The Rufford Foundation
Final Report

Congratulations on the completion of your project that was supported by The Rufford Foundation.

We ask all grant recipients to complete a Final Report Form that helps us to gauge the success of our grant giving. The Final Report must be sent in **word format** and not PDF format or any other format. We understand that projects often do not follow the predicted course but knowledge of your experiences is valuable to us and others who may be undertaking similar work. Please be as honest as you can in answering the questions – remember that negative experiences are just as valuable as positive ones if they help others to learn from them.

Please complete the form in English and be as clear and concise as you can. Please note that the information may be edited for clarity. We will ask for further information if required. If you have any other materials produced by the project, particularly a few relevant photographs please send these to us separately.

Please submit your final report to jane@rufford.org.

Thank you for your help.

Josh Cole, Grants Director

Grant Recipient Details	
Your name	Magdalene Namondo Ngeve
Project title	Local Gene Flow Dispersal, and Establishment of Mangroves in Cameroon: Combined Revelations from Morpho-Physical Observations
RSG reference	**17335-1**
Reporting period	May 2015 – May 2016
Amount of grant	£5000
Your email address	ngevem@yahoo.com
Date of this report	April 18th 2016

1. Please indicate the level of achievement of the project's original objectives and include any relevant comments on factors affecting this.

Objective	Not achieved	Partially achieved	Fully achieved	Comments
Assessment of the fate of mangrove propagules after abscission			√	Please refer to comment 1 and comment 4
Investigation of the optimal ambient (physical) conditions for mangrove establishment			√	Please refer to comment 2 and comment 4
Investigation of local gene flow through capture-mark-release-flow experiments			√	Please refer to comment 3
Investigation of gene flow through the use of molecular (microsatellite) markers.			√	Please refer to comment 3

Comment 1: For this objective we investigated the fate of propagules post abscission from their mother trees. This was done considering 5 probable environmental conditions that a propagule could deal with when it is released from its parent tree in the wild: (1) Fresh water, (2) Half saline water (salinity of about 17 ppt), (3) Saline water (salinity of 35 ppt), (4) Dry and seldom inundated soil (salinity: 0.002ppt), (5) Mud (salinity: 0.002ppt), and (6) Sand (salinity: 0.002ppt). Each experimental condition was in a separate plastic bath. We monitored temperature and air humidity (as well as light) every day. Propagule growth was measured every 10 days. This allowed us to fully understand the viability of propagules under variable environmental condition and how they tend to respond under different constraints. Propagules in Mud started growing first, but those in the sand grew longer roots overall, indicating that irrespective of an early unset of rooting in the mud, both sediment types trigger rooting very well. Nevertheless, delayed dormancy was observed for propagules in fresh and half-saline water (where no sediment was present), where propagules only started rooting after the 4th and 5th weeks, and earlier in the fresh water, than in the half-saline water, where growth was much faster and longer ones started. Currently propagules in the saline water still have not developed any roots. It is interesting to note that this high salinity experiment resulted in the highest propagule mortality, especially for treatments in the light (direct sun light). These results indicate that propagules that remain afloat for too long, without finding suitable sediment to strand, could begin rooting after a period of dormancy if they are in the catchments where salinity levels are not low since rooting is highly

reduced in plain sea water. Root bumps and a few roots also were observed in a few of the propagules in the treatment of dry soil, indicating high resistance to drought for *Rhizophora racemosa* propagules. Data analyses on these are still being carried out for this study, but a look at Fig 1 below summarises the preliminary results.

