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Alternative cultivation of *Nepenthes*

Hydroponic trials

Development of nutrient solutions for Nepenthes

Alternative growing media

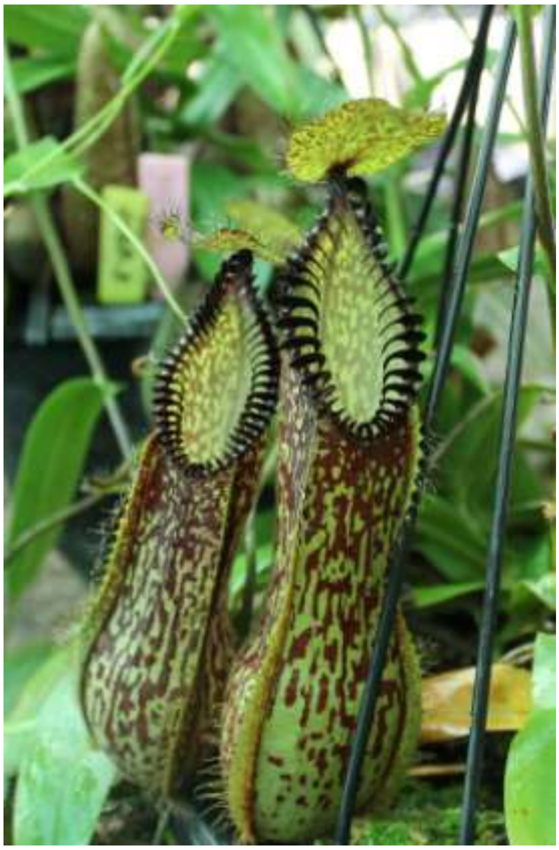
All research conducted at:
Borneo Exotics Highland Nursery
Lindula
Nuwara Eliya
Sri Lanka

Contents

Introduction

Nepenthes are a genus of plant which have adapted to growing in nutrient deficient soils, supplementing the nutrients they uptake through a diminished root system by trapping and digesting insects and small animals. The terrestrial species grow in acidic media such as pure laterite or live *Sphagnum* sp. but often *Nepenthes* adopt an epiphytic habit, receiving a regular flow of water acidified and enriched by humus across a typical mat of moss to which the roots cling. These conditions prove challenging to replication in cultivation, especially in regard to nutrition.

Several species grow serpentine soils found on ultramafic rock and experience concentrations of heavy metals and trace elements that would be toxic to most plants *[1]*. With many species becoming increasingly threatened due to deforestation and poaching, ex-situ conservation is now a very important method of protecting a species. For these reasons this project was proposed in order to research combinations of nutrients that will prove beneficial to the genus. The trails will be carried out at Borneo Exotics highland nursery, Sri Lanka. In this project we will be running two sets of trials simultaneously. The first is using hydroponics for the mass production of commercial hybrids, and the second will be testing alternative growing media for individual threatened species which are difficult to cultivate. We aim to test the effectiveness of different compositions of liquid feed at varying concentrations, applied to a range of growing media through hydroponics. The trials will be carried out over a six month period in order to allow sufficient time to record notable changes in plant growth and development. Through this work we seek a greater understanding of the nutritional requirements of this unusual and fascinating genus.



