The bioinformatics method of protein dehydration induced drought stress in *Hevea brasiliensis* Muell. Arg.

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**Abstract.** *Hevea brasiliensis* Muell. Arg. (rubber plant) is one of the most significant sources of natural rubber in the world. Planting material used is vegetative propagation that called a clone. Each rubber clone has specific adaptability. Adaptation of rubber in drought area is a complex trait. The current study assesses of the bioinformatics on the NCBI database as well as expected the physicochemical of protein dehydration. Several parameters of physicochemical protein dehydration in *H. brasiliensis* were varied among the genes observed. There is twenty (20) protein dehydration induced from *H. brasiliensis* deposited in NCBI. The length of the genes was alternated with the genes ascertained. Several lines of coded amino acids were 370 to 1490 bp. The mitochondria target peptide value diversified from 0.020 to 0.342, signifying that it is expected to be a presence. The present resulted indicated the prominence of the variation and role of a physical and chemical characteristic of the distinct amino acids in protein dehydration genes as drought stress in rubber.

1. **Introduction**

Abiotic stress or dryness was the main problem causes poor the plant growth and decrease crop yields [1]. Drought affects 45% of agricultural land in the world [2]. The drought caused by salt soil areas and higher atmospheric temperatures combined with the overhead of sun and low relative humidity (RH). Drought tolerances was a complicated quality to influenced by genes, the interaction between genes and environmental factors [3].

The genomic differences in the presence of plant responses contain the expression of induced genes in actual plants to protect from drought [4-6]. A full length of abiotic stress to induced the genes classify two functional the categories and the regulation of proteins. Firstly category includes the gene encoding to a protein membrane, enzymes for, detoxification (dehydrons, dehydrorseorbate reductase, superoxide dismutases, quinone reductase, and ascorbate peroxidase), osmolyte biosynthesis and protein to macromolecular protecting (LEA protein, chaperons, and anti-freezing protein also mRNA binding protein) [2]. Another group is encoding genes for transcription factor, protein kinases (SRK2E) and ribosom, receptor protein kinases also signal proteinase transduction (phospholipase C and phosphodiesterases).
The rubber was consisted previously of quantifying level expression genes and several abiotic stress responsive to transcripts [7,8,9,10]. Indicates the relationship of DRE or CRT binding factors and ABC transporters from drought tolerance about the analysis of protein expression to different clones of rubber. The peroxidase and WRKY transcriptions factor and slowly embryogenesis profuse 5 (LEA5) protein have been reported the first association of drought tolerance [7-9].

Development of improved molecular biology methods and new functional genes to expand tolerance mechanisms in rubber tree drought. Furthermore, HbCuZnSOD and HbMmSOD have been cloned in PB 260 conferred enhance drought tolerance [11].

Despite the abundance of drought tolerance in *Hevea*, limited studies have been focused on protein dehydration induced in a rubber plant. Protein dehydration has been reported to increase of drought tolerance with overexpression of the gene. This study aims to evaluate bioinformatics of dehydrated proteins induced by rubber plant (*H. brasiliensis*).

2. Materials and Method

2.1. Materials

Twenty (20) calculated of protein dehydration induced of rubber (*Hevea brasiliensis*) deposited from NCBI database. The NCBI reference sequence of DNA and amino acids was used in this study:

