Postdoc Fellowships for non-EU researchers

Final Report

Name	Md. Qumruzzaman Chowdhury
Selection	2012
Host institution	Wood Biology Service, Royal Museum for Central Africa (RMC A), Tervuren,
Supervisor	Dr. Hans Beeckman
Period covered by this report	from 03/09/2013 to 28/2/2015
Title	Long-term growth dynamics in mangrove forests of Bangladesh under climate change

1. Objectives of the Fellowship (1/2 page)

The overall objectives of the fellowship were to provide opportunities for research on the selected topic on Bangladesh mangroves and to improve the research potential of the fellow to pursue through further training in to exchange ideas and information about the latest advances in wood biology.

The selected topic focused on the Sundarbans, the largest single tract mangrove forest in the world situated in the south-western border of Bangladesh. It has enormous significance in coastal protection, biodiversity conservation and livelihood supports to the local communities. Since 1893, the forest has been managed under consecutive periodic (i.e., every 10-year) management plans which have been prepared from the inventory data and setting up exploitable girth/diameter guessing their ages from traditional girth measurements. Apart from anthropogenic disturbances, depletion of the stock has resulted from over harvesting due to over estimation of growth. Mangrove ecosystems are threatened globally due to the adverse effect of climate change. Time series based on dated growth rings offer the opportunity for age and growth rate estimation, fixing rotation and annual allowable cut, and to explore climate-growth relationships for paleo-climate reconstruction as well as for the prediction of tree growth under future climate change. However, the general assumption on indistinct growth rings in mangrove species, mostly hinders the potential of dendrochronological applications. Therefore, we aimed i) to elucidate the ring structure and growth periodicity integrating cambial marking experiments and cambium activity analysis in *Heritiera fomes*, the flagship species of Bangladesh mangroves ii) to investigate the dendroclimatological potentials and iii) to understand wood anatomical traits variations (intra and inter- specific) in different environmental conditions.

2. Methodology in a nutshell (1/2/ page)

2.1 Cambial marking experiment Sample collection (objective 1)

Cambial zone of twenty *H. fomes* trees in four study sites (five in each location) were marked on 6-8th January 2013, using a hypodermic needle (18G; 1.2 mm diameter) at 130 cm above ground level. A total of 13 stem discs were collected from three sites on 30th March and 1st April 2014. Fresh samples were collected from another fifteen trees from three sites and immediately preserved in container with 1:1 alcohol and glycerin. A total of 40 stem discs from eight different locations was collected at breast height. All discs were sanded using a sanding machine with gradually increasing grit from 150 to 1200. Transversal sections of xylem and cambial samples were papered and images were taken with a microscope, equipped with camera system. Moreover, images were taken using polarized light optics as well as epi-fluorescence with mercury arc lamp and an Olympus WU filter cube.

2.2. Sampling for ring analysis (objective 2)

A total of 40 stem discs from eight different locations were collected at breast height. On the sanded discs, tree-ring boundaries were marked with pencil under a stereomicroscope on three radii from pith to bark to check for ring anomalies. The anomalous ring number (s) was (were) archived to facilitate corrections during subsequent tree-ring analysis. After marking, all discs were scanned at 2400 dpi and the ring widths were measured using Fiji ImageJ software. Crossdating was performed among the radii of each disc. The influence of local climate (temperature and precipitation) on tree growth was analyzed using DendroClim 2002 over the common time span from 1948 to 2011.

2.3. Sampling for traits analysis (objective 3)

A total of 210 wood cores were collected from three species from sixty three salinity zones. Microtome slides were prepared and image analysis was carried out using a software (see objective 1 & 2).

2.4. Site variables analysis

Composite soil samples were collected from each sampling location (objective 1-3) from a depth of 15 cm, they were kept in polythene bags, we analyzed the electrical conductivity (EC, in a solution of 1:5 soil-water mixtures using a conductivity meter, Extech 341350A-P Oyster) and converted this to salinity (ECe). Inundation classes were designated as I, II, III and IV where inundated by 100–76%, 75–51%, 50–26%, 25–5% of the high tides, respectively.

3. Results (6-8 pages)

3.1 Tree ring characters in *Heritiera fomes*

Wound observation

The cambial mark appeared as moderate reaction in wood due to wounding (Fig. 1). The inserted needle produced a puncture canal (up to 3 mm in length) in the xylem which is characterized by a combination of wood anatomical features, such as crushed cells, an amorphous zone and oxidized xylem. The dark layer was formed by the residues of crushed cells which are usually referred to as the 'stripes of cell wall residues'. The amorphous substances accumulated between the crushed cells and the oxidized wood.