Nepenthes hamata

Nepenthes robcantleyi

Ex Situ Conservation

The illegal market for trading wild collected plants and seed is still growing, and in recently year's internet access has become readily available in numerous third world countries, such as Indonesia and Malaysia. Many poachers are now finding customers through online auction sites and plant forums. Also lots of local people are coming to realise the value of a rare species and destroying the populations for the huge profits offered by plant collectors. When Borneo Exotics obtains seed of any *Nepenthes* it is immediately sown in-vitro to ensure the species can be multiplied easily in the future. The sudden increase in availability of a rare or highly sought after species quickly reduces the demand on the black market. But with many of these rare species very few people know how to cultivate them successfully. Meaning it can take several years for the multiplication and cultivation techniques to be established. One example is *N. attenboroughii* which was discovered by Stewart McPherson in 2007 in Palawan, Philippines. Since its discovery, the wild populations have suffered severely from excessive seed collection every year, pushing this species to the IUCN status of "critically endangered". Fortunately Borneo Exotics now have this species in tissue culture, although they have had difficulties raising this species we hope that our trials will prove successful and this will contribute to the protection of this species. Another good example of the importance of ex-situ conservation is the late discovery of N. robcantleyi, which was found in 1997 by Robert Cantley on the island of Mindanao, Philippines. The habitat of these plants was being logged at the time of discovery and only a few seed were obtained from one plant. Several years later Borneo Exotics managed to flower two of their six plants producing masses of seed. After the Chelsea flower show in 2010 Robert Cantley showed a sample of the plant to Dr Martin Cheek who immediately recognised it as a new species. It was named *N. robcantleyi* *[2]* in recognition of the work Rob Cantley has done, propagating this new species to produce thousands of plants which could one day repopulation the wild. *N. robcantleyi* is now thought to be either extinct or critically endangered. *[2]*

Borneo Exotics tissue culture facilities

Robert Cantley is the chairman of the IUCN Carnivorous Plants specialist group *[3]*. He endeavours to continue conserving *Nepenthes* species through cultivation

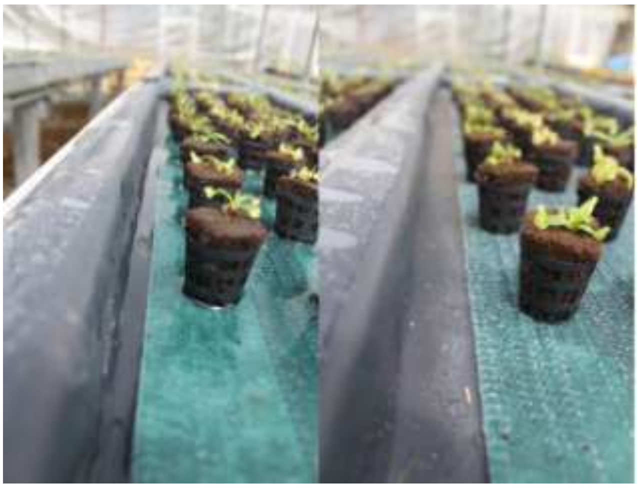
Hydroponic trials

The observed conditions in a typical natural habitat of this genus are often found to be a loose, open substrate with a high saturation of acidified water. These conditions are concurrent with hydroponic methods of growing. There are various methods of applying a nutrient solution to the roots and it would be useful to conduct experiments on these systems in order to ascertain the optimum procedure. However, the aim of this project is to research the nutrient solutions themselves and, following consultation with other experienced growers, we adopted a system proven to work effectively for this genus.
The solutions were tested in 48 hydroponic trays each containing 32-40 plants. The sections were further subdivided into a selection of commercial hybrids, highland species and four species with difficult cultivation requirements. The hydroponic trials we set up use just two different media's. Pure coco coir and peat polymer plugs. While we object the usage of peat on a large scale commercial basis, this is for only used to see that the polymer does not damage the *Nepenthes* in anyway. If this is not an issue a coco peat polymer plugs can be developed for future trials.

Due to the commercial nature of the nursery that provided us with the opportunity to run these trials, we are unable to name products or exact details of nutrient concentrations or combinations. *See Appendix I.*

Method

For the mass production of commercial hybrids we set up trials using a hydroponic system for controlled watering and feeding. The design was based on a system that is known already to effectively maintain suitable hydration for many *Nepenthes* hybrids. The bottoms of the hydroponic trays were lined with capillary matting and the plants were placed in either coco coir or peat plugs, both in small net pots. The nutrient solution was poured into the trays so it covered the capillary matting by approximately 2-3mm. The contact between the growing media and capillary matting meant that the plants could only dry out if the matting did. Our protocol for watering was to top up 1 litre of water per tray once the capillary matting could be pressed without water forming a well around the finger.