1. XM_021802160 and XP_021657852.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 6-like (LOC110648060), transcript variant X2).
2. XM-021802159 and XP_021657851.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 6-like (LOC110648060), transcript variant X1).
3. XM_021790164 and XP_021645856.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 3-like (LOC110639282)).
4. XM_021781572 and XP_021637264.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 4-like (LOC110633098)).
5. XM_021780257 and XP_021635949.1 (*H.brasiliensis* thioredoxin-like protein CDSP32, chloroplastic (LOC110632135)).
6. XM_021779394 and XP_021635086.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 4-like (LOC110631527), transcript variant X2).
7. XM_021779393 and XP_021635085.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 4-like (LOC110631527), transcript variant X1).
8. XM_021814868 and XP_021674178.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 4-like (LOC110660253)).
9. XM_021814058 and XP_021669750.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 3-like (LOC110657014), transcript variant X3).
10. XM_021814057 and 021669749.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 3-like (LOC110657014), transcript variant X2).
11. XM_021814056 and XP_021669748.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 3-like (LOC110657014), transcript variant X1).
12. XM_021799893 and XP_021655585.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 6-like (LOC110646422), transcript variant X7).
13. XM_021799886 and XP_021655578.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 6-like (LOC110646422), transcript variant X6).
14. XM_021799879 and XP_021655571.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 6-like (LOC110646422), transcript variant X5).
15. XM_021799871 and XP_021655563.1 (*H.brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 6-like (LOC110646422), transcript variant X4).
16. XM_021799864 and XP_021655556.1 (*H. brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 6-like (LOC110646422), transcript variant X3).
17. XM_021799858 and XP_021655550.1 (*H. brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 6-like (LOC110646422), transcript variant X2).
18. XM_021799853 and XP_021655545.1 (*H. brasiliensis* protein DEHYDRATION-INDUCED 19 homolog 6-like (LOC110646422), transcript variant X1).
19. KX661028 and AOO86553.1 (*H. brasiliensis* cultivar 93-114 ErbB-3 binding protein 1 (EBP1)).
20. HQ687666 and AEE81756.1 (*H. brasiliensis* cultivar CATAS 7-33-97 metallothionein (MT2)).

2.2. Physical and chemical characteristics of the protein dehydration induced

The structure and physicochemical characteristics from protein dehydration induced from *H. brasiliensis* was analysis with Protparam online searched (web.expasy.org/protparam/). The calculated factor designate of molecular mass, a total number of atoms, theoretical isoelectric point values, extinction coefficients, predictable half-life, aliphatic index, instability and high average of hydropathy beforehand reported [12].

2.3. Possible peptide transfer and subcellular localization of induced protein dehydration

Transit peptide analyzed by accessing the online P1.1 target server (www.cbs.dtu.dk/services/targetp/). This position corresponds to the estimated presence of one of the chloroplast pre-sequential N-terminal terminals of transit peptide (cTP), mitochondrial targeting peptide (mTP), and also the peptide signal signal (SP) peptide pathway. The prediction tool for localization subcellular proteins (PSORT) is used to access online predictions with psort.hgc.jp/form.html to control the subcellular determinations of protein-induced dehydration as previously [13].

3. Results and Discussion

3.1. Physicochemical characteristics of the protein dehydration induced in *H. brasiliensis*

Several characters of physicochemical protein dehydration induced by *H. brasiliensis* was shown in Table 1. The genes length changes with ascertained genes. Individual lines are coded of amino acid from 370 to 1490 bp. The heterogeneity related in molecular weight, the point of theoretical isoelectric, total number atom, coefficient extinction and instability, and average throughout hydropathicity analyzed genes. Despite the availability of thousands of stress associated ESTs of *H. brasiliensis*, quantitative gene expression analysis of these genes has only recently been attempted for the identification of candidate genes/factors that are contributing to drought tolerance. With the advent of the qPCR technique, it is easier to quantify each gene and establish their relevance under the given stress situations. For quantitative expression analysis in RRII 105 (susceptible) and RRIM 600 clones, RRII 208 and RRII 430 (tolerant) clones using quantitative PCR (qPCR) found peroxidase was overexpressed many folds in tolerant clones compared to the susceptible one. LEA 5 gene also showed a similar trend but with a lesser extent than peroxidase (9). The energy of biosynthesis expression and ROS forage systems associated with genes, including HbAPX, HbCAT, HbCuZnSOD, HbACAT, HbMnSOD, HbCOA, HbATP, and significantly regulated by the stress of drought [14].