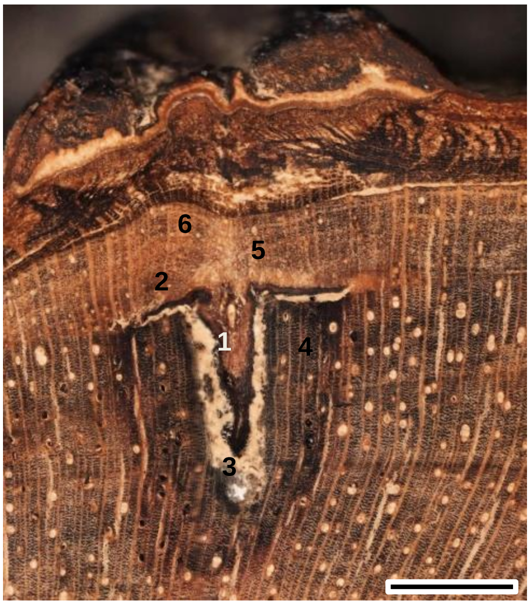

Fig. 1 Microphotograph of the cambial wounding; 1, puncture canal; 2, crushed cambial derivatives 3, amorphous layer; 4, oxidized wood; 5, parenchyma tissue; 6, restored xylem. Scale bar = 2 mm.

The oxidized wood produced around the puncture canal up to 1.5 mm distance after the amorphous zone. Above the layer of crushed cells, callus-like parenchymatous tissue indicates the actual wound response of the tree, after which wood production was restored. The amorphous substances accumulated between the crushed cells and the oxidized wood. Above the layer of crushed cells, a large area of parenchymatous tissue indicates the actual wound response of the tree, after which wood production was restored. The newly formed wood produced a two-fold higher vessel density and significantly (*t*-test, $p<0.01$) smaller size vessels after wounding. The number of the grouped vessels and ray density was higher in the post-wounding zone. Moreover, most of the vessels in the oxidized wood were filled with gummy substances.

Tree ring structure and periodicity

Of the 13 samples collected from the cambial marking experiment, one growth ring was found in each tree since in January 2013 (Fig. 1A). The ring is composed with a band of marginal parenchyma which is predominantly one cell wide but occasionally up to three and mixed with fibers (Figs. 1 & 2). Newly formed resorted wood was not found before the ring formation after wounding (Figs. 1 & 2), and it can therefore be assumed that wood formation is ceased in January. Of the 13 marked trees, 38% trees developed a complete concentric growth ring after the cambium marking.

Fig. 2 Sanded disc of a pinned tree, the arrow indicates a ring formed after cambial marking (A). Anatomical characters of a ring (B). Scale bars, A = 10 mm and B= 500 µm.

The width of an individual ring is not constant along the circumference. Growth ring anomalies, i.e., wedging and partially missing rings were also found which is related to the reduced growth rate and the asymmetrical stem form. In most of the cases, the opposite part of tension wood had low radial increment (<0.75 mm) and therefore current year ring in that area merged with the previous one and produced wedging or sometime (partially) missing ring.

Cambial activity

In this study, the term cambial zone applies to the entire region of tissue generation and includes their intermediate derivatives between the xylem and phloem. The demarcation boundary between xylem and cambium zone was abrupt. The cambial zone was characterized by 4-7 cambial cells as well as thin tangential and thicker radial walls (Fig. 3). Moreover, there was no enlarging or differentiating cambial derivative. There was no site-specific variation in cambial zone characteristics. These criteria indicated the presence of the dormant cambium during the sampling period.