After watering *Before watering*

The EC will be measured every fortnight and recorded on a spread sheet. Once the EC drops below the specified level for each trial, the appropriate nutrient solution will be added.

Originally we had not anticipated the number of trials necessary to give a comprehensive understanding of the nutrients *Nepenthes* need. The 16 sections we could use for trials each contained a drainage hole and tap in the right corner. However each section was then separated into three, resulting in 48 trial sections. The drainage system then became void resulting in no means of flushing the system through. Due to the lack of an effective drainage system unused nutrients will build up and these could cause damage if this system is used for long term growth. The principle of this method is simply to allow fast development of commercial hybrids over a period of 6 months. The data from the trails will be recorded in September 2012. This is allows six months for the plants to develop and the trials may be left for a further six months to see when they're quality starts to decline due to nutrient build up. Taking these issues into consideration one must remember that these are only preliminary trials to find what nutrients, and at what strengths these plants respond to best. In that aspect we can expect satisfactory results and the above issues can be resolved for large scale production.

<u>Development of nutrient solution’s for *Nepenthes* Sp.</u>

To determine what concentration of macronutrients are most suitable we set up four trials using a balanced N-P-K at four different conductivity levels (EC). Whichever of these shows the most significant growth will be chosen for our appropriate base macronutrient solution. We then expect to conduct further trials with varying levels of each nutrient meeting the EC level of whichever of the preliminary trials is most successful.

As a control solution we used a conductivity which is below the level where plants may be damaged, but not so low that it would have little impact on the growth. *[Exact EC information withheld]* In their natural environments many *Nepenthes* Sp. experience high levels of micronutrients and trace elements, sometimes at toxic levels. To imitate these we would require a spectroscopic analysis of the soil where several species naturally occur. Instead we included in the control balanced levels of calcium, magnesium, zinc and iron with additional trace elements. This covers all possible requirements and increased levels of individual micronutrients and trace elements will hopefully determine what is most beneficial. For specific issues such as root development we included root stimulators to our trials *See appendix I for complete.*

Method

For the majority of these trials the individual macro and micronutrients were mixed together at balanced ratios and a trace element mix was added to create the control solution. The following micronutrients, Mg, Ca, Fe were systematically increased at three difference strengths, this process was then repeated a further three times with increased levels of trace elements. For the hydroponic trials the nutrient solutions are applied once the EC drops below a specified level, this varies depending on the trial and EC of the initial solution applied. Readings are taken biweekly by removing 2-4 empty peat plugs and squeezing them into a vessel, inserting the EC truncheon and recording the recording the data onto the appropriate spread sheet. The solution is then poured back into the bench to avoid discrepancies in nutrient application. One major flaw in this design is that the empty plugs will contain more nutrients than one containing a plant. Ideally we would remove a plug with a plant and take a reading from that, however on the scale of which we conducted these trials it would not be possible as that method would damage the root system and impair the accuracy of our results.

To ensure no mistakes were made in the nutrient applications while we were away from the nursery all solutions had to be prepared in advanced. As we could not estimate the rate of nutrient uptake, concentrated solutions had to be prepared for monthly application, six syringes for each trial. These was tested individually to ensure once diluted the EC levels matched those on our records. This should hopefully bypass any human error which could occur from the staff at the nursery. Certain trials which required enzyme feed, root stimulators and vitamin supplements had to be prepared in separate syringes to prevent any unexpected chemical reactions.

