultivated mats. However, because of its lower cost and lower labour requirement, sowing and cuttings are the most diffuse and cost-effective way to plant a green roof, especially in the case of large roofs and extensive green roofs (Dunnett and Kingsbury, 2008).

Few published research studies investigate the effect of the substrate depth on plant growth and establishment in green roofs systems. In all the cases plants were planted from cuttings, plugs or pot grown plants (Dunnett and Nolan, 2004; Durhman *et al.*, 2007; Dunnett *et al.*, 2008; Getter and Rowe, 2008; Thuring *et al.*, 2010); and when planted from seeds no attention to plant performance and germination was paid (VanWoert *et al.*, 2005). However, when plant growth is concerned deeper substrates have been shown to promote plant survival, growth and species richness (Dunnett and Nolan, 2004; Durhman *et al.*, 2007; Dunnett *et al.*, 2008; Getter and Rowe, 2008; Dunnett *et al.*, 2010). Besides that, deeper substrates also decrease the water runoff, having greater water holding capacity (VanWoert *et al.*, 2005; Mentens *et al.*, 2006).

Moreover, although the germination response to sowing depth has been investigated (Martinez *et al.*,1992; Martinez *et al.*,2002), and despite sowing is a diffuse way to plant green roofs, no studies have been performed on the influence of the substrate depth on seed germination, neither on green roof conditions nor using green roofing substrates.

Plant seed germination is a complex aspect of plant physiology, involving many eco-physiological aspects. Physiologically, the germination starts with the water adsorption by the seed, through a process called imbibition. Consequently, the fundamental factor allowing seed to germinate is water (Alpi *et al.*, 2000). To a great extent, supporting the vital role played by water in the germination process, Dasberg and Mendel (1971) observed the decrease in germination rate and the final germination percentage in *Oryzopsis holciformis* with increasing water stress.

Following their dissemination from the plant, seeds exhibit one of these three conditions: they may be non-dormant and germinate immediately; non-dormant and quiescent; or dormant. The dormancy condition depends entirely on the seed itself, therefore on the species and their inherited properties; while in non-dormant species the seed quiescence depends on the environmental conditions (i.e. water availability, temperature, etc.) (Baskin and Baskin, 1998; Alpi *et al.*, 2000). Generally, in temperate regions, temperature is the factor regarded as the main ones regulating seeds dormancy (Baskin and Baskin, 1998). Additionally, beside water availability, temperature is confirmed to be the main factor affecting seeds germination also in arid and semi-arid regions (Bonvissuto and Busso, 2007).

Arguably, not only the depth may influence the water content of a substrate but also its composition and structure. It is also true that the matric potential, being directly related to soil texture and structure characteristics and being the common expression of soil moisture content, significantly influences seeds germination (Dasberg and Mendel, 1971; Giardini, 1992).

It was thus hypotesized that substrate depth may affect plant establishment from seeds, where lower substrate depths are expected to do not promote as deeper substrates seeds germination.

The aim of the present work was to provide information about seed germination of a broad range of plant species widely used to plant green roofs on green roof conditions. Therefore due to the previous considerations, this study was designed to evaluate the effect of green roof substrate depth on germination and initial plant establishment of 42 perennial species, forbs and grasses, most of which have never been tested in a maritime climate nor in green roof environmental conditions.

4.2.2 MATERIALS AND METHODS

Experimental site

The study was conducted on the top of a three-story building at Sheffield University, in Sheffield, between July and September 2010 (53°38'N, 1°48' W). Mean annual rainfall is 824 mm quite uniformly distributed throughout the year (http://www.metoffice.gov.uk/climate/uk/).

Plant material

The species selected for this study are among those most commonly found on green roofs. Species are in total 42, all of which are perennials, distinguishable in 32 forbs and 10 grasses (Tab.4.6).

Table 4.61: Taxa studied and their main phenotypical characteristics.

Species	Height (cm)	Flower Colour
Achillea millefolium (wild form)	40	white
Aethionema grandiflorum	20	pink
Armeria maritima	15	rose - red
Calamintha nepeta	40	light blue - pink
Campanula rotundifolia	30	light blue - pink
Centranthus ruber var. coccineus	60	red
Chrysanthemum leucanthemum	60	pure white
Delosperma cooperi	10	brilliant yellow
Dianthus carthusianorum	40	purple - red
Dianthus deltoides	20	dark rosy - red
Echium vulgare	80	bright blue
Galium verum	30	yellow
Jasione montana	20	blue
Knautia arvensis	80	dark red
Leontodon autumnalis	20	yellow
Linaria vulgaris	75	yellow with orange
Lotus corniculatus	10	yellow
Oenothera caespitosa spp. *eximia*	20	white
Origanum vulgare	35	pale purple
Penstemon pinifolius	20	orange - red flowers
Petrorhagia saxifraga	20	pink
Prunella vulgaris	20	violet
Sedum acre	6	yellow
Sedum album	10	white
Sedum kamtschaticum	15	orange - yellow
Sedum rupestre	30	yellow
Sedum sexangulare	8	lemon yellow
Silene uniflora	15	mostly white

Talinum calycinum	10	neon - pink
Teucrium chamaedrys	30	pink
Thymus pulegioides	25	purple
Veronica spicata	60	blue

GRASSES

Species	Height (cm)	Flower Colour
Anthoxanthum odoratum	30	-
Briza media	50	-
Carex flacca	25	-
Festuca amethystina	20	-
Festuca glauca	25	-
Festuca ovina	25	-
Koeleria glauca	25	-
Melica ciliata	40	-
Poa alpina	10	-
Sesleria caerulea	25	-

Germination trial and experimental design

Seeds of 42 perennial species were sown according to a completely randomized design on a substrate mix made up of 55% Zincolit® crushed brick, 30% pumice, 15% organic matter, a blend of 5% composted bark and 10% coir. Trays of internal dimensions L73,5xW42,5xH16 cm were used as planting units. A drainage layer DVB-12 was placed at the bottom of the tray, and over this was placed a filter sheet, both were placed underneath the substrate layer.