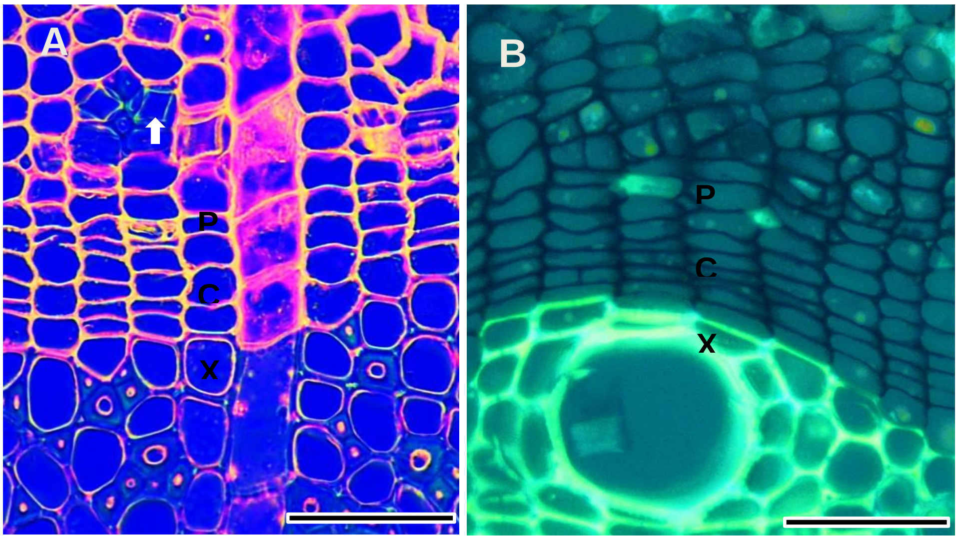

Fig. 3 Morphology of dormant cambium. A, transverse section of light micrograph; B florescence light. X, xylem; C, cambial zone; P, phloem. Arrow indicates crystal. Scale bar = 50 µm.

Phloem characteristics

The conductive and non-conductive phloem was characterized based on anatomical characteristics considering the conductive potential of the sieve tube elements by Esau (1969). The non-conducting phloem is the part of phloem where the sieve elements are no longer functional and distinguished by complete or partially collapse of sieve elements (obliteration), ray dilation, presence of sclereids and crystals. The collapsed sieve tubes were found at some distance from the cambium zone (Fig. 4A). The sclereids were thicker walled, lignified and isodiametric or slightly extended cells with variable shape. The ray cells were enlarged (dilatation) in the phloem toward the periderm (Fig. 4B). The prismatic and rhomboidal shape crystals were abundant and occurred in the axial parenchyma solitary as well as chambered in the phloem (Figs. 4C & D).

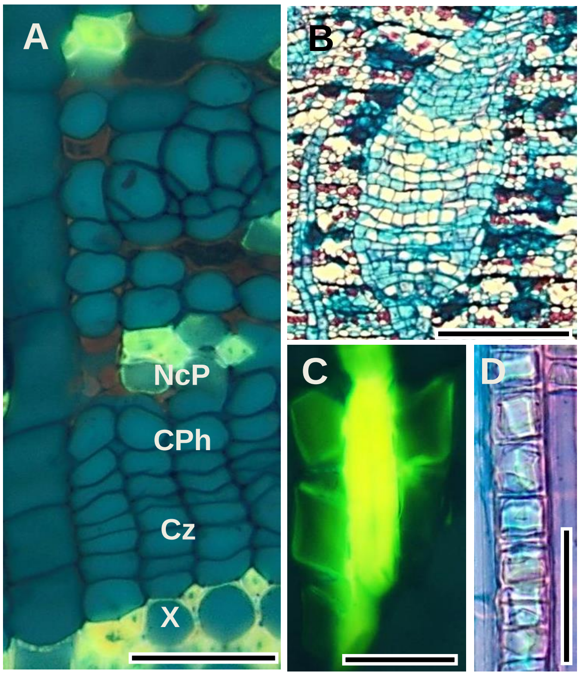

Fig. 4 Morphology of the phloem. A, transverse section under florescence light; B, widening ray; C, crystals under florescence light in radial section and D; tangential section with chambered crystals. Xy, xylem; Cz, cambial zone; Ph, phloem. Scale bar (for A-C) = 50 µm; and for D is 500 µm.

Most of the phloem was characterized by non-functional features (Esau 1969), except narrow part close to the cambium where the sieve elements were intact with the companion cells (Fig. 4A).

The data presented here regarding to the first objective, using cambial marking, as well as cambial dormancy offer strong evidence for the presence of annual growth rings in *H. fomes* and showing their potential for analysis of tree age, carbon sequestration and reconstructions of forest growth dynamics. *H. fomes* is still the most important species of the Bangladesh mangroves and spreading over 67% of the vegetated area. From the last century due to increase of human disturbances, and reducing the allowable harvesting diameter in the successive management plans might has decreased large diameter trees in the forest. However, there are still trees more than 50 cm diameter available with nearly 200 years life-span, which might offer the opportunity to construct long chronologies.

3.2. Dendroclimatogical potential of *Heritiera fomes*

Chronology development

All discs showed distinct rings and marking tree-ring boundaries was possible from all sites (Fig. 5A). Partially missing (ring width drops in some part of the disc) and wedging rings were found in most of the samples. False rings and double rings were also detected and corrected by following the whole circumference of the disc. After correction, all three radii were averaged to one tree-ring series for each tree. The ring width significantly varied between the sites and the eastern site showed higher value (1.22±0.16 mm) than the western site (0.96±0.18 mm). However, ring width variation within the site was not significant in both cases. Average tree-ring width was 1.13±0.15 mm.