Pre-prepared nutrients

Alternative growing media

The Borneo Exotics nursery use standard media for all of their *Nepenthes* Sp. and hybrids. For adult plants the mix consists of 1:1 coco coir and coco chips, while for young plants they use pure coco coir. The coconut palms where this material originates are grown inland and to avoid salt concentrations which would be detrimental to Nepenthes. While this mix provides good drainage and works for most plants it is not ideal for ultramafic species such as *N. villosa*. To establish the best media and nutrient combinations for these species I contacted Dr Charles Clarke who directed me to an article on inorganic growing media *[4]*, suggesting that it was perhaps aeration that the species needed. Following this advice several trials were set up testing different media with varying nutrient applications (S*ee appendix II for full details - 49-58 and 69.)*

Due to limited availability of materials in Sri Lanka the media components were carried over by hand, therefore we were unable to set these trials up on a larger scale. If the plants show signs of improvement in growth and development then further trials will be set up. The information that we gathered stated that ultramafic or serpentine soils are derived from igneous and meta-igneous base rock *[5]*. We can concluded that pumice and volcanic rock will have similar qualities. In addition to this crushed volcanic rock can be added to increase trace elements and aid microbial activity in the soil; but this is another important factor to consider in future projects.

One of the issues Borneo Exotics asked us to resolve was the problem of *N. lowii, N. villosa, N. macrophylla* and *N. x trusmadiensis* not developing root systems in the nurseries. This issue we believe was due to the high humidity inhibiting the rate of transpiration and thus decreasing the need for root development. As this could not be changed we ran three trials *(66-68 Appendix II)* using course sand and open mixes to allow aeration to the roots. Trial No. 66 is using the coco coir as a control, the three trials will be examined after the six month period and the length and root mass will be compared for improvement.

The trials set up to understand the requirements of ultramafic species were of a specific interest to us. These soils naturally contain magnesium, iron and trace metals at levels which are often toxic to many species *[1]*. We managed to set up several trials testing different nutrient concentrations in a media similar in texture and consistency as those found on Mt. Kinabalu, Borneo.

Nepenthes rajah growing in inorganinc media

Method

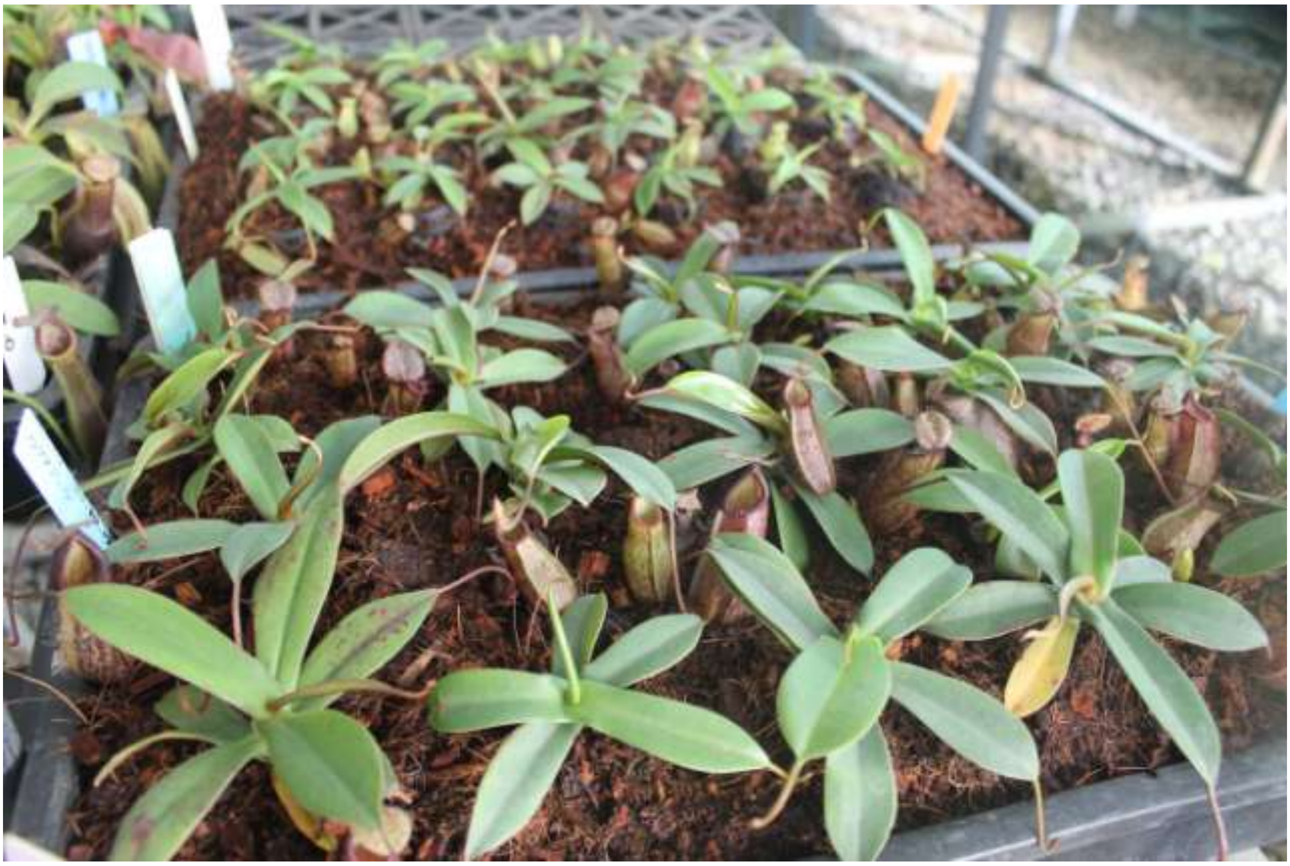
Conducting these trials was a little more complicated than the hydroponics. The inorganic media for example was very open and free draining and therefore required to be left in a tray of water. This was lined with capillary matting and a small drainage whole added at 2cm from the bottom to prevent over filing. The rest of the trials were potted into their specified media and placed in a tray of coco coir to prevent them drying out too quickly. When solutions are applied the coir is also watered to prevent nutrients diffusing into the plane media.