Plants were sown on trays with three different substrate depths (5, 10 and 15 cm). To maintain uniformity between trays in the upper surface of the substrate, the depth was adjusted by inserting a false base beneath the drainage layer, created using polystyrene sheets. Four replicates plant trays for each taxon were assessed.

A total number of 28 plants were allowed to grow in each tray, at a spacing of 10 cm distance between plants. Randomization was performed using a random number generator (Research Randomizer). Twelve positions in total were assigned to each species, thus 240 seeds were counted and further split up into 12 portions, having approximately 20 seeds per species and sowing point. To facilitate the sowing activities a sowing template made up of cardboard was created. Finally, to complete the sowing a layer of fine particles was placed over the top in order to help seeds germination.

Measurements

The number of total seedlings germinated was counted every week for one month. Percentage of germination was calculated with the following formula:

$$(n°of\ seedlings\ germinated/n°of\ seeds\ sowed) * 100$$

Statistical analysis

All data was statistically analyzed using SPSS 16.0 software package for Microsoft Windows Vista.

ANOVA analysis of variance was performed using a General Linear Model, defining species and depth as fixed variables. The result of ANOVAs was subjected to the Duncan's new multiple range test at P = 0.05 on each of the significant variables measured.

4.2.3 RESULTS AND DISCUSSION

By the second week from sowing, plant germination was significantly affected by taxon, substrate depth and their interaction. Averaged over taxa the percentage of germination ranged from a minimum of 16% recorded in 5cm depth, to 23% recorded in 10 cm depth, to a maximum of 26% recorded in the deepest trays. Except taxa which were not affected at this relatively early stage of germination by substrate depth variation, the other taxa showed overall significant increase in germination rate as the substrate depth increased (Tab 4.2). Among taxa *Achillea millefolium* (54%), *Chrysanthemum leucanthemum* (60%), *Petrorhagia saxifraga* (63%) and *Poa alpina* (54%) had greatest percentage of germination, while the lowest value was recorded for *Teucrium chamaedrys* (0.7%), followed by *Carex flacca* (1.8%), *Oenothera caespitosa* (1.5%) and *Delosperma cooperi* (1.6%).

It is also noteworthy that within sedums, *S.acre* recorded the highest germination at 2 weeks from sowing, and that as the substrate depth was reduced the greater was its germination (45% at 5cm, 36% at cm10, 26% at 15cm). It is well-known that sedums grow in very shallow and well-drained soils, and the shallower substrate fulfilled strongly these requirements. Furthermore, it is possible to consider that the 5cm depth tray reached higher soil temperature than the deeper ones, a condition which may had promoted sedums germination. It has been found in *Sedum* seeds belonging to two different species that when sowed immediately after seed collection both germinated at relative low temperature (around 15°C), while when stored, as the length of storage was prolonged the optimal temperature for germination increased to 25°C (Kentaro *et al*., 2006). Mean temperature value of August in Sheffield is reported to be around 15°C (http://www.worldclimate.com), thus it is possible to think that *Sedum acre* seeds germination was positively affected by the 5cm depth for the depth-related increasing temperature besides possible seed storage-handling factors (seeds were provided by a German seed company Jelitto Perennial Seeds).

After four weeks from sowing, germination was again affected by the factor taxa, substrate depth, and the interaction between taxa and depth. By this time most of the species medially reached ≈50% of germination (Tab.4.7). In general, deeper substrates confirmed to promote germination, 45% at 5cm, 49% at 15 cm, 50% at 10 cm, and over time the effect between the deeper substrates was comparable. Over time there were significant differences between taxa in terms of germination capacity, where *Talinum calycinum* (5%), *Carex flacca* (8%), *Oenothera caespitosa* (6%) and *Delosperma cooperi* (7%) confirmed their low germination rate, whereas grasses *Festuca glauca* and *Poa alpina* had the higher germinations (88% and 83%, respectively) and performed greater in all type of substrates. Some taxa in two weeks time notably increased the number of seedling emerged reaching by the 4th week significantly high percentage of germination and were: *Sesleria caerulea* (92% increase, 72% germination), *Festuca amethystina* (88% increase, 69% germination), *Campanula rotundifolia* (78% increase, 64% germination), *Knautia arvensis* (73% increase, 64% germination), *Festuca glauca* (69% increase, 88% germination), *Prunella vulgaris* (69% increase, 73% germination).

Concerning the taxa with scarce germination, *Talinum calycinum* normally require more time than other species to germinate (30-90 days) (http://b-and-t-world-seeds.com), while *Carex flacca* has been studied as a seed dormant species, having a combination of a strict or conditional primary dormancy, a light requirement for germination, low germination at constant temperatures, a positive response to diurnal temperature fluctuations and an induction of secondary dormancy in late spring by increasing environmental temperatures and showed to be able to germinate after one year from sowing in normal conditions (Shutz, 1998; Shutz, 2000). On the other hand, *Oenothera caespitosa* and *Delosperma cooperi* are usually considered as easy species to grow and quite fast to germinate, 10-40 days and 15-30 days. However, temperatures required for a good germination are 20 and 23°C, respectively. The value of germination recorded in *Oenothera* and *Delosperma* at 5cm depth was in both cases twice the one recorded in deeper trays, this may give support to the thesis that temperature found in the shallower trays was probably more favourable to germination for some species. But, for these species temperature or environmental conditions were perhaps not sufficient for a good germination. As explained before, seed provenance and storage can also affect seeds germination, thus, the results found for the species under discussion might lead to think that some variations in their germinability could have occurred previous to the trial.

Table 4.7: Mean percentage of germination of taxa at 5cm, 10cm, and 15cm depth at two and four weeks from sowing.