Table 1. Physicochemical characteristic of the protein dehydration induced in *H.brasiliensis*

| Variant | R1   | R2   | R3   | R4   | R5   | R6   | R7   |
|---------|------|------|------|------|------|------|------|
| Length of genes/bp | 1177 | 1125 | 1249 | 1166 | 1161 | 1345 | 1427 |
| Number of amino acids | 207  | 208  | 215  | 215  | 299  | 234  | 234  |
| Molecular weight | 98228.64 | 93977.81 | 105210.33 | 97003.39 | 95717.73 | 114374.97 | 121292.71 |
| Theoretical isoelectric points values | 5.05 | 4.98 | 5.04 | 4.98 | 5.06 | 4.95 | 4.94 |
| Total number of atoms | 12636 | 12086 | 13554 | 12493 | 12310 | 1345 | 1427 |
| Extinction coefficient | 15125 | 14500 | 16000 | 14000 | 14625 | 17750 | 18875 |
| Half-life period | 30 h | 7.2 h | 4.4 h | 7.2 h | 1.2 h | 7.2 h | 7.2 h |
| Instability coefficient | 46.10 | 46.03 | 39.57 | 44.78 | 44.56 | 42.03 | 42.76 |
| Aliphatic index | 29.31 | 28.53 | 28.02 | 26.16 | 30.84 | 28.70 | 28.59 |
| Grand average of hydropathicity | 0.757 | 0.738 | 0.720 | 0.643 | 0.783 | 0.747 | 0.746 |

Table 1. Continued

| Variant | R8   | R9   | R10  | R11  | R12  | R13  | R14  |
|---------|------|------|------|------|------|------|------|
| Length of genes/bp | 1198 | 1085 | 1157 | 1170 | 1024 | 1029 | 1063 |
| Number of amino acids | 216  | 192  | 215  | 220  | 206  | 217  | 220  |
| Molecular weight | 99610.33 | 90845.71 | 96726.11 | 97645.09 | 85215.03 | 85618.49 | 88591.73 |
| Theoretical isoelectric points values | 4.98 | 5.09 | 4.99 | 5.08 | 5.08 | 4.99 | 5.08 |
| Total number of atoms | 12873 | 11766 | 12529 | 12650 | 10964 | 11016 | 11407 |
| Extinction coefficient | 14000 | 12625 | 13250 | 13375 | 13000 | 13125 | 13375 |
| Half-life period | 7.2 h | 4.4 h | 7.2 h | 4.4 h | 4.4 h | 7.2 h | 4.4 h |
| Instability coefficient | 42.65 | 31.26 | 31.20 | 30.41 | 47.98 | 47.79 | 48.28 |
| Aliphatic index | 28.55 | 29.03 | 28.78 | 28.83 | 29.49 | 29.54 | 29.26 |
| Grand average of hydropathicity | 0.679 | 0.683 | 0.672 | 0.670 | 0.755 | 0.756 | 0.743 |

Table 1. Continued

| Variant | R15  | R16  | R17  | R18  | R19  | R20  |
|---------|------|------|------|------|------|------|
| Length of genes/bp | 1078 | 1078 | 1107 | 1114 | 1490 | 372  |
| Number of amino acids | 221  | 221  | 230  | 231  | 395  | 78   |
| Molecular weight | 89205.42 | 89700.01 | 92255.77 | 92819.42 | 121231.95 | 29749.03 |
| Theoretical isoelectric points values | 5.08 | 5.07 | 5.07 | 4.98 | 5.04 | 5.27 |
| Total number of atoms | 11488 | 11545 | 11881 | 11951 | 15690 | 3817 |
| Extinction coefficient | 13500 | 13625 | 13875 | 14000 | 16250 | 4375 |
| Half-life period | 4.4 h | 1.2 h | 4.4 h | 7.2 h | 30 h | 1.2 h |
| Instability coefficient | 47.42 | 47.21 | 4.78 | 47.55 | 33.34 | 37.64 |
| Aliphatic index | 29.66 | 29.59 | 29.18 | 29.17 | 32.01 | 32.80 |
| Grand average of hydropathicity | 0.750 | 0.754 | 0.738 | 0.740 | 0.730 | 0.810 |