Methods of recording data

One issue which has been noted by Borneo Exotics is over fertilisation, which often causes sale problems. Not, like one may expect, in the case of root burning and damage to the plant. Instead the sudden increase in growth, resulting in a decreased laminar thickness, pitcher size and both leaf and pitcher colouration. The purpose of developing this fertilisation technique is to find a way of balancing all the desired characteristics of the plant by recording correlation between them with the correct nutrients. This could mean that multiple feeds of different nutrients are required to give the desired result. E.g. increased EC of macronutrients achieves maximum leaf development while a decreased level of macronutrients and an increase level of trace elements and Iron for example, may increase pitcher size and colouration. Ideally a perfect combination of all of these will be used. It could beneficial to isolate these separately to increase growth first and then develop the desirable characteristics, providing the increased growth is not at the expense of leaf quality.

Measurements of pitcher height and colouration, leaf thickness and colouration and plant diameters would be an effective method of comparison. However due to shortages of time and natural variability between species and seed grown hybrids a monthly photographic record will be kept as the main source of data. While not a very scientifically valid method, photographing the plants and judging improvements by eye will allow us to find sufficient evidence to see if it is more effective than Borneo Exotics current fertilisation regime.

The most effective method to see how successful the trials are would be to measure wet and dry mass. Unfortunately this is not an option as many of the plants we are using are both critically endangered and extremely valuable. While many species within this genus grow epiphytically with diminished root system we do intend to remove the media where possible and take photographs and measurements. This will depend on the speed of deterioration of the peat polymer plugs.



Conclusion

The conclusion of this will be completed in September 2012. We are unable to observe any increase in the plant development as many *Nepenthes* in cultivation have a slow rate of growth. I remain in regular correspondence with Robert Cantley and the managers who helped with this project. Both of whom send updates on how the trials are going and photos of their observations.