Comment 2: From our experiments on the fate of propagules post abscission, our data indicated that mud and low saline conditions (fresh water) are the strongest root triggers in propagules. Therefore, to investigate the optimal growth and establishment conditions of propagules we had six experimental setups, we tested 3 experimental conditions: (1) Mud in fresh water conditions, (2) Mud in half-saline conditions and (3) Mud in saline conditions in Light (mimic of high light availability in mangrove forest edges). A second treatment of all three experimental conditions above was done in the shade (mimic of limited light due to mangrove dense canopy cover), to investigate propagule growths interactions of salinity and light (and thus a temperature gradient). This was done considering two major Light-varying conditions in order to investigate the role of Light availability at the forest edges of mangroves versus the shade of the dense canopy on mangrove seedling growth and establishment. Statistical tests are still on-going but our preliminary results show that propagules in the shade compensate for the absence of light by remaining ever-green, to make use of the little light that filters through, for optimal photosynthesis. Propagules in light generally turned brownish after 1 - 2 weeks even when still viable. After 3 weeks, the numbers of rooting propagules (as well as the mean length of the longest root) in the light treatments were generally significantly higher than their shade counterparts, for all 3 test conditions. Also the mean number of roots per propagules was the same for both shade and light Freshwater mud treatments but increased in shade half saline and saline mud conditions than Light counterparts. Indicating an initial assignment of more energy to increase root numbers in the shady saline environments than light freshwater environments, at the early rooting weeks. After 6 weeks, the mean number of roots per propagules was highest in the light-fresh water mud treatment, and almost equal in the other treatment. Average root length after six weeks was highest in light-freshwater mud and light-half saline mud treatments and lowest in the saline mud conditions of both shade and light. The Light-Freshwater mud treatment was the first and only to have propagules which were leafing after 6 weeks. The overall growth after 15 weeks decreased with increasing salinity for the light treatments and propagules in the light generally grew better than those in the shade (except for those in sea water mud (max salinity of 34 ppts) where those in the shade grew better than those in the Light). This indicates a strong interaction between high salinities and limited light of dense mangrove forest on seedling growth. This implies propagules that get to strand in the dense mangrove canopy experience reduced growth, but will surely survive if other stressors, such as high salinity, are absent. In the presence of high saline conditions, propagules in the shade (under dense canopy) as well as those in the light (forest edges) are both restricted to an equal extent. Therefore the major determinant of propagule growth is salinity and moisture rather than light availability (although in comparing sites with low salinities, those in the light grow better than those in the canopy shade).

Comment 3: We harvested 600 matured propagules, marked (painted) and released them to observe seed dispersal direction and landscape connectivity for the highly fragmented mangroves of the Wouri River. The insight on the dispersal dynamics of both seeds and pollen (see comment 4 below) is very crucial for the conservation of these populations. Capture-release experiments indicate dispersal is bidirectional along the Wouri River (Fig. 2), mediated by the tides in the upstream direction and by the river currents in the downstream direction. We collected leaf material from 650 individuals (2 leaves per tree) and in another case study we collected 30 propagules and 2 leaves per tree from 10 trees in a plot (3 plots were sampled). From these samples we obtained high quality DNA in the laboratory, which with molecular procedures we were able to assess genetic diversity and gene flow and found that patterns are in congruence with the capture release experiments of propagules. The manuscript written from these data has been completed and submitted. We found that for this estuary, mangroves of the protected area (Douala-Edea Reserve) have the lowest genetic diversity. This highlights the impact of historical pressures on "recently" protected areas, and calls for the need to increase the number of protected areas within this area. Interesting, we also found that the smallest and most vulnerable populations near the wouri bridged have the highest genetic diversity. Since genetic diversity has a positive correlation with (theoretical) resilience, these populations need to be protected and the use their propagules in different afforestation schemes should be considered so as to conserve their unique genetic heritage. These findings were also presented in an oral presentation at an international conference (see below) and have been recently submitted to *Hydrobiologia* journal for publication. In the second case study of propagules and leaves of mother trees to assess cross pollination and (gene flow via pollen transfer), data analyses are being carried out.

Comment 4: This was normally not one of our objectives, but while carrying out our work, we had the opportunity to asses "uncommon" threats to mangrove seedling survival and growth in the wild for the mangrove populations of the Wouri River. Focus group discussions and field observations revealed that mangrove populations of the Wouri River are plagued with "uncommon" threats, in addition to the widely reported (illegal) wood extraction and pollution. These threats include: intensive sand extraction, removal of the mangrove sediments (from mangrove forest floor) for creation of anti-erosion barriers and land reclamation, and invasive species (*Nypa fruticans* which reduces available habitat and water hyacinth: *Eichhornia crassipes*, which traps and retain propagules in their mats and prevent them from locating potential suitable habitats to strand). The extent to which these different threats affect these mangroves shall be assessed.