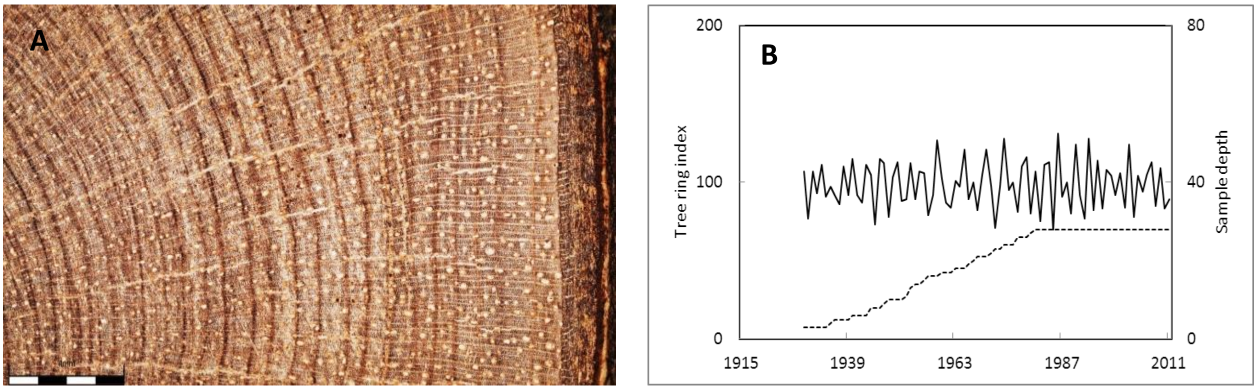

Fig. 5 Transversal view of a sanded disc showing tree-ring boundaries (A). Scale bar =4 mm. Standardized regional chronology (B).

Cross-dating was not successful for all trees in both sites. Based on the selection criteria (*GLK* ≥ 60 and $t \geq 2.0$), we excluded 7 trees from the eastern part and 3 trees in the western part. Tree-ring series displaying poor correlation with the master series were also removed and correlation between two site chronologies was high (0.64, p<0.001, n=82).Finally, a regional chronology was constructed using 28 trees from both sites with a length of 82 years (Fig. 5B).

Climate-growth analysis

In general, we found positive correlations between tree growth and precipitation from the corresponding meteorological station for both zones (data not shown). Increased precipitation in May to September and November enhanced local tree growth in the eastern site.

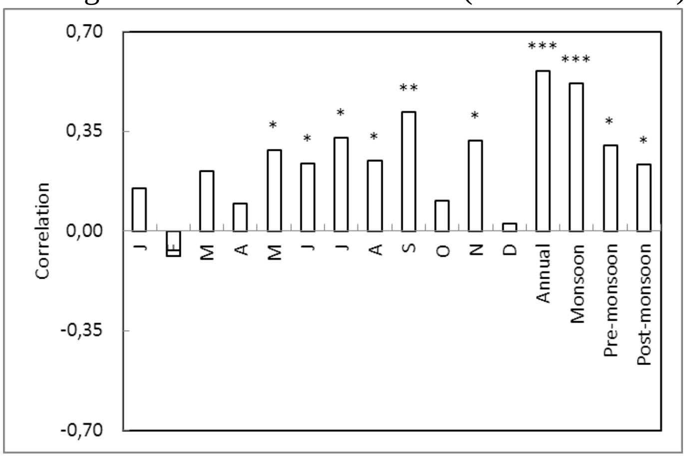

Fig. 6 Correlations between regional chronology the monthly, seasonally and annual precipitation. Significance levels *p<0.05, **p<0.01, ***p<0.001.

Trees in the western site showed similar relationships except the precipitation of May and August. On the regional scale, precipitation of May to September and November showed positive correlation (Fig. 6).

In case of precipitation, annual precipitation yielded the highest correlations (east, r = 0.51; west, r = 0.60; regional, r = 0.56, p<0.01) with the three chronologies followed by monsoon (east 0.47, west 0.60 regional 0.52, p<0.01) and pre-monsoon (east, r = 0.30; west, r = 0.28; regional, r =0.30, p<0.05) precipitation (Fig. 5). Post-monsoon precipitation was slightly correlated only with regional chronology (r = 0.23, p<0.05). The monthly temperature and relative humidity did not show significant correlations with any chronology.

We found significant correlations for regional precipitation with gridded sea surface temperature (SST) for over the Pacific from September to December and Indian Ocean from August to November (Fig. 6). The SST of both oceans during this time period or other periods of the year was not directly correlated with the regional chronology.