Taxa	2nd week					Taxa	4th week				
	% Germination Mean		Depth				% Germination Mean		Depth		
			5cm	10cm	15cm				5 cm	10 cm	15 cm
Teucrium chamaedrys	0,69	a	5,33	1,78	2,08	*Carex flacca*	7,74	a	16,54	5	1,67
Carex flacca	1,81	ab	5	1,87	0,42	*Talinum calycinum*	4,8	a	8,46	4,29	1,67
Delosperma cooperi	1,56	ab	3,85	1,78	0,83	*Delosperma cooperi*	6,87	ab	10,71	5,71	4,17
Oenothera caespitosa spp. eximia	1,54	ab	3,08	0,71	0,83	*Oenothera caespitosa spp. eximia*	6,4	ab	9,62	5	4,58
Silene uniflora	1,79	ab	3,46	1,07	0,83	*Teucrium chamaedrys*	11,24	bc	13,08	13,57	7,08
Talinum calycinum	2,94	ab	4,17	4,64	2,22	*Echium vulgare*	11,34	bc	13,85	8,93	11,25
Jasione montana	2,52	abc	2,86	1,07	3,64	*Silene uniflora*	14,73	cd	12,69	16,07	15,42
Linaria vulgaris	2,76	abc	3,55	0,77	7,5	*Sedum sexangulare*	18,85	cd	18,46	26,43	11,67
Sedum sexangulare	4,58	abc	2,66	5	8,75	*Linaria vulgaris*	19,11	d	20,71	20,36	16,25
Aethionema grandiflorum	4,68	abcd	3,21	5	5,83	*Origanum vulgare*	27,64	e	32,86	27,14	22,92
Echium vulgare	4,35	abcd	2,14	2,14	8,75	*Aethionema grandiflorum*	32,58	ef	30	36,07	31,67
Origanum vulgare	5,1	abcd	6,54	6,25	2,5	*Armeria maritima*	36,28	ef	38,08	30,36	40,42
Penstemon pinifolius	6,08	abcd	11,92	4,23	2,08	*Sedum album*	36,22	ef	30,36	37,86	40,45
Sesleria caerulea	5,83	abcd	1,25	5	11,25	*Jasione montana*	40	efg	36,07	38,93	45
Festuca amethystina	7,68	bcde	2,92	6,79	13,33	*Melica ciliata*	40,4	fgh	42,14	38,21	40,83
Sedum kamtschaticum	9,34	cde	10,45	9,64	7,92	*Koeleria glauca*	47,4	ghi	47,14	49,64	45,42
Briza media	11,39	def	8,75	11,25	14,17	*Sedum kamtschaticum*	51,39	hil	46,54	54,29	53,33
Armeria maritima	12,4	efg	5,83	14,29	17,08	*Briza media*	50,58	hil	49,29	47,86	54,58
Campanula rotundifolia	13,5	efg	5,83	18,85	15,83	*Leontodon autumnalis*	50,1	hil	42,5	53,21	54,58
Knautia arvensis	17,51	fgh	11,67	18,93	21,92	*Sedum acre*	53,56	hil	55,71	56,79	48,18
Koeleria glauca	22,36	ghi	7,5	27,5	32,08	*Anthoxanthum odoratum*	54,68	ilm	51,54	52,5	60
Sedum album	21,82	ghil	26,92	18,08	20,45	*Galium verum*	57,04	ilmn	53,21	55	62,92
Galium verum	21,46	ghl	13,85	18,46	32,08	*Veronica spicata*	54,96	ilmn	41,67	58,21	65
Calamintha nepeta	29,96	hilm	14,17	33,21	42,5	*Sedum reflexum*	55,69	ilmn	44,58	62,86	59,62
Festuca glauca	27,03	hilm	15,38	30,71	35	*Thymus pulegioides*	57,36	ilmno	51,54	69,29	51,25
Leontodon autumnalis	23,25	hilm	13,93	25	30,83	*Centranthus ruber var. coccineus*	62,28	lmnop	51,07	65,36	70,42

Prunella vulgaris	25,48	hilm	16,25	23,93	36,25	***Lotus corniculatus***	61,11	lmnop	54,29	68,21	60,83
Veronica spicata	29,49	hilm	12,14	32,08	44,23	***Achillea millefolium***	61,07	lmnop	56,79	66,43	60
Melica ciliata	27,52	hilmn	14,64	32,92	35	***Dianthus deltoides***	64,06	mnop	50,38	66,79	75
Centranthus ruber var. coccineus	31,46	ilmn	24,09	26,54	43,75	***Campanula rotundifolia***	63,77	mnopq	58,57	73,57	59,17
Sedum reflexum	30,37	ilmn	22,73	36,07	32,31	***Penstemon pinifolius***	63,35	mnopqr	67,31	68,57	54,17
Dianthus carthusianorum	33,23	lmno	23,75	39,29	36,67	***Knautia arvensis***	64,22	mnopqr	64,64	65,71	62,31
Sedum acre	35,7	mno	44,58	36,15	26,36	***Calamintha nepeta***	66,69	nopq	53,33	72,14	74,58
Thymus pulegioides	33,89	mno	24,17	43,75	33,75	***Festuca amethystina***	68,55	opqr	66,43	69,64	69,58
Anthoxanthum odoratum	37,98	no	27,08	36,43	50,42	***Dianthus carthusianorum***	71,54	pqrs	57,31	78,57	78,75
Dianthus deltoides	43,79	o	31,67	51,07	48,64	***Prunella vulgaris***	72,85	qrs	74,62	71,43	72,5
Festuca ovina	44,5	o	28,08	50	55,42	***Festuca ovina***	72,49	rs	70,38	75	72,08
Lotus corniculatus	41,11	o	30	47,08	46,25	***Petrorhagia saxifraga***	73,86	rs	65,38	72,86	83,33
Achillea millefolium	53,99	p	50,77	60,36	50,83	***Sesleria caerulea***	79,31	st	74,64	82,86	80,42
Chrysanthemum leucanthemum	59,73	p	52,69	66,92	59,58	***Chrysanthemum leucanthemum***	78,48	st	76,92	78,93	79,58
Petrorhagia saxifraga	63,26	p	45,77	66,92	77,08	***Poa alpina***	82,96	tu	72,5	86,79	89,58
Poa alpina	53,73	p	37,08	62,86	61,25	***Festuca glauca***	88,15	u	83,85	88,93	91,67
Significance	**Taxa**			***					***		
	Depth			***					***		
	Taxa*Depth			***					*		
	$LSD_{0.05}$			**16.89**					**15.57**		

*, *** significant at P< 0.05, and 0.001, respectively. Means distinguished by Duncan's multiple range test (P<0.05) within rows.