2. Please explain any unforeseen difficulties that arose during the project and how these were tackled (if relevant).

Increase in the prices of fuel was the major challenge encountered but this was taken care of by supplementary funding received from our institution. Also unexpected rains and primate predation of our propagules disturbed the early stages of some of our growth experimental set ups. So we adapted to deal with this and carried out our experiments in more controlled environments.

3. Briefly describe the three most important outcomes of your project.

i. Unveiling genetic diversity and local gene flow (by pollen and by seed dispersal) among these *Rhizophora racemosa* populations of the Cameroon Estuary complex has been achieved. This will then allow the managers of these ecosystems to consider this very important concept to conservation, in future conservation efforts and schemes.
ii. Seed dispersal along the Wouri River is bidirectional, mediated in the downstream direction by the river currents and in the upstream direction by tides. The overall gene flow along this estuary is very high such that they form one genetic cluster, likely due to the strong hydrodynamic patterns of this estuary. This high rate of gene flow implies that these populations are not experiencing any drift and thus remain a self-sustaining forest ecosystems (and have the potential to recover) should the current challenges be removed, with no human intervention on tree planting.
iii. A propagule's fate post abscission from the mother tree is determined by that of the type of environment it finds itself. Although these propagules have large reserved of water and nutrients, survival chances greatly decline with aridity and with increasing salinity. Immediate growth occurs for propagules in moist sediment (both sand and mud), while delayed dormancy occurs for floating propagules in fresh and half saline media. Propagule mortality is extremely high for those floating for long periods in open sea water. This information is also relevant to managers of these ecosystems in the development of nurseries for these species and subsequent reintroduction of nursed seedlings in the wild, where care should be takes on local conditions to enable optimal survival of reintroduced seedlings.
iv. Salinity rather than light availability determines propagule survival and establishment, where humidity is optimal. Nevertheless limited light availability is restrictive on early growth of seedlings. Bearing this in mind, managers can strongly advocate against the canalization and damming of rivers that feed mangrove creeks and catchments. This so because with sea level rise and the subsequent saline intrusion into catchments, these low salt-tolerant species are highly at risk.
v. Highlighting the existence of the "uncommon" threats to mangroves seedling survival, in addition to the knowledge obtained from physical growth conditions, is very crucial in strategizing for the management of the mangrove forest of the Wouri particularly (and everywhere such threats do occur). Should policy makers focus on alleviating sand extraction pressure and mangrove sediment removal (as these greatly reduces available substrate for the stranding of propagules), recovery of these systems will be quicker.

4. Briefly describe the involvement of local communities and how they have benefitted from the project (if relevant).

Local communities helped to keep intruders away from the experimental setups and others volunteered (to join the paid field assistants) to helping in the retrieval of propagules, while swimming for leisure. Local communities also participated in focused group discussion and some interviews to give us preliminary insights on the ecosystem and the (current) human-induced threats they face.

5. Are there any plans to continue this work?

Yes we do have plans to continue the work. Our field work revealed that community perception towards mangrove ecosystems and their conservation very negative. Also, we received several intriguing questions from the local communities and some school children about mangroves. This calls for dire need to increase capacity building and create awareness, through campaigns, training workshops and seminars. This will enhance the involvement of these stakeholders in participatory management. Together with local NGO's, we plan to carry out a thorough assessment of local ecological perception, valuation and knowledge of mangrove ecosystems, and how this can be improved through education and training and how all of these can be used in formulating better conservation scheme for these mangroves. We also intend to create awareness in primary and secondary schools, to educate the coming generations on mangrove ecosystems and other ecosystems. We also have plans to incorporate other mangrove areas along the coast of Cameroon; this will facilitate regional integration of the different mangrove areas in management efforts. Therefore we have plans to apply for a 2nd Rufford Foundation Small Grant to carry out our project on the education of local communities and students on mangrove ecosystem conservation.

6. How do you plan to share the results of your work with others?

Some findings were presented in an oral presentation at the *European Conference of Tropical Ecology: "Tropical diversity, ecology and land use,* which held on 23rd of February 2016 at the University Göttingen, Germany; organized by the Society for Tropical Ecology. A poster was presented showing some of the results at the PhD day (5th July 2015) of the Doctoral School of Natural Sciences and (Bioscience) Engineering at the VUB. There are plans to share more of the results in other workshops and seminars with NGO's in Cameroon and other scientific forum. One manuscript has been submitted (in *Hydrobiologia*) and it the acknowledgement section, the Rufford Foundation was recognised as the funding body. Statistical analyses are still being carried out and more manuscripts are in preparation for submission and subsequent publication in scientific journals, and they all will bear the name of the Rufford Foundation as the funding body of the research.