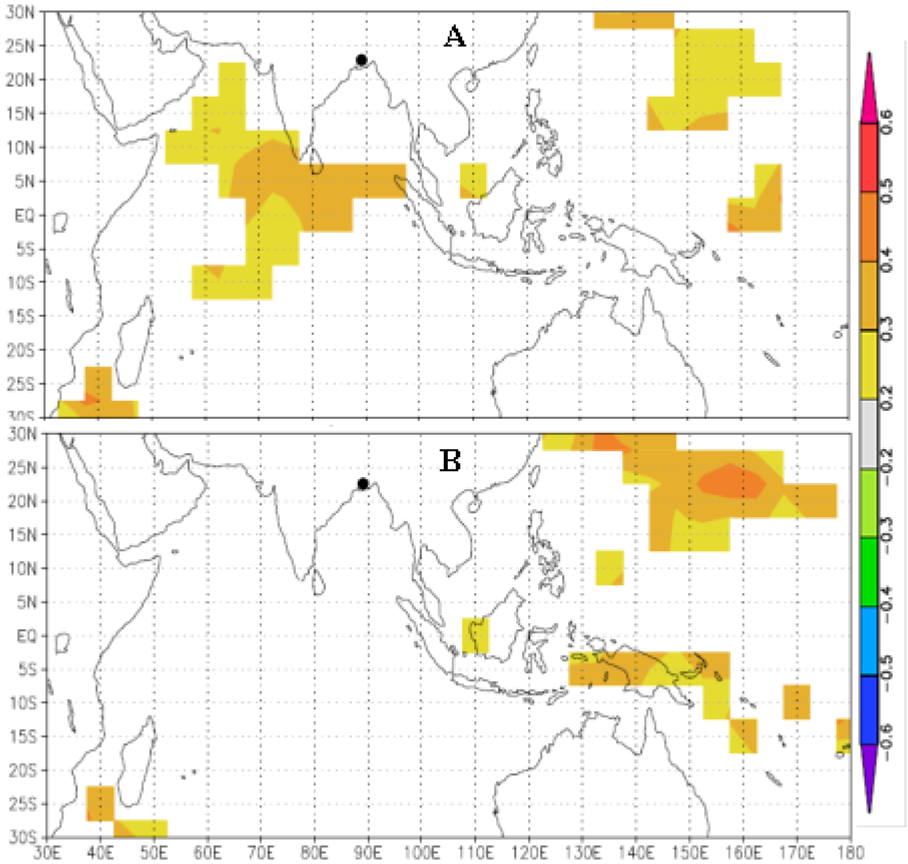

Fig. 7 The correlation map (1948–2011) between the gridded sea surface temperature (SST) and precipitation during August–November (A) and September–December (B).

At a global scale, precipitation of May to July and October to December was negatively influenced by ENSO (Fig. 7). However, tree growth was not influenced by this force, even during this time period. Moreover, tree growth in strong El Niño and La Niña years did not differ significantly from the corresponding ‘normal’ years.

For the first time, we present the dendroclimatological potential for *H. fomes* from Bangladesh mangroves. Its tree rings are well-defined and cross-datable which lead to develop regional chronology. Tree growth is mainly influenced by local climate and suggests the potentials of development of longer chronologies for paleo-climatic data reconstruction in Bangladesh. Moreover, longer ring-width series with a good site replication will improve our understanding of the climate/growth association of this species and extension of tree-ring network within its natural range.

3.3. Hydraulic traits variation in three dominant species in the Sundarbans

Despite mangroves have been extensively studied, little is known about their ecological wood anatomy. Vessel architecture is an important source of information about how trees are coping with the varying environmental conditions such as salinity to optimize the hydraulic conductivity and safety. We surveyed of three important mangrove species from three different salinity zones such as low (<30 dsm^{-1}; ECe), medium (30-54 dsm^{-1}; ECe) and high (>54 dsm^{-1}; ECe) from the Sundarbans. For this study, wood cores of 105 trees of *Excoecaria agallocha*, 65 of *H. fomes* and 48 of *Xylocarpus mekongensis* were collected from sixty three permanent sample plots (PSP) covering from three salinity zones. From each PSP at least one wood sample was collected from the available species at breast height level.