4.2.4 CONCLUSION

It can be concluded that as hypotisezed substrate depth has a strong influence on the first phase of plant germination, as illustrated by results from the first weeks after sowing. Deeper substrate conditions promoted seeds germination allowing seeds to germinate faster than the shallower depth. This was except for *Sedum acre* which in the first phase of germination was favoured by 5cm depth. Nevertheless, as has been widely demonstrated deeper substrates are beneficial for both increased water holding capacity and more effective mitigation of extreme temperature fluctuations. Since roof conditions are not so favourable to plant growth, especially for seedlings, a fast and good establishment through deeper substrates could represent a key element for the future success of the green roof.

These preliminary results also show that while germination of some taxa was affected by the variation of the substrate depth, others were not. Moreover, there was significant difference in the response among taxa in percentage of germination in roof conditions, where species like *Talinum calycinum* and *Carex flacca* showed a requirement for considerable more time to germinate than the other species, while species like *Festuca glauca*, *Poa alpina*, *Chrysanthemum leucanthemum*, and *Sesleria caerulea* showed by the end of the experiment high germination rates at any substrate depth.

The results obtained concern a limited period of observation of one month, within which was not possible to complete the observation of the entire germination phase. Hence these results need to be considered as preliminary and require to be completed with further growth and plant survival observations.

4.3 VEGETATION SURVEY ON AN EXTENSIVE GREEN ROOF IN SHEFFIELD

4.3.1 INTRODUCTION

Research on plant survival and adaptation to the green roof environment has been investigated by several authors (Koheler, 2003; Emilsson and Rolf, 2005; Monterusso *et al*., 2005; Durhman et al., 2006; Nagase and Thuring, 2006; Getter and Rowe, 2008; Dunnett *et al*., 2008b; Emilsson, 2008). However, most of these studies are short-term and often focused on survival and growth of commonly used taxa like Sedums. This fact, may provide a misleading or incomplete picture of the long-term survival of the individual plants, and of the dynamics and changes in the vegetation as a whole (Dunnett *et al*., 2008b).

Monitoring plants communities' evolution on roof is important because the vegetation cover composition can significantly alter the green roof's functionality both from an aesthetical and ecological stand point (Oberndorfer *et al*., 2007; Dunnett *et al.*,2008b). From the ecological perspective, the monitoring of plant survival is important as also of the invasion by non-planted species, and the description of the process of vegetation success and change can be valuable to investigate for several reasons. Mainly, this information would help to complete that picture of plants growth and dynamics on green roofs, which is pivotal also from a design perspective.

As Koheler (2003) reported, there are many key factors to which plant development on green roofs are related, such as water storage and capacity of the green roof, type of plant establishment, local climate etc. This list has been extended through the years by other researches, which investigated also on other factors affecting plants development as the substrate composition (Emilsson, 2008) and depth (Dunnett et al., 2008b; Getter and Rowe, 2008).

Substrate depth has been identified as a major limiting factor for plant establishment on green roofs (White and Snodgrass, 2003; Dunnett and Nolan, 2004; Getter and Rowe, 2008; Benvenuti and Bacci, 2010; Thuring et al., 2010). Long-term studies also indicate that substrate depth has a great influence on plant performance, survival and diversity dynamics, which may change significantly compared to the initial planting composition (Dunnett et al., 2008b).

The aim of this study was to investigate the relationship between substrate depth and plant diversity and cover on an extensive vegetated roof.

4.3.2 MATERIALS AND METHODS

Site description and structural planting

The Cube, situated in Sheffield's Cultural Industries Quarter (CIQ), is an excellent example of a sustainably designed building, having at its top (5 storey) an extensive green roof system installed over a 661 m^2 area.

The green roof was planted 2006 on a locally sourced substrate (70% crushed brick to 30% compost). Despite the specifications wanted the material spread evenly across the whole roof to a depth of 70 mm, the contractors, failing, spread it unevenly creating a variation in depth of substrate across the roof. The bricks used were sourced seconds reducing the pressure on landfill through the recycling of aggregates. The compost was made from green waste material.

The leading design idea consisted in the implementation of a biodiversity roof in the form of a wildflower meadow, to create a more natural effect using a short flowering seed mix specially formulated by Dr. Dunnett via Pictorial Meadows and Green Estate. Avoiding the idea of a more traditional sedum mat, the seed mix of drought-tolerant grasses, perennials and alpines selected has ensured a wider diversity, which today supports even bee cultivation.

The plants chosen for this realization are presented in the following table (4.8).

Table 4.8: Plant species sown in the green roof expressed in % of seeds by weight

%	**Taxa**
10	Achillea millefolium
10	Allium schoenoprasum
5	Anthemis tinctoria
10	Dianthus carthusianorum
5	Echium vulgare
5	Galium verum
10	Leontodon hispidus
5	Origanum vulgare
5	Petrorhagia saxifraga
5	Reseda luteola
5	Reseda odorata
1	Sedum acre
2	Sedum album
2	Silene uniflora
5	Thymus vulgaris
5	Verbascum nigrum
10	Pictorial Meadows 'Short Mix'

Vegetation survey and sampling design

The vegetation was surveyed in autumn 2010, when some species were still flowering, according to methods described by Kent and Coker (2000).