7. Timescale: Over what period was The Rufford Foundation grant used? How does this compare to the anticipated or actual length of the project?

The Rufford grant was used for the entire period of the 12 months project. Funds were received in May 2015 and the project commenced immediately.

8. Budget: Please provide a breakdown of budgeted versus actual expenditure and the reasons for any differences. All figures should be in £ sterling, indicating the local exchange rate used.

Item	**Budgeted Amount (£)**	**Actual Amount (£)**	**Difference (£)**	**Comments**
15 Large plastic baths/containers, one for each setup and replicates and controls and water drums	400	300	100	The excess £100 was used for other items that did cost more than initially budgeted for (see below)
Permanent Fluorescent markers to mark propagules to study dispersal (water-insoluble Biodegradable paints were used instead, which are more costly but safer for the environment), paint brushes, paint dyes, paint dissolvent	150	350	200	
Fuel and hiring of speedboats and canoes for navigation within the mangroves to harvest propagules, regular checks of experimental set ups, and harvesting of propagules and sampling for plant material for genetic data	1500	2000	500	This was much higher than expected because of two reasons (1) increased cost of fuel during the period of the research and (2) the intensive search that was needed for propagule recapture after they had been dispersed over wide distance
Transport to different mangrove sites and payment of local assistants	1500	1000	500	
4. Buying of needed equipment's such as refractometer, Leaf-Area Index meter, GPS, tape, digital Camera with geo-tagging, rain-boots for research team members, notebooks, stationary, envelops for storing plant tissue for genetic material	1000	1150	150	
Food and accommodation	250	250	0	
Miscellaneous	200	200	0	
Total	5000	5450	450	

Note: The additional £450 was covered by our institution.

9. Looking ahead, what do you feel are the important next steps?

The next steps would be to educate local communities and other stakeholders through campaigns, training workshops and seminars and to thoroughly assess local ecological perceptions and how these can be integrated into improved conservation schemes. It is also of great relevance for our work to be extended to the three other mangrove areas that span the entire coast of Cameroon. This is because although the Cameroon Estuary (the current study area of this project) is the most disturbed mangrove area, there is a need for regional mangrove protection, because only 7.1% of mangroves are protected in Cameroon, and there is genetic connectivity between some mangrove areas. There is dire need to create nature education programs in the near future to be aired in local and national TV and radio stations. And we have some thoughts which we hope when developed will allow for this.

10. Did you use The Rufford Foundation logo in any materials produced in relation to this project? Did the RSGF receive any publicity during the course of your work?

Yes, the Rufford Foundation logo was used on every page of the oral and on all the poster presentations of our findings. The Rufford logo was inserted as the funding body of the project in all presentations. The Rufford Foundation was also acknowledged in the submitted manuscript (and will be included in subsequent ones under preparation) as the funding body of the project.

11. Any other comments?

I would like to express my sincere gratitude to the Rufford Foundation (UK) for funding this project and allowing me to gain great insights on the field situation of mangroves of the Cameroon Estuary. The combination of both morpho-physiological observations and genetic/or genotypic diversity is very crucial for the long term management of these mangroves; and the Rufford grant has enabled this through funding this project. It has been a very rewarding experience and very great opportunity and privilege to have been funded by the Rufford Foundation.

