

***Arabidopsis thaliana* plant natriuretic peptide active domain forms amyloid-like fibrils in a pH-dependent manner**

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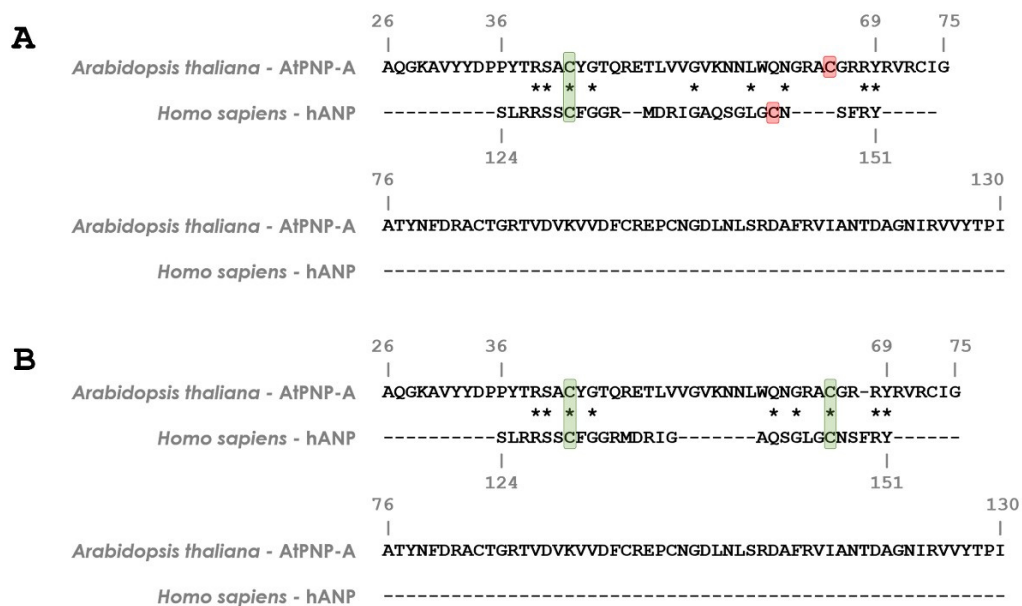
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Supplementary File 1

Sequence alignments

Sequence alignment of hANP and AtPNP-A

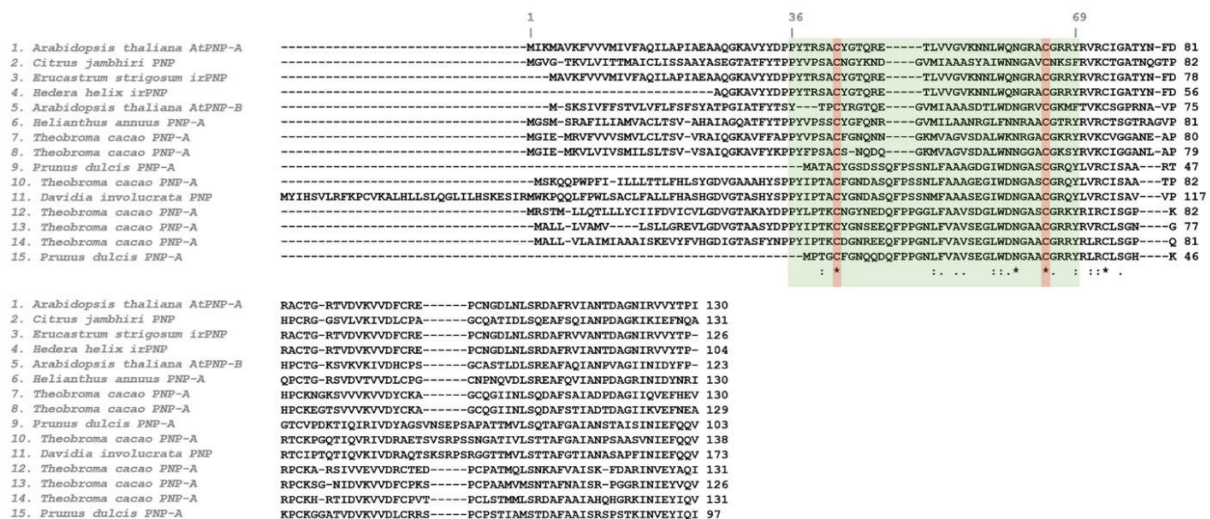
The amino acid sequences of the AtPNP-A (Q9ZV52) and the hANP (P01160) were extracted from UniprotKB, free from their signal peptide or pro-peptide sequences. The two peptide sequences were then aligned applying the Needleman-Wunsch algorithm [87] and the result was manually edited utilizing Jalview [88].



Supplementary Figure S1. Sequence alignment of hANP and AtPNP-A. (A) The amino acid sequence homology between hANP and AtPNP-A as suggested after the molecular identification of the AtPNP-A [29]. Green box: aligned cysteines, Red box: not aligned cysteines (B) The alignment of hANP and AtPNP-A we propose based on the available experimental data supporting the importance of the disulfide bond to obtain their functional structure (green boxes). Asterisks (*) indicate positions which have a single, fully conserved residue.

Multiple sequence alignment of all know plant natriuretic peptides (PNPs)

Amino acid sequences of all known plant natriuretic peptides (PNPs) were extracted from UniprotKB [89], with the following accession numbers: Q9ZV52 (*Arabidopsis thaliana*), Q9M0C2 (*Arabidopsis thaliana*), Q9ZP41 (*Citrus jambhiri*), A0A251V4M4 (*Helianthus annuus*), A0A061GQD7 (*Theobroma cacao*), A0A061GQ10 (*Theobroma cacao*), A0A061GAM9 (*Theobroma cacao*), A0A061GQI5 (*Theobroma cacao*), A0A061GQU2 (*Theobroma cacao*), A0A061GNK9 (*Theobroma cacao*), Q8RWA0 (*Erucastrum strigosum*), A0A4Y1QUF7 (*Prunus dulcis*), A0A5B7B5A2 (*Davidia involucrata*), A0A4Y1RVQ8 (*Prunus dulcis*), Q84V62 (*Hedera helix*). Multiple sequence alignment of all PNPs sequences was performed utilizing Clustal Omega [90] and Jalview [88].



Supplementary Figure S2. Multiple sequence alignment of all plant natriuretic peptides. Each line represents an entry, obtained from the protein database UniProt [90]. A green box shows the amino acid residues corresponding to the 34 amino acid active domain of the AtPNP-A sequence and the homologous regions corresponding to the rest of the PNPs. As it is observed, the cysteine residues are conserved in all sequences (orange boxes). The corresponding UniProt Accession Numbers (ACs) are given: 1. Q9ZV52, 2. Q9ZP41, 3. Q8RWA0, 4. Q9M0C2, 5. Q84V62, 6. A0A251V4M4, 7. A0A061GQD7, 8. A0A061GQ10, 9. A0A061GQI5, 10. A0A061GQU2, 11. A0A061GAM9, 12. A0A061GNK9. Asterisks (*) indicate positions which have a single, fully conserved residue. Colon (:) indicates conservation between groups of strongly similar properties (> 50% in the Gonnet PAM 250 matrix). Period (.) indicates conservation between groups of weakly similar properties (≤50% and >0 in the Gonnet PAM 250 matrix) [90].