I am writing a full analysis of how each trial performed against their appropriate controls, and counter controls. E.g. trial 7 with increased Iron will be compared against control 3, trial 8 with a second increase of Iron can then be compared against trials 7 to see if higher levels of Iron are detrimental to the plants or if it actually augments growth in any way. Due to the commercial nature of this business the information cannot be published within this public report. Any scientific institution that wishes to see this paper must contact me privately.

Completed hydroponic trials 1-48

Acknowledgments

This project was only made possible from the benevolent funding supplied by the Merlin Trust, which I am eternally grateful for. Thank you to my colleague Joe McGahan for all his help and support throughout the project, Robert Cantley and Diana Williams for their supervision, assistance and for the opportunity to work at their beautiful nursery. Nick Johnson at Kew, for his help, guidance, and excellent letter of recommendation. Dr Charles Clarke for his help and advice, directing us to some very useful articles and publications, and all the staff at Borneo Exotics nursery for their assistance in setting up and running these trials.

References

Publications

2. Dr Martin Cheek. (2011). Nepenthes robcantleyi sp. nov. (Nepenthaceae) from Mindanao, Philippines. Nordic Journal of Botany. 29 (1), 1.

Websites

1. Sabah Parks. (2010). Kinabalu vegetation profile. Available: http://www.sabahparks.org.my/eng/kinabalu_park/vegetation_profiles.asp. Last accessed 27 May 2012

3. IUCN. (2012). Available: http://www.iucn.org/about/work/programmes/species/who_we_are/ssc_specialist_groups_and_red_list_authorities_directory/plants/. Last accessed 27 April 2012.

4. Heiko Rischer. (2000). Growing Nepenthes in a Completely Inorganic Substrate. Available: http://www.carnivorousplants.org/cpn/samples/Cult292NepInorSub.htm. Last accessed 27 April 2012.

5. F. Q. Brearley. (2005). NUTRIENT LIMITATION IN A MALAYSIAN ULTRAMAFIC SOIL. Available: http://info.frim.gov.my/cfdocs/infocenter/jtfsonline/jtfs/v17n4/596-609.pdf. Last accessed 26 April 2012.

Appendix I

Nutrient Solutions

Macronutrients:

(N) Nitrogen : (P) Phosphorus : (K) Potassium

TE = Trace Elements

- 1TE = minimum dosage (1ml/L)
- 2TE = maximum dosage (2ml/L)
- 3TE = exceeded dosage (3ml/L)

Fe = Iron

- 1Fe = minimum dosage (0.75ml/L)
- 2Fe = medium dosage (1ml/L)
- 3Fe = maximum dosage (2ml/L)
- 4Fe = exceeded dosage (3ml/L)

MgO = Magnesium

- 1Mg = minimum dosage (1ml P/L)
- 2Mg = maximum dosage (2ml P/L)
- 3Mg = exceeded dosage (3ml P/L)

Ca = Calcium

- 1Ca = minimum dosage (1ml P/L)
- 2Ca = maximum dosage (2ml P/L)
- 3Ca = exceeded dosage (3ml P/L)

Ready mixed solutions

- Liquid Hydroponic Orchid feed
- Powder hydroponic orchid feed
- Powdered seaweed feed
- Standard commercial plant feed

Additives:

Micro-mix – Balanced MgO, Ca, Fe
Root stimulator
Vitamin supplement
Trichoderma strains
Natural enzymes
Coffee