The vegetation was recorded using a subdivided quadrat 50x50cm (5x5 subdivisions). Plant cover was estimated visually and two values of the substrate depth were measured for each quadrat. The belt transect approach was applied and in total 4 transects were designed on the roof for sampling: two through the longer side of the roof and the other two perpendicularly to it. Vascular plants, moss, bare ground were recorded. Plant maximum height was measured and recorded on the most species reach transect as measured by the species presence.

Statistical analysis

All data was statistically analyzed using SPSS 16.0 software package for Microsoft Windows Vista. A Bivariate Correlation and regression was performed. Plant cover data collected was transformed appropriately using an arcsine squareroot transformation.

4.3.3 RESULT AND DISCUSSION

Sedum cover was negatively related to depth ($R=-0.625$, $P<0.001$) (Fig.4.5), plants were found to grow at lower depth where the competition and shade from higher and more competitive plants was lower. This result confirm that Sedums are originated from and suited for habitats with shallow substrates, giving better results in this conditions, while do not take any advantage from deeper substrates (Dunnett and Nolan, 2004; Thuring, 2010). Also what has been found by Dunnett *et al*. (2008b) seems to confirm the impression that sedum scarcity at higher substrate depths is due to their low ability to compete with forbs and grasses, which exhibit greater growth in deeper substrates.

Similarly, moss and areas with lack of vegetation were also negatively related to depth ($R=-0.728$, $P<0.001$ and $R=-0.190$, $P<0.05$, respectively) (Fig.4.6). This result indicate that where substrate is shallow and only sedums can grow, when green coverage is not optimal or optimally maintained, alternative forms of non vascular plants develop. Emilsson (2008) in a long-term survey study found that sedums, moss and bare ground cover is dependent on time, and while sedums cover generally decreased, moss development highly increased over time.

Figure 4.5: Cover-depth relationship in *Sedum*.

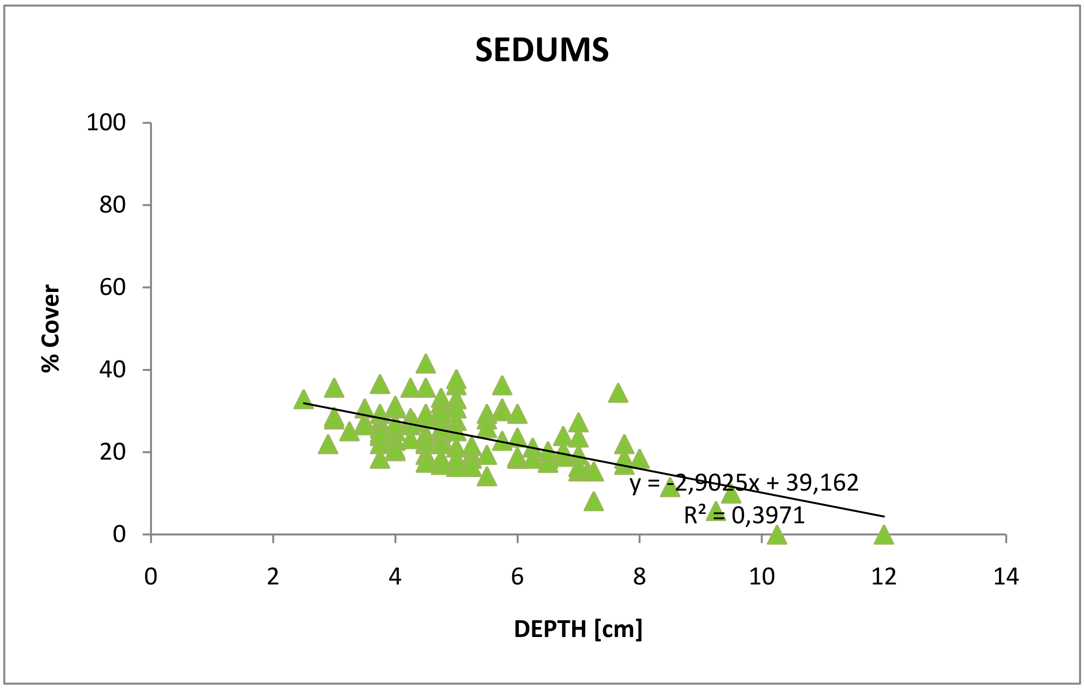
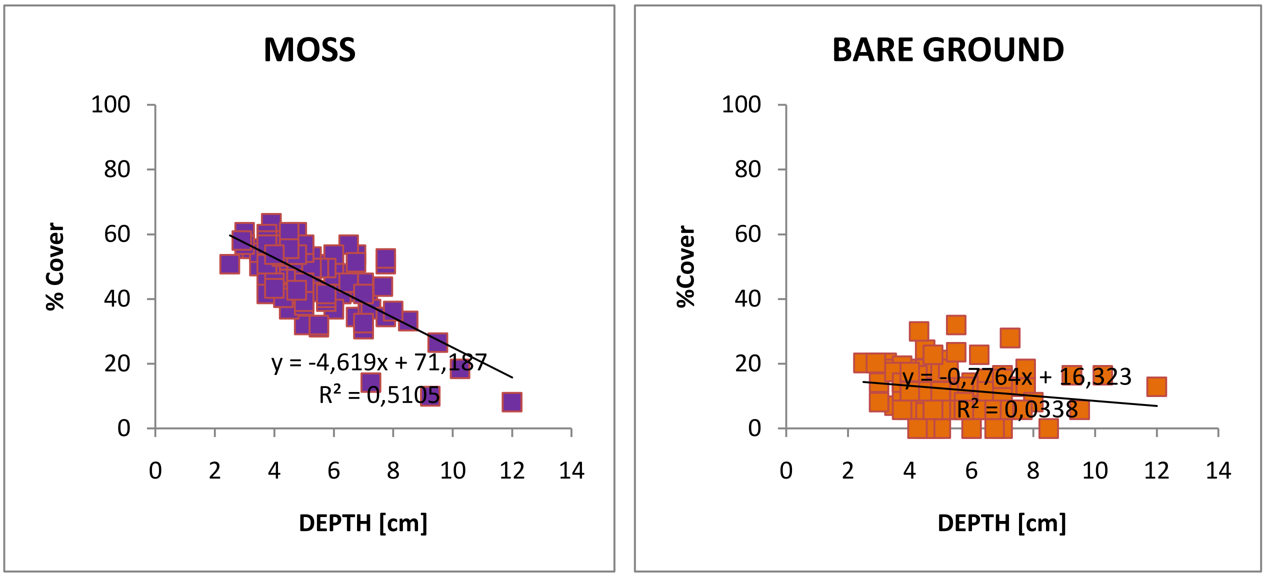