3.2. Possible transfer of peptide and subcellular locations
The possibility of a potential transfer peptide from peptide transfer reside in the polyprenol gene reductase. Twenty (20) certainties were defined: the chloroplast transit of peptide and the
mitochondrial target peptide and the signal of pathway secretory also the prospect prediction. On the other hand, the chloroplast peptide was not indication peptides because short, showed different of chloroplast transit of peptide but great indicator relative of the peptide from pathway secretion in rubber tree protein dehydration induced. It is remarkable of mitochondria target peptide divided from 0.020-0.342, it is expected to attend was significant. The mitochondrial target peptide (X2), rubber protein dehydration induced. HbCuZnSOD overexpression in clone PB 260 indicated as drought tolerance (11). These result showed that ROS-scavenging enzymes as roles crucial in the mechanism of drought tolerance. The genes representative in mitochondria (HbAPX), (15) and HbATP (16), a gene of ascorbate peroxidase were cloned.

Table 2. The promising of potential transit peptide of in H. brasiliensis

| Variant | Chloroplast transit peptide | Mitochondrial target peptide | Signal peptide of secretory pathway | Reliability prediction |
|---------|-----------------------------|-----------------------------|-------------------------------------|----------------------|
| R1      | 0.331                       | 0.024                       | 0.709                               | 4                    |
| R2      | 0.080                       | 0.096                       | 0.052                               | 3                    |
| R3      | 0.305                       | 0.020                       | 0.172                               | 5                    |
| R4      | 0.088                       | 0.044                       | 0.273                               | 5                    |
| R5      | 0.344                       | 0.129                       | 0.030                               | 5                    |
| R6      | 0.376                       | 0.035                       | 0.096                               | 5                    |
| R7      | 0.329                       | 0.055                       | 0.132                               | 5                    |
| R8      | 0.171                       | 0.122                       | 0.054                               | 4                    |
| R9      | 0.157                       | 0.253                       | 0.365                               | 4                    |
| R10     | 0.155                       | 0.043                       | 0.486                               | 4                    |
| R11     | 0.151                       | 0.028                       | 0.042                               | 5                    |
| R12     | 0.270                       | 0.139                       | 0.035                               | 5                    |
| R13     | 0.211                       | 0.134                       | 0.026                               | 5                    |
| R14     | 0.271                       | 0.241                       | 0.026                               | 5                    |
| R15     | 0.271                       | 0.241                       | 0.026                               | 5                    |
| R16     | 0.261                       | 0.341                       | 0.027                               | 5                    |
| R17     | 0.210                       | 0.342                       | 0.037                               | 5                    |
| R18     | 0.163                       | 0.123                       | 0.067                               | 5                    |
| R19     | 0.213                       | 0.051                       | 0.069                               | 5                    |
| R20     | 0.115                       | 0.269                       | 0.106                               | 5                    |

The location subcellular of protein dehydration induced in rubber shown in Table 2. The localization subcellular of the genes are frequently miserable in the mitochondrial matrix space (Mms). There are 18 variants were located in the nucleus (N). The R5 was a presence in cytoplasmic (C). There are six variants (R3, R5, R9, R10, R11, and R20) in chloroplast stroma (Cs). Seven options (X5, X10, X11, x14, X17, X18, and X18) were located in chloroplast thylakoid membrane (Ctm). The R20 was a presence in chloroplast thylakoid space (Cts). Twelve variants (R1, R2, R6, R7, R12, R13, R14, R15, R16, R17, R18, and R19) were located in microbody/peroxisome (M/p). Eight samples (R1, R2, R3, R4, R6, R7, R8, and R9) were located in the mitochondrial inner membrane (Mim). Two samples (R4 and R8) were located in mitochondrial intermembrane space (Mim). Five samples (R12, R13, R15, R16, R19) were found in endoplasmic reticulum (Er) membrane. The drought variations in effects on physiological metabolism different invariant growing and the stage of development of rubber trees (17). The coefficient of transpiration (18), integrity membrane (19), osmoregulation, the pressure of laticifer turgor (20), potential solute low (21) found related to drought tolerance. Malondialdehyde protein accumulation mostly causes damage membrane cells in the plant or animal. The content of MDA increase coincides with being broken down inside seedlings of the rubber trees. Chlorophyll comfortable indicated drought rephased the chloroplasts structure, photosynthesis influence and expression of HbRbsS gene (14). Wheat leaves showed a similar result under moderate drought
pressure, acidification of the thylakoid lumen in dryness stress and the leaves associated with activity and increasing fractions about PSI.

