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Research Article

Lichen Transplantation on Polyporales Fungi. An Attempt to Verify the Compatibility after Translocation from a Hill Forest to Anthropized City Parks at Livorno (Tuscany, Italy)

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Abstract

The transplantation of genus *Cladonia* terricolous lichens on Polyporales parasitic xylophagous fungi was attempted. Three species of *Cladonia* (*C. pyxidata*, *C. rangiformis*, *C. foliacea*) have been moved (translocated) from Livorno hill woods (Tuscany, Italy) to some public Livorno parks which suffer by an excessive anthropization. After a short period (a week) of adaptation to the new environment we transplanted thirteen specimens of *Cladonia* lichens on four different species of Polyporales fungi (*Fomitopsis betulina*, *Phellinus torulosus*, *Stereum hirsutum* and *Ganoderma applanatum*) identified on the trees of the parks. The transplants obtained good engraftment and every periodic qualitative control showed a persistent photosynthetic activity even eight months after the start. Only one transplant (on *Ganoderma applanatum*) failed due by the toxicity of a secretion after incision. These observations lead us to consider those experiments as a demonstration of the compatibility of the implied tissues on transplantation and also allow us to introduce the difference of the term transplantation (the insert of an organism to another) from the often-confused translocation (relocation of an organism in different habitat). Future checks will indicate how and for how long this artificial association complex might persist.

Introduction

Some public parks of Livorno (Tuscany, Italy) suffer from excessive anthropization due by social activities, attendance of numerous visitors, a proximity to the urban traffic and sometime by the human incorrect behavior. After we ascertained the presence of epiphytic lichens on the bark and mostly on the trees branches and observed the absence of terricolous lichens in the ground of all the parks (lichen soil desert), we decided to focus two interesting aspects: translocation and then transplantation. Firstly, we identified some terricolous lichens living in abundance on the hills far from the city [1]. We therefore chosen terricolous lichen species *Cladonia pyxidata*, *Cladonia rangiformis* and *Cladonia foliacea* retained useful for their translocation in several points of the ground awaiting their subsequent transplantations on Polyporales fungi just identified in the ecosystem of two city parks. In the specific case of transplantation, we consider the terricolous lichen as “donor” and the Polyporales fungus as “recipient”. Few specimens of Polyporales are growing in the parks where we identified *Fomitopsis betulina*, *Phellinus torulosus*, *Stereum hirsutum* and *Ganoderma applanatum*. So, we attempted the lichen transplantation on each one of that fungi. After the engraftment we systematically planned to check qualitatively the status of the experimental conditions every fifteen days.

The location

The ten public parks studied are located in Livorno (Tuscany, Italy) at an altitude varying between 5 and 35 m a.s.l. Their names are Villa Regina, Villa Fabbriotti, Villa Corridi, Villa Mimbelli, Villa Henderson, Parco Pertini, Parco Masini, Parco Luschi, Bosco dei Cappuccini, Fortezza Nuova.

Material and Methods

In May 2022 we selected and collected small amount of three species of the terricolous lichen: *Cladonia pyxidata* (L.) Hoffm., *Cladonia rangiformis* Hoffm., *Cladonia foliacea* (Huds.) Willd., all growing in the Livorno hill forest at 230 m a.s.l. about 15km from the city. The environment and altitude of the hills is much more different from the urban (5 m a.s.l.) and the periferic (Villa Corridi, 35 m a.s.l.) parks where we translocated the lichens. Other differences regard the soil structure, the temperature and humidity. Considering that all we thought that the lichens can adapt themselves to this ecosystem change for a short period (a week) useful for acclimatization in situ. More other uses may be possible by the translocation such as species conservation, habitat restoration [2] or for biomonitoring pollution [3]. After the acclimatization we decided to proceed with the transplant. All the three terricolous lichen species collected for the transplant differ morphologically each other. The first one *C. pyxidata* is more complex than the others because it contains several fruticose structures forming cup like stalk (Podetia) on the thallus. This different phenotypic character really made difficult the manipulation needed during their use in the transplantations. The *Cladonia* species, determined by microscopy and also chemical spot tests are considered in the case of transplant as “donor”. We also identified and chosen as “recipients” of the transplantation *Fomitopsis betulina* (Bull.) B K Cui. M.L. Han et al., *Ganoderma applanatum* (Pers.) Pat. growing on the bark of *Quercus ilex* (L.) and *Fraxinus ornus* L. both located in the park of Villa Regina. Then we found *Phellinus torulosus* (Pers.) Boirdot & Galzin and *Stereum hirsutum* (Willd.) Pers. respectively on the base of a *Platanum occidentalis* L. and on the severed cut trunk of *Quercus ilex* in Villa Fabbriotti. The transplantation methodology [4,5] began in its first phase with the incision (on average of 4 cm wide and 3 cm deep) in the upper part of the mushroom from the cuticle to the medulla strate without removing any tissue, leaving the space raised to allow the lichen insert (Figure 1). The second phase consisted in manually