Figure 4.6: Cover-depth relationship in moss and bare ground.



Concerning forbs, cover was significantly and positively affected by depth (R=0.751, P<0.001) (Fig.4.7). In the same way the abundance of grasses was positively related to substrate depth (R=0.625, P<0.001) (Fig.4.8). This result confirms the beneficial effect of deeper soil media on these types of plants and their growth. In this case forbs required more than 4 cm to have a minimum of 20% of coverage, while grasses resulted more exigent needing at least 7 cm.

Figure 4.7: Cover-depth relationship in forbs.

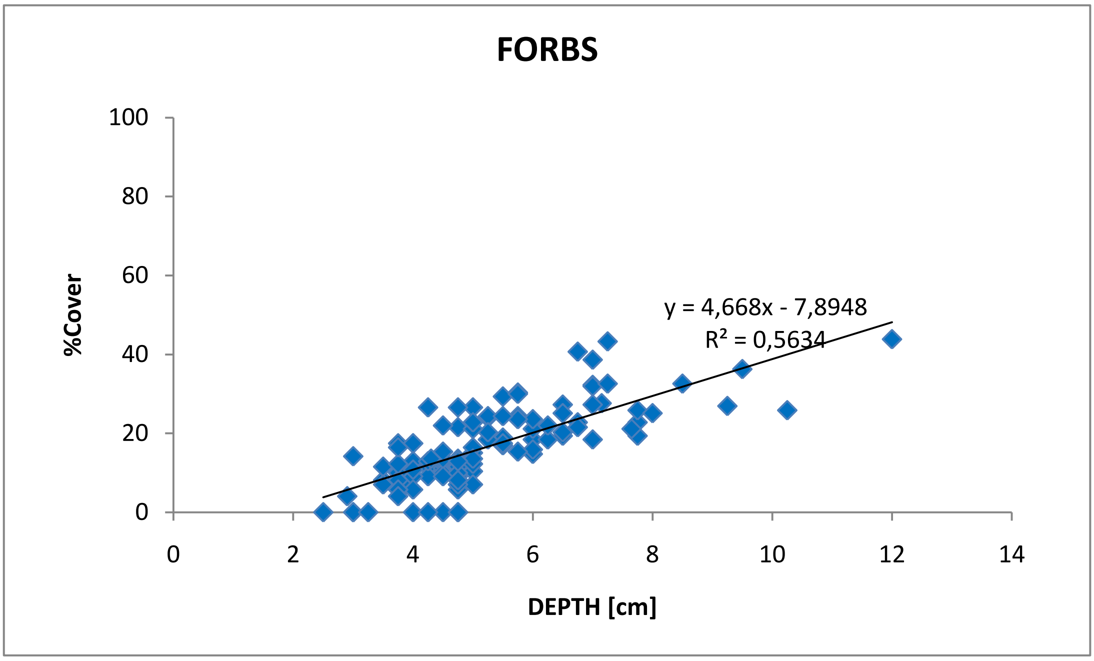
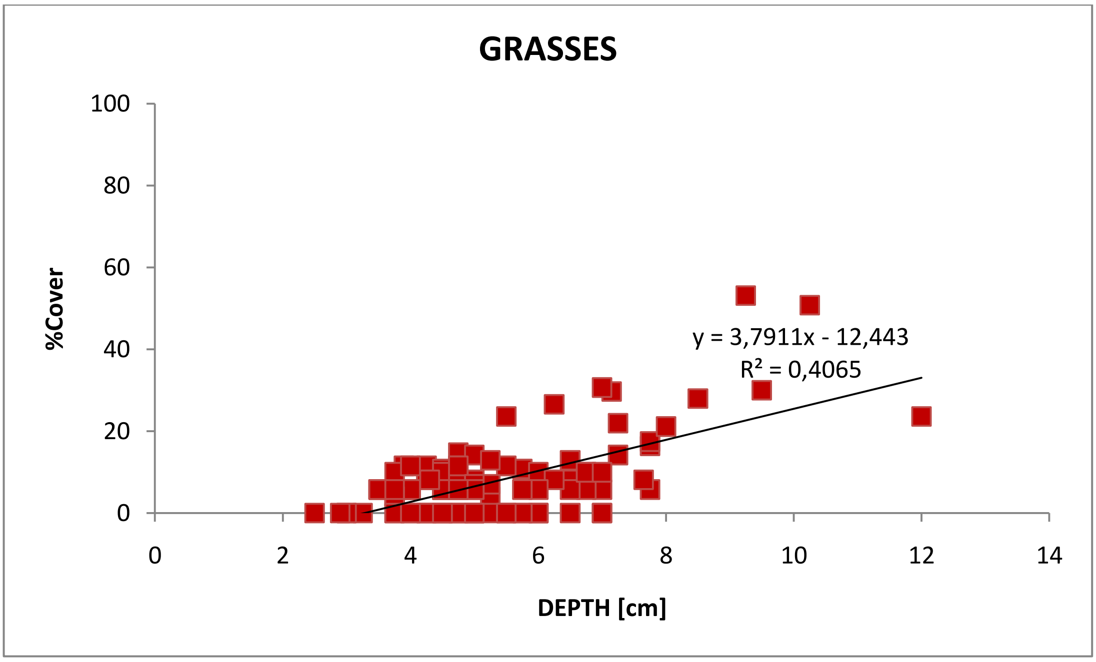