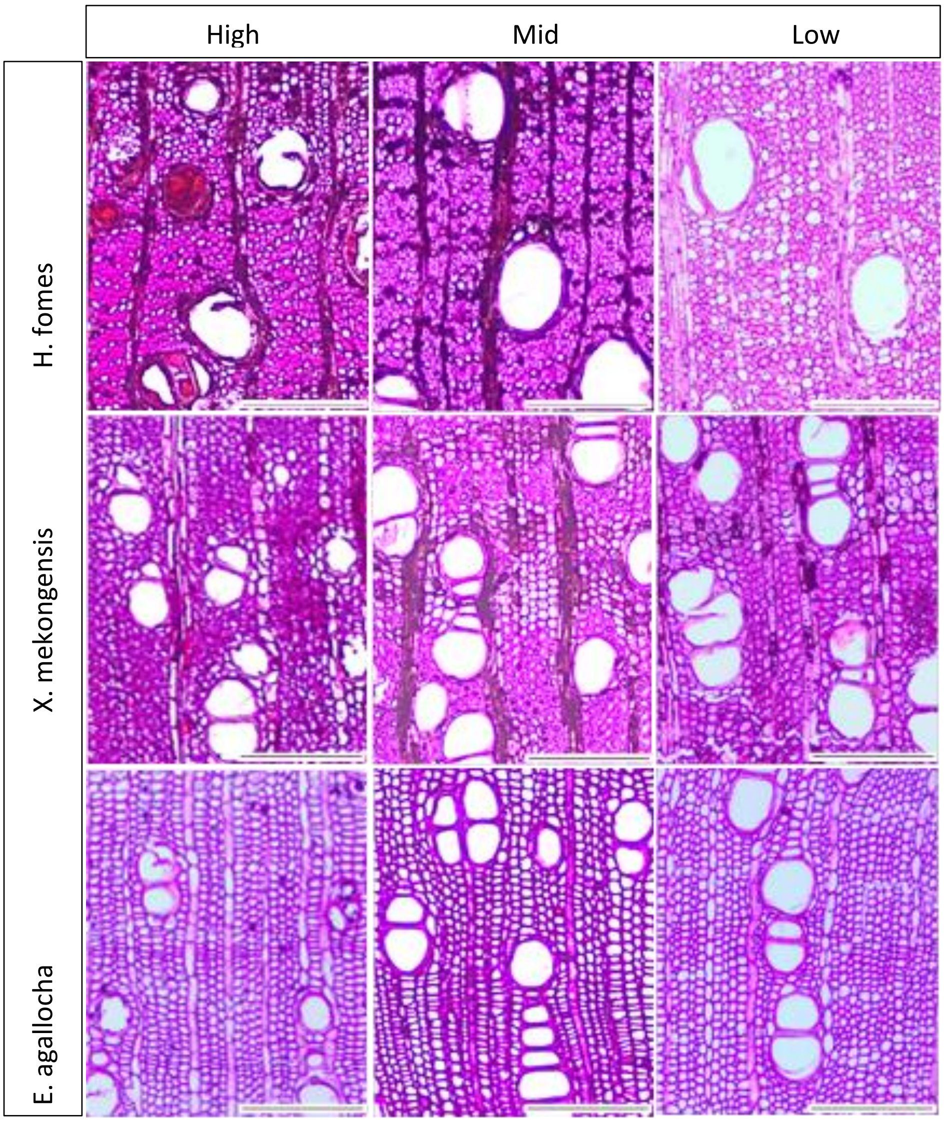

Fig. 8 Vessel characteristics variations in three different species in different salinity.

We focused our examinations on the outermost portion of the cores, which contained the recent formed xylem and were most likely to be related to the environmental conditions as we measured in salinity in the soil samples. Vessel density (number of vessel per mm^2), grouping (ratio of group vessel to total number of vessel) and diameters (radial, tangential and average) of vessels were measured and compared.

Vessel characteristics varied among the regions, but not all characteristics had the same spatial patterns (Fig. 8). Vessel diameter was higher and in the low and medium salinity area than in the higher salinity zone for *H. fomes* and *X. mekongensis*. On the other hand, *E. agallocha* showed a slightly wider vessel size in the lower salinity zone. Vessel density variation was not significant among the three salinity zones for each species. However, lower salinity area had higher number of vessels. Grouping index also showed a similar pattern among the zones for the studied species. Frequency distributions of the average vessel dimensions in *H. fomes* showed a similar pattern in mid and low salinity zones, however in the higher salinity zone the pattern was slightly left skewed (Fig. 5a). In *X. mekongensis* and *E. agallocha*, the average vessel diameter showed similar pattern (normal distribution) but in the higher salinity zone the wider vessels were less (Fig. 9B &C).