introducing and pressing adequate amount of the lichen specimens into the cavity (Figure 2 & 3). Glue has never been used. The next phases were represented by the systematic qualitative macro photographical comparison measurements of the “in itinere” states of the newly formed complex.



Figure 1: Incision type in old specimen of *Fomitopsis betulina*.



Figure 2: Incisions and insert of *Cladonia pyxidata* on *Fomitopsis betulina* juvenile state.



Figure 3: Two incisions and one first insert of *Cladonia rangiformis* on *Fomitopsis betulina* juvenile state specimens.

Result and Discussion

We made eighteen lichen specimens translocations in the parks. Only three of that gave bad results, perhaps due to the overturning with consequent reduction or fragmentation of the lichen mass by the birds present in the environment. Despite this, the other fifteen lichen colonies showed a good vitality even seven months after their translocation. Concerning the transplantation, really more important for the aim of this work, we carried out twelve successful lichen transplantations on Polyporales versus only one failed probably due to bad manipulation. Two weeks after the transplants we noticed an interesting behavior of one “recipient” mushroom *Fomitopsis betulina* which tended to absorb a very small part of the thallus of the “donor” lichen *Cladonia pyxidata*. This fact can be attributed to the rapid growth of the tissues in juvenile status of this mushroom (Figure 4). On the contrary another old *Fomitopsis betulina* specimen maintained well the transplanted lichen without any absorption because its very slow growth was unable to wrap the lichen. All the successful transplanted lichen specimens, including *C. rangiformis* and *C. foliacea*, showed excellent photosynthetic viability when wet naturally or artificially watered until the latest follow-up in December 2022 (Figure 5). Surprisingly *Cladonia pyxidata* transplanted on *Ganoderma applanatum* did not resist to the toxic liquid released by the mushroom cuticle after the incision, thus this transplantation failed. At last all the lichens live quite well eight months after transplantation, however we think that the final results are to be recorded in more than a year. So, at this point, we believe that the ancient Latin Cicerone’s phrase “*Pares cum paribus facillime congregantur*” quite confirm the compatibility between the two different organisms. In fact the “recipient” and the “donor” of transplantation present a common structure (Hyphae). Polyporales fungi and Lichens (commonly defined as a symbiosis between a fungus and an alga) both contain a fungal structure. We believe that the major importance of this preliminary work consists in proposing a new type of lichen transplantation useful both for studying the tissue compatibility of the graft and for observing the reaction of the over time. It remains unlikely to hypothesize the use of this type of transplant to modify an anthropized environment, albeit limited which lacks terricolous lichens. It could however be useful for monitoring pollution near the ground, without neglecting the didactic aspect concerning the uses of the lichens.



Figure 4: *Cladonia pyxidata* six months after the transplantation on *Fomitopsis betulina*. Partial absorption of the lichen thallus by Poliphorales fungus cuticle.



Figure 5: *Cladonia rangiformis* and *Cladonia foliacea* eight months after transplantation on *Phellinus torulosus* at the base of a Plane tree at Villa Fabbricotti.

Conclusion

On the basis of the results obtained, albeit limited to few experimental cases of transplantations, we think that the compatibility between terricolous *Cladonia* lichens and Polyphorales fungi has been confirmed both the permanent photosynthetic viability eight months from the start of the transplant. If these facts go beyond “scientific speculation” or experimental curiosity, however may provide insight into the possible creation of new models of association between fairly similar organisms. We also believe that this transplantation methodology together the translocation could give a help to increase the presence of those important photosynthetic organisms and their thermodynamics [6] in the city park disturbed areas. Finally, once again, we want to mention the ancient Latin phrase of Cicerone in *De Senectute* “*Pares cum paribus facillime congregantur*” (equals with equals very easily associate themselves) which well could be adapt to this work. In fact, the Lichen (symbiosis between fungus and algae) has the component (Hyphae) as well the Polyphorales fungus [7,8].

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