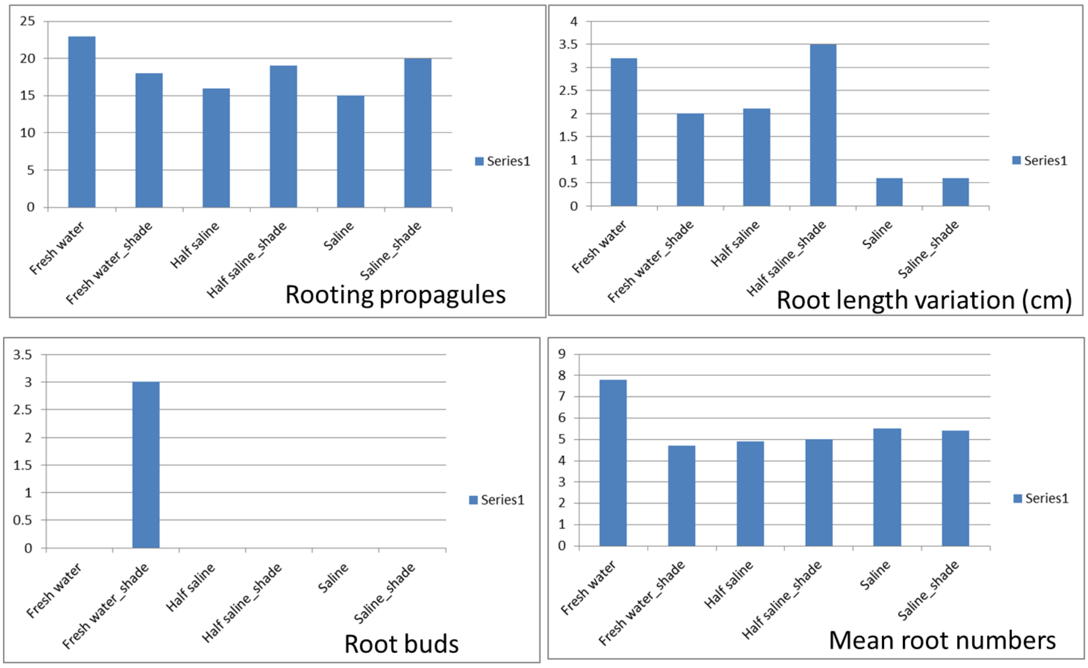

Fig 1. Preliminary results or growth experiments (after 6 weeks in different growth treatment conditions).

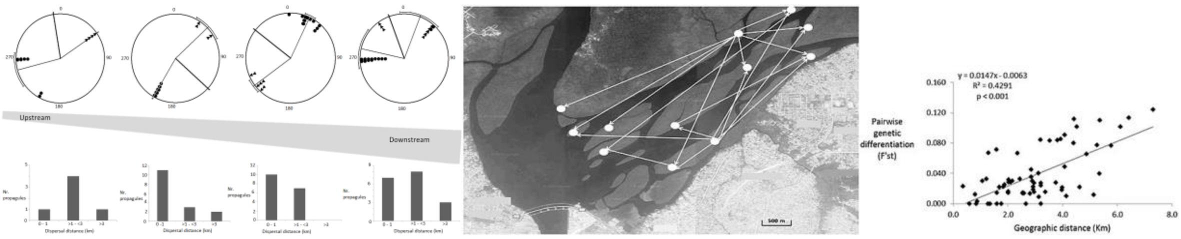

Fig 2. Summary of propagule dispersal directions and distances, contemporary migration rates, and dispersal model for mangroves of the linear landscape of the Wouri

Fig 3. Removal of mangrove sediment severely destabilizes sediments and this will hamper stranding of propagules (A). Heap of compact mangrove sediment used for anti-erosion barriers and for land reclamation into the Wouri River. (B, C). Sand extraction (D) is also a major threat on these mangroves. This could go a long way in altering the natural hydrodynamics of the river and will affect mangroves

Fig. 4. Measuring propagule growth with a young enthusiast who is fascinated about mangroves and nature science (A). Growth experiments, examining new roots and root bumps after the first week (B). Success in our growth experiments evidenced from long roots in optimal conditions (C).

Fig 5. Painted propagules for ready for the capture-release dispersal experiments (A). On the voyage of searching for dispersing propagules over the Wouri River (B). Two of the propagule search team members pose for a picture during a rest/lunch break on a small island on the Wouri River (C).