Hydroponic Trials

1. Pure water = Control 1 (C1)
2. Osmocote pellets 10:11:18 +MgO +TE = Control 2 (C2)
3. N:P:K medium EC + micro-mix + 1TE = Control 3 (C3)
4. N:P:K low EC + micro-mix + 1TE
5. N:P:K intermediate EC + micro-mix + 1TE
6. N:P:K high EC + micro-mix + 1TE
7. Control 3 + 1FE
8. Control 3 + 2FE
9. Control 3 + 1MgO
10. Control 3 + 2MgO
11. Control 3 + 1Ca
12. Control 3 + 2Ca
13. Control 3 + 1FE +1TE
14. Control 3 + 2FE +1TE
15. Control 3 + 1MgO +1TE
16. Control 3 + 2MgO +1TE
17. Control 3 + 1Ca +1TE
18. Control 3 + 2Ca +1TE
19. Control 3 + 1FE +2TE
20. Control 3 + 2FE +2TE
21. Control 3 + 1MgO +2TE
22. Control 3 + 2MgO +2TE
23. Control 3 + 1Ca +2TE
24. Control 3 + 2Ca +2TE
25. Control 3 + Root stimulator Single dosage
26. Control 3 + Root stimulator applied Bi-monthly
27. Control 3 + Vitamin supplement
28. Control 3 + Vitamin supplement
29. Control 3 + Natural enzymes Single dosage
30. Control 3 + Natural enzymes applied Bi-monthly
31. Control 3 + Trichoderma strains - Applied once on first feed
32. Standard commercial plant feed – low EC
33. Standard commercial plant feed – medium EC
34. Standard commercial plant feed – high EC
35. Liquid Hydroponic Orchid feed - low EC
36. Liquid Hydroponic Orchid feed - high EC
37. Powder hydroponic orchid feed - low EC
38. Powder hydroponic orchid feed - high EC
39. Seaweed feed
40. Osmocote low EC
41. Osmocote medium EC +1FE +1MgO
42. Osmocote high EC
43. Coffee
44. Control 3 + Coffee
45. Control 3
46. Control 3
47. Control 3

48. Control 3

Media trials

49. Control 2
50. Control 3
51. Seaweed feed +micro-mix
52. Control 2
53. Control 2
54. Control 2
55. Seaweed feed
56. Seaweed feed +micro-mix
57. Control 3
58. Control 3 +micro-mix +TE
59. Control 2
60. Control 2 + 1Ca
61. Control 2 + vitamin supplement
62. Control 2 + natural enzymes
63. Seaweed feed
64. Seaweed feed +1Ca
65. Seaweed feed +1Ca +1TE
66. Control 3 + root stimulator
67. Control 3
68. Control 3
69. Control 3

BRAND NAMES NOT LISTED

Appendix II

GROWING MEDIA

1-44. Peat Polymer Plugs

1-44. Coco coir

45. Peat plugs with tray

46. Chopped foam, paddy husk and coco chips (1:1:1)
47. Growth technology peat plugs
48. Foam plugs
49. Burnt Earth: Padi Husks: Seramis, 1:1:1
50. Burnt Earth: Padi Husks: Seramis: Hydroleca: Volcanic Rock Chips, 1:1:1:1:1
51. Burnt Earth: Padi Husks: Seramis: Hydroleca: Volcanic Rock Chips: Rock Dust, 1:1:1:1:1:1 (Rock dust applied at 5g/L)
52. Burnt Earth: Padi Husks, 1:1
53. Burnt Earth: Padi Husks: Rock dust 1:1 (Rock dust applied at 5g/L)
54. Burnt Earth: Padi Husks: Hydroleca: Seramis: Rock dust, 1:1:1:1 (Rock dust applied at 5g/L)
55. Burnt Earth: Padi Husks: Seramis: Rock dust 1:1:1 (Rock dust applied at 5g/L)
56. Burnt Earth: Padi Husks: Seramis: Rock dust 1:1:1 (Rock dust applied at 5g/L)
57. Burnt Earth: Padi Husks: Seramis: Rock dust 1:1:1 (Rock dust applied at 5g/L)
58. Burnt Earth: Padi Husks: Seramis: Rock dust 1:1:1 (Rock dust applied at 5g/L)
59. Coco coir
60. Coco coir
61. Coco coir
62. Coco coir
63. Coco coir
64. Coco coir
65. Coco coir
66. Coco coir
67. Burnt Earth: Padi Husks: River sand 1:1:1
68. Burnt Earth: Padi Husks: Seramis: River sand 1:1:1:2
69. Hydroleca: Volcanic Rock Chips: Seramis 1:1:1