Table 3. Subcellular localization of protein dehydration induced in *H. brasiliensis*

| Variant | Ns | C | Cs | Ctm | Cts | M (p) | Mim | Mms | Mis | Er (m) |
|---------|----|---|----|-----|-----|-------|-----|-----|-----|-------|
| R1      | 0.700 | nd | nd | nd | nd | 0.300 | 0.121 | 0.437 | nd | nd |
| R2      | 0.700 | nd | nd | nd | nd | 0.300 | 0.121 | 0.437 | nd | nd |
| R3      | 0.674 | nd | nd | nd | nd | 0.403 | 0.720 | nd | nd |
| R4      | 0.674 | nd | nd | nd | nd | 0.600 | 0.920 | 0.600 | nd |
| R5      | nd | 0.450 | 0.200 | 0.200 | nd | Nd | 0.360 | nd |
| R6      | 0.300 | nd | nd | nd | nd | 0.300 | 0.211 | 0.495 | nd |
| R7      | 0.300 | nd | nd | nd | nd | 0.300 | 0.211 | 0.495 | nd |
| R8      | 0.360 | nd | nd | nd | nd | 0.345 | 0.650 | 0.345 | nd |
| R9      | 0.880 | nd | 0.498 | nd | nd | Nd | 0.464 | nd |
| R10     | 0.960 | nd | 0.498 | 0.246 | nd | nd | 0.464 | nd |
| R11     | 0.880 | nd | 0.498 | 0.246 | nd | nd | 0.464 | nd |
| R12     | 0.600 | nd | nd | nd | nd | 0.300 | 0.100 | nd | 0.000 |
| R13     | 0.600 | nd | nd | Nd | nd | 0.300 | 0.100 | nd | 0.000 |
| R14     | 0.600 | nd | nd | 0.100 | nd | 0.329 | 0.100 | nd | 0.000 |
| R15     | 0.600 | nd | Nd | nd | 0.300 | nd | 0.100 | nd | 0.000 |
| R16     | 0.600 | nd | Nd | nd | 0.300 | nd | 0.100 | nd | 0.000 |
| R17     | 0.600 | nd | nd | 0.100 | 0.335 | nd | 0.100 | nd |
| R18     | 0.600 | nd | nd | 0.100 | 0.331 | nd | 0.100 | nd |
| R19     | 0.300 | nd | nd | nd | nd | 0.300 | 0.100 | nd | 0.000 |
| R20     | nd | nd | 0.794 | 0.625 | 0.96 | Nd | Nd | 0.100 | nd |

Note:
N: Nucleus
C: Cytoplasm
Cs: Chloroplast stroma
Ctm: Chloroplast thylakoid membrane
Cts: Chloroplast thylakoid space
M (p): Microbody (peroxisome)
Mim: Mitochondrial inner membrane
Mms: Mitochondrial matrix space
Mis: Mitochondrial intermembrane space
Er (m): Endoplasmic reticulum (membrane)
nd: not detected

Conclusions
This study clarified the variable, physical role, and specific chemical or rubber protein dehydration induced. Our result is preceding that the studies on the location subcellular of drought tolerance genes in the plasma membrane.