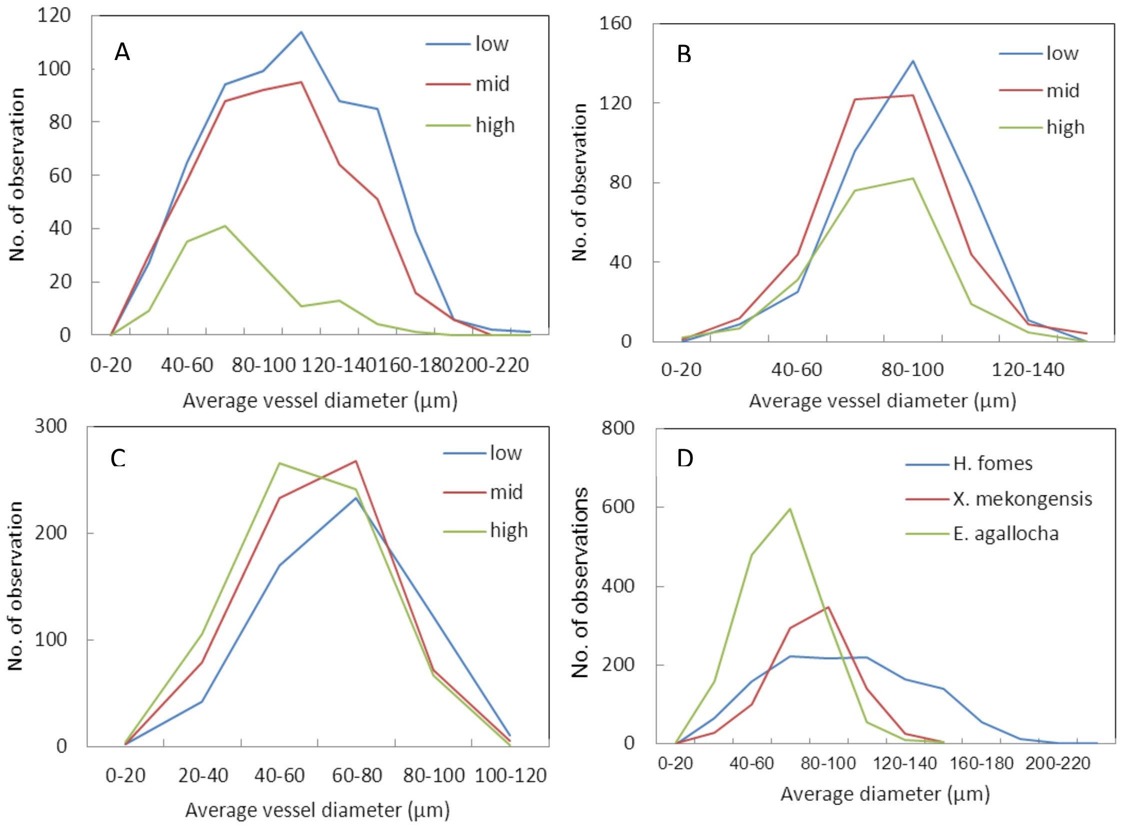

Fig. 9 Frequency distribution of the average vessel diameters. A, H. fomes; B, X. mekongensis; C, E. agallocha in the three salinity zones; D, comparison among the three species.

Compared to *H. fomes*, the frequency distributions in *E. agallocha* and *X. mekongensis* showed a left skewed curve for average vessel diameter (Fig. 9D). However, E. agallocha showed higher number of vessels with smaller diameters than *X. mekongensis*. *H. fomes* showed larger size vessels but the trade-off is narrow between the vessel size and density. As *E. agallocha* not only has smaller vessels than other two species but also has a larger proportion of the tiniest vessels that explains a link between small vessel diameter and low vulnerability to cavitation.

From this analysis, it can be explained that vessel diameter is more sensitive to changes in salinity. The water transport system of *H. fomes* is composed with higher vessel diameters but with low vessel density, and grouping compared to two other species that is expected to be vulnerable based on a physiological interpretation of the combination of observed vessel characteristics due to the cavitation risk of the system.

4. Perspectives for future collaboration between units (1 page)

The currently completed project by the fellow was initiated from a previous collaborative pilot project in 2006 on Bangladesh mangroves at RMCA. This project developed baseline growth data and methods to explore the dynamics on Bangladesh mangroves. From this project, both units mutually agreed to broaden the horizon on mangroves and other ecosystems extending to South Asia. From the proposed project, the host unit is benefited from the fieldworks of the fellow with more number of Bangladeshi mangrove species which is an important mission of RMCA, in addition to scientific contribution.

The Wood Biology Service at Royal Museum for Central Africa (RMCA) is equipped with modern laboratory facilities for wood analysis with different perspectives and high quality scientists, microtomist, collection manager and other staffs. Six additional researchers, who are formally member of the scientific staff of different universities in Belgium, are conducting the major part of their research in the Laboratory. Moreover, many PhD students from different Belgian and foreign universities are part of the research team. RMCA collaborates with its local and international partners mainly in the natural sciences area focusing on climate variability and its impacts on the ecosystems and society. Even though the main focuses of RMCA on African regions but has long collaborations to other tropical areas across the world. Therefore, scope for new collaborations with the fellow to resolve forest and environmental problems in South Asian regions that certainly coincides with the current initiatives of the host unit on the global environmental change and UN REDD+ programs.