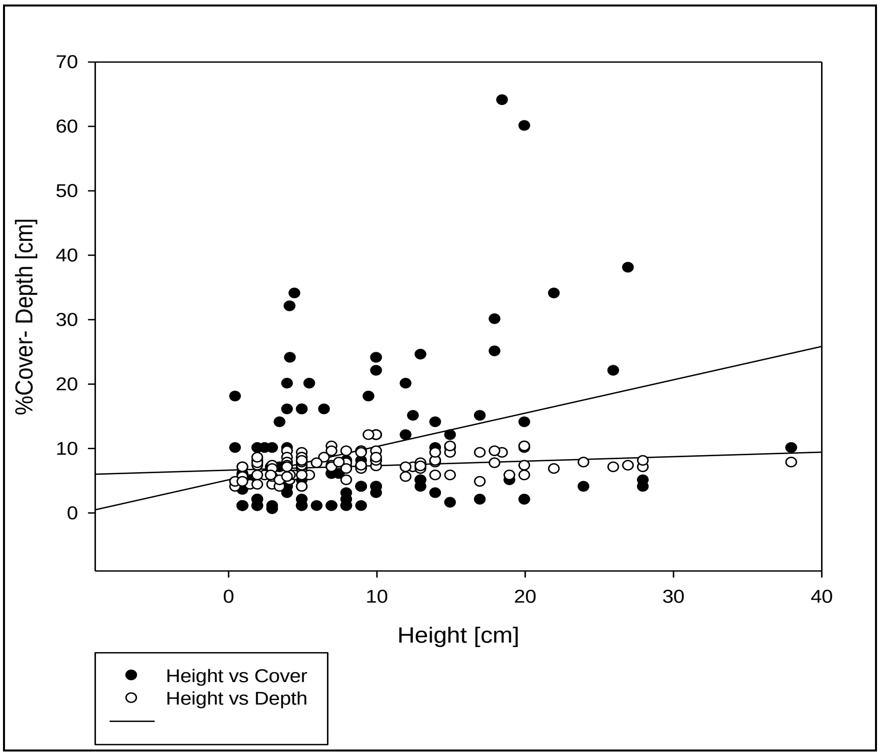
Figure 4.8: Cover-depth relationship in grasses



1.4

When plant cover was correlated over depth and maximum height, a positive relation with both depth (R=0.287, P<0.01) and cover (R=0.359, P<0.001) was found with plant height, while cover and depth resulted no significantly related because of the opposite trend observed in the different plant types object of study (Fig.4.9).

Figure 4.9: Relations between height and cover and height and substrate depth.



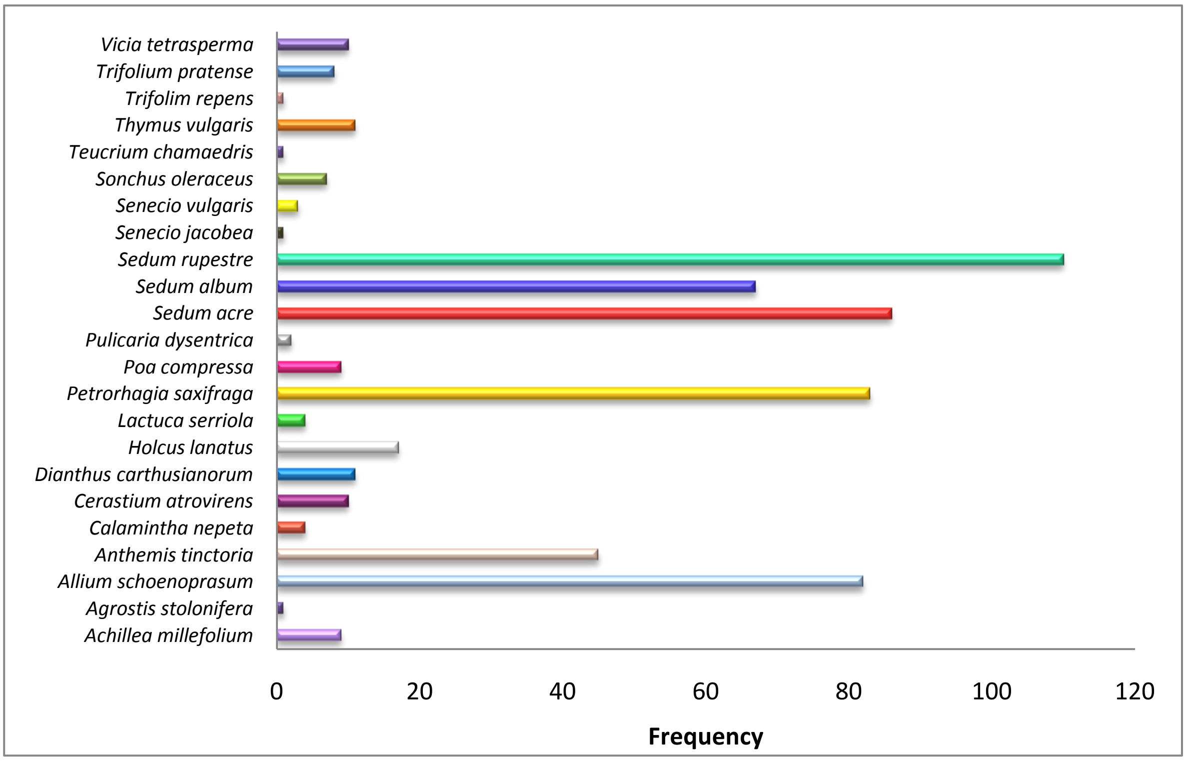
In forbs and grasses, as substrate depth increased, coverage increased (R=0.278, P<0.01), and as cover increased also plant height significantly increased (R=0.489, P<0.001), while to changes in depth did not correspond significant changes in plant height.