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References
[1] Acquaah G 2012 Principles of plant genetics and breeding 2nd ed. Oxford UK Wiley 20 pp
[2] Dos Reis SP, Lima AM, de Souza CRB 2012 Recent molecular advances on downstream plant responses to abiotic stress *Intl. J. Mol. Sci.* 13 8628–8647.
[3] Tardieu F, Tuberosa R 2010 Dissection and modelling of abiotic stress tolerance in plants *Curr. Opin. Plant Biol.* 13 206–2012.
[4] Colmer TD, Pedersen O 2008 Underwater photosynthesis and respiration in leaves of submerged wetland plants: gas films improve CO2 and O2 exchange New Phytol. 177 918–926.
[5] Mittler R 2006 Abiotic stress, the field environment and stress combination Trends Plant Sci. 11 15–19.
[6] Fujita M, Fujita Y, Noutoshi Y, Takahashi F, Narusaka Y, Yamaguchi-Shinozaki K, Shinozaki K 2006 Crosstalk between abiotic and biotic stress responses: a current view from the points of convergence in the stress signaling networks Curr. Opin. Plant Biol. 9 436–442.
[7] Thomas M, Shatik MBM, Saha T, Jacob J, Schaffer AR, Luke LP, Kurvilla L, Annamalainathan K, Krisnakumar R 2011 Screening of drought-responsive transcripts of Hevea brasiliensis and identification of candidate genes for drought tolerance J. Plant Biol. 38&39 111–118
[8] Thomas M, Shatik MBM, Luke LP, Sumesh KV, Sathesh PR, Annamalainathan K, Jacob J 2012. Stress responsive transcripts and their association with drought tolerance in Hevea brasiliensis J. Plant Crop. 40 180–187
[9] Shatik MBM, Luke LP, Thomas M, Sumesh KV, Sathesh PR 2011 Identification of drought tolerant genes by quantitative expression analysis in Hevea brasiliensis IRRDB International Rubber Conference, 15-16 Dec. Chiang Main, Thailand
[10] Shatik MBM, Kurvilla L, Thomas M, Luke LP, Sathesh PR, Annamalainathan K, Jacob J 2012 Quantitative expression analysis of stress responsive genes under cold stress in Hevea brasiliensis Rubber Sci. 25 199–213
[11] Leclercq J, Martin F, Sanier C, Clement-Vidal A, Fabre D, Oliver G, Lardet L, Ayar A, Peyramard M, Montoro P 2012 Over-expression of a cytosolic isoform of the HbCuZnSOD gene in Hevea brasiliensis changes its response to a water deficit Plant Mol. Biol. 80 255–272
[12] Basyuni M, Wasielah M, Sumardi 2017. Bioinformatics study of the mangrove actin genes J. Phys. : Conf. Ser. 801 012013
[13] Basyuni M, Wati R 2018 Bioinformatics analysis of the predicted polyphenol reductase genes in higher plants J. Phys. : Conf. Ser. 978 012050
[14] Wang LF 2014 Physiological and molecular responses to drought stress in rubber tree (Hevea brasiliensis Muell. Arg.) Plant Physiol. Biochem. 83 243-249
[15] Mai J, Herbette S, Vandame M, Kositsup B, Kasemso P, Cavaloc E, Julien JL, Ameglio T, Roeckel-Drevet P 2009 Effect of chilling on photosynthesis and antioxidant enzymes in Hevea brasiliensis Muell. Ar. Trees 23 863–874
[16] Chye ML, Tan CT 1992 Isolation and nucleotide sequence of a cDNA clone encoding the beta subunit of mitochondrial ATP synthase from Hevea brasiliensis Plant Mol. Biol. 18 611–612.
[17] Devakumar A, Gururaja Rao G, Rajagopal R, Sanjeeva Rao P, George M, Vijayakumar K, Sethuraj M 1988 Studies on the soil-plant-atmosphere system in Hevea: II. Seasonal effects on water relations and yield Indian J. Nat. Rubber Res. 1 45–60.
[18] Nair DB, Dey SK, Rajagopal R, Vijayakumar KR, Sethuraj MR 1996 Synergistic effect of heat and osmotic stress in causing membrane injury in Hevea brasiliensis J. Plant Biol. 39 177e181.
[19] Reddy Y 2000 Effect of moisture stress on stability of membrane integrity in Hevea brasiliensis across temperature regimes Indian J. For. 23 110–111
[20] Ranasasinghe MS, Milburn JA1995 Xylem conduction and cavitation in Hevea brasiliensis J. Exp. Bot. 46 1693–1700.
[21] Ayutthaya SIN, Do FC, Pannangpetch K, Junjittakarn J, Maeght JL, Rocheteau A, Cochard H 2011 Water loss regulation in mature Hevea brasiliensis effects of intermittent drought in the rainy season and hydraulic regulation Tree Physiol. 31 751–762.