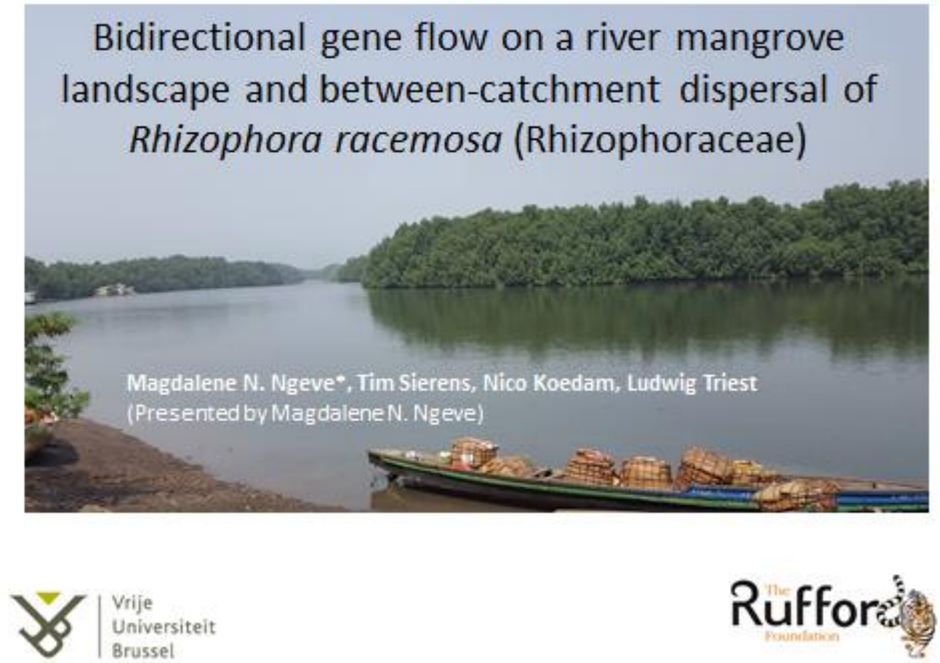

Fig 6. Tittle slide of the presentation of first results at the European Conference of Tropical Ecology (February 22nd 2016).

Acknowledgments

Funding

Vrije Universiteit Brussel

The Rufford Foundation

Fig 7. Closing slide of the presentation at the European Conference of Tropical Ecology (February 22nd 2016).

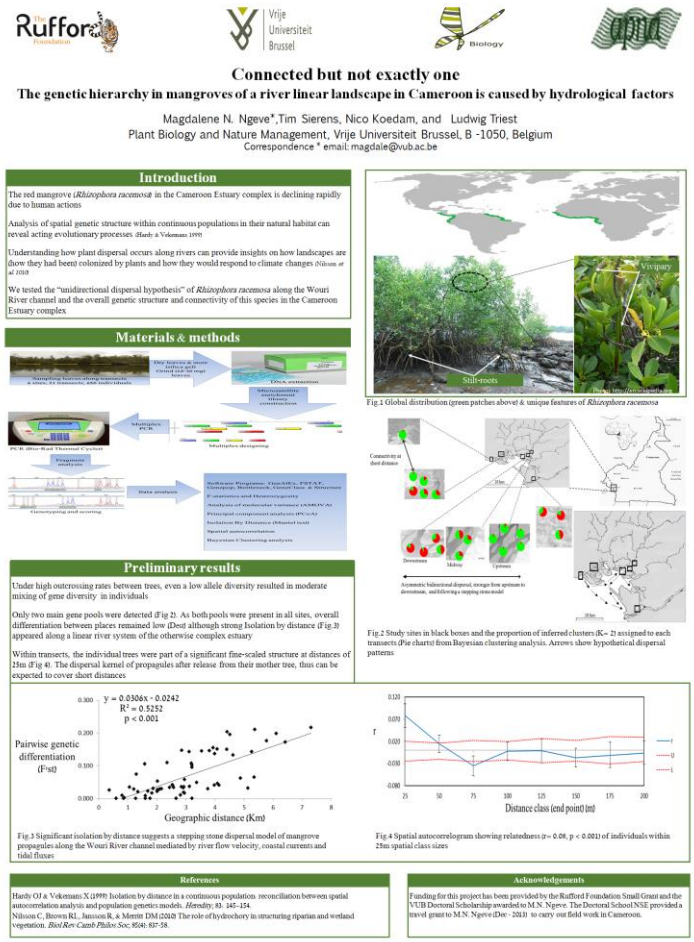

Fig 8. Preliminary results on gene flow presented on a poster during the PhD Day of the Doctoral School of Natural Sciences and (Bioscience) Engineering at the Vrije Universiteit Brussel.