While with sedums, the ground cover and plant height resulted positively related (R=0.782, P<0.001), the substrate depth did not significant affect changes in plant coverage and height.

Looking at the plants compositions and presence on the roof it is evident how the most dominant species were sedums (Fig.4.10). Although sedums' percentage of seeds in weight was not high when the roof was planted, after 4 years from the sowing they dominated the roof. Particularly, the most represented *Sedum* species was *S.rupestre*, while concerning the other two species, *S.acre* and *album,* were also abundantly present, but their contribution to cover was minimal. Forbs were more abundant than grasses, and within forbs, especially in lower substrates depth the most represented species were *Petrorhagia saxifraga* and *Allium schoenoprasum*. For grasses, *Poa compressa* and *Holcus lanatus* were the most abundant species.

All these species were part of the mix designed for this roof, except some volunteers that established over time: *Cerastium atrovirens, Lactuca serriola, Pulicaria dysentrica, Senecio Jacobea, Senecio vulgaris, Sonchus oleraceus Trifolium repens Trifolium pratense* and *Vicia tetrasperma,* where these last species along with *Cerastium atrovirens* were the most present.

Figure 4.10 : The relative presence of species found on the roof.



Species as *Echium vulgare, Galium verum, Leontodon hispidus, Reseda luteola, Reseda odorata, Silene uniflora, Verbascum nigrum* were missing. These taxa are new for the green roof planting are since are not reported in any of the list available from literature (Snodgrass and Snodgrass, 2006; Dunnett and Kingsbury, 2008) excepting for *Galium verum* and *Silene uniflora*. However, these roof conditions had not been favourable to their survival or perhaps their establishment.

The persistence and abundance of the above mentioned forbs and grasses species is in agreement with results found by Koheler (2006), which through a long-term evaluation of vegetation dynamics on two extensive green roofs in Berlin, found that *Allium shoenoprasum* and *Poa compressa* were two of the most dominant species found on the sites studied over the entire period of study. Furthermore, as demonstrated in another long-term experiment conducted in UK, plant survival of a mix of dry-tolerant forbs and grasses taxa showed to not decline significantly in deeper substrate depth, while in 10 cm depth most of the species recorded a decline in abundance

(Dunnett *et al.*, 2008a). However, authors observed that this decline was not true for some species which were able to reproduce themselves efficiently through self seeding or agamic propagation. In the examined roof the success of *Petrorhagia saxifraga* and *Allium schoenoprasum* can be probably related to the fact that these species are good self sowing which are also probably less exigent than others in term of substrate depth requirement.

4.3.4 CONCLUSIONS

The results of this brief investigation have highlighted the important effect of substrate depth on plant development and growth on green roofs. A few centimetres of substrate depth can make a large difference on the roof's aesthetic value and benefits. In fact, it was confirmed that shallower substrate depths are more preferable for sedums growth, in these conditions forbs and grasses cannot compete in the same way as in deeper depth where instead sedums are scarcely represented.

Among the non-succulent species *Petrorhagia saxifraga, Allium shoenoprasum*, and *Anthemis tinctoria* were the most abundant. Whereas, because of the roof extensive design having mostly shallow substrate depths, *Sedums* species and moss were dominant. However, it is worth noticing that the above-cited non-sedum species can persist over time also in shallow substrate depth of 4 to 6 cm, thus they strongly confirm to be successful for extensive green roofs.

Results from this study, may suggest further investigations on specific depth-plant species relationships, in order to identify for each species the optimal depth of growth and the lower limit to which plants can survive and persist over time. Especially for extensive green roofs, where substrate depth and severe conditions are limiting and plant choice is very narrow, widening the knowledge of plants dynamics in the long-term, of their cover and abundance variation and the vegetation diversity in relation to time and substrate depth can be useful to improve the green roof planting design.

5 GENERAL CONCLUSIONS

To summarize, it can be concluded that as result of plant selection in Mediterranean climate several drought-tolerant perennial taxa, native and non-native, chosen by aesthetical, eco- and physio-morphological criteria, were found as suitable for green roof planting.

Particularly taxa able to stand to long drought exposure and to survive in roof conditions also through self-seeding capacity were: *Armeria maritima* 'Splendens'*; Cerastium tomentosum; Delosperma cooperi; Sedum spp.; Thymus spp. and Cotynus coggygria*.

In maritime conditions the investigation focused on *Thymus* and *Sedum* led to indentify which taxa are potentially more adaptable to the roof environment and furthermore to provide additional information about the possible influence of plant provenance on plant performance. Results obtained from the study concerning local plants collected from different altitudes suggest an area for further works.

In both climates sedums confirmed to be the best planting option for extensive green roofs. The tested taxa had fast establishment and good growth, and often performed better than other perennial taxa.

Overall in the course of plant selection studies it was found that wild forms were often more adapt to the roof environment than cultivars and that native species are not necessarily more suitable than non-native species to these high demanding environments. However, findings suggest to investigate more this field and to consider both native and non-native species as potential resources.

The additional studies undertaken under maritime climate, germination trial and long-term vegetation survey, pointed out the influence of substrate depth on plant initial growth as well as on long-term vegetation dynamics, where plant germination, growth and diversity are favoured by deeper depths.

Data on surface temperature reductions by different green roofs systems under Mediterranean climate conditions showed that green roofs during summer can decrease the maximum temperature of the roof surface by up to 30°C, while in winter the temperature never goes below zero. Furthermore, vegetation type and presence seems to have an influence on green roofs thermal properties by providing additional benefit.

From a practical point of view, the results obtained in the present thesis can help green roofer in planting design and management, especially in Mediterranean climate conditions.

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