Considering the range of scientific areas covered by RMCA, new collaboration can be in different avenues, such as sharing resources, contacts, and information. For example, RMCA has a large collection database of species that may be useful to the fellow and other researchers of Forestry and Environmental Science Department at Shajalal University of Science and Technology (SUST), Bangladesh. In addition, RMCA has links with other in many Asian countries that may be useful for SUST to stimulate sustainable regional scientific action. The researchers of RMCA can profit from fieldworks and having more number of samples from different species for collection purpose as well as for comparative studies. In particular, one of the important issues of SUST is capacity-building of the students and researchers. This can be done through joint PhD projects via collaborative universities of RMCA. Sources of funding for the project/s can be explored jointly externally to the donor organisation/s such as EU, UNESCO, BELSPO etc. It is to the interest of both units that efforts to continue collaboration and be carried in future.

5. Valorisation/Diffusion (including Publications, Conferences, Seminars, Missions abroad...

Peer reviewed Journals

1. Based on the review of tree-ring characters of 105 tree species growing in Bangladesh, we prepared a manuscript paper titled on 'Tree rings in Bangladesh - a perspective'.

2. One manuscript has been prepared on 'Tree ring characters in *Heritiera fomes* in Sundarbans' that will be submitted to *Trees- Structure and Function* soon.

3. A manuscript has been prepared on 'Dendroclimotogical potential of *Heritiera fomes* in Sundarbans' for *PlosOne*.

4. A manuscript is under preparation on 'Hydraulic traits variation in the Sundarbans' for relevant journal.

Conference

1. One abstract on 'Exploring Bangladesh mangroves - ordinary glitches but new opportunities for dendrochronology' has been accepted for oral presentation in 4thAsian Dendrochronological Conference which will be held on 09-12 March 2015 at Kathmandu, Nepal.

2. An abstract on 'Tree rings in *Diploknema butyracea* (Roxburgh) H. J. Lam - a potential tool for understanding the land use history and growth dynamics in Nepal' has been also accepted for poster session in 4thAsian Dendrochronological Conference which will be held on 09-12 March 2015 at Kathmandu, Nepal.

Missions

1. In January 2013, I have established a pinning experiment in the targeted species in seven sites. So, fieldworks were carried out from 10-03-2014 to 08-04-2014 in Sundarbans, Bangladesh. During this mission pinning sample trees were harvested and fresh samples were collected for cambial analysis from three addition sites.

2. From 30-10-2014 to 7-11-2014, again field works were carried out in the Sundarbans, Bangladesh. During this mission a total of 255 wood cores were collected from 60 permanent sample plots in collaboration with Mr. SK Sarker from University of Glasgow, UK. Detail anatomical study will be carried out with these samples to understand their anatomical variations with changing environmental conditions to address our third objective.

Collaborations

1. Collaboration established with Professor Pieter Zuidema, Forest Ecology and Management Group, Wageningen University, The Netherlands. Under this collaboration one of MSc student is now conducting thesis on one of mangrove species from the Sundarbans under joint supervision.

2. Collaboration has been started with Professor Jason Matthiopoulos and Mr. SK Sarker, Spatial and Population Ecology Research group, University of Glasgow, UK. Under this

collaboration we currently write a joint manuscript on hydraulic traits variation in the Sundarbans.

<u>Collections</u>

1. Within the framework of this project 115 stem discs and 275 wood cores covering 20 species were collected and indexed in the collection of museum.

2. A total of 300 slides were prepared from the collected wood samples that are also indexed in the collection of museum.

6. Skills/Added value transferred to home institution abroad (1/2 page)

The fellow learned new laboratory techniques of dendrochronology and wood biology at RMCA. Moreover, the fellow had an opportunity to have experience on mangroves of different continents studying samples from the collection. Take home experiences on mangrove dendrochronology such as tree-ring measurement, cross dating, climate-growth analysis etc. and techniques related to wood biology such as cambial, phloem analysis, microtomy, microscopy etc. are the added advantages for home institution because that can help to extend horizon of the existing wood biology laboratory at the home institution. The newly generated knowledge on Bangladesh mangroves is a spin-off to update the reading and lecture materials of the on-going Mangrove Ecology and Wood Anatomy courses at home institution. Besides that the fellow gained hands on training on specimen collection, cataloging and conservation techniques that will help the fellow to develop a xylarium at home which is long term goal of the fellow's home institution.