

Damian:

[Plant J.](#) 2013 May;74(3):363-71. doi: 10.1111/tpj.12128. Epub 2013 Apr 5.

Generation of an artificial ring chromosome in Arabidopsis by Cre/LoxP-mediated recombination.

[Murata M](#), [Shibata F](#), [Hironaka A](#), [Kashihara K](#), [Fujimoto S](#), [Yokota E](#), [Nagaki K](#).

Abstract

A eukaryotic chromosome consists of a centromere, two telomeres and a number of replication origins, and 'artificial chromosomes' may be created in yeast and mammals when these three elements are artificially joined and introduced into cells. Plant artificial chromosomes (PACs) have been suggested as new vectors for the development of new crops and as tools for basic research on chromosomes. However, indisputable PAC formation has not yet been confirmed. Here, we present a method for generating PACs in the model plant *Arabidopsis thaliana* using the Cre/LoxP and Activator/Dissociation element systems. The successfully generated PAC, designated AtARC1 (*A. thaliana* artificial ring chromosome 1), originated from a centromeric edge of the long arm of chromosome 2, but its size (2.85 Mb) is much smaller than that of the original chromosome (26.3 Mb). Although AtARC1 contains only a short centromere domain consisting of 180 bp repeats approximately 250 kb in length, compared with the 3 Mb domain on the original chromosome 2, centromere-specific histone H3 (HTR12) was detected on the centromeric region. This result supported the observed stability of the PAC during mitosis in the absence of selection, and transmission of the PAC to the next generation through meiosis. Because AtARC1 contains a unique LoxP site driven by the CaMV 35S promoter, it is possible to introduce a selectable marker and desired transgenes into AtARC1 at the LoxP site using Cre recombinase. Therefore, AtARC1 meets the criteria for a PAC and is a promising vector.

[Plant J.](#) 2013 Mar 8. doi: 10.1111/tpj.12169. [Epub ahead of print]

Heat stress induction of miR398 triggers a regulatory loop that is critical for thermotolerance in Arabidopsis.

[Guan Q](#), [Lu X](#), [Zeng H](#), [Zhang Y](#), [Zhu J](#).

Abstract

microRNAs (miRNAs) play important roles in plant growth and development. Previous studies have shown that down-regulation of miR398 in response to oxidative stress permits up-regulation of one of its target genes, CSD2 (copper/zinc superoxide dismutase), and thereby helps plants to cope with oxidative stress. We report here that heat stress rapidly induces miR398 and reduces transcripts of its target genes CSD1, CSD2 and CCS (a gene encoding a copper chaperone for both CSD1 and CSD2). Transgenic plants expressing miR398-resistant forms of CSD1, CSD2 and CCS under the control of their native promoters are more sensitive to heat stress (as indicated by increased damage at the whole-plant level and to flowers) than transgenic plants expressing normal coding sequences of CSD1, CSD2 or CCS under the control of their native promoters. In contrast, *csd1*, *csd2* and *ccs* mutant plants are more heat-tolerant (as

indicated by less damage to flowers) than the wild-type. Expression of genes encoding heat stress transcription factors (HSF genes) and heat shock proteins (HSP genes) is reduced in heat-sensitive transgenic plants expressing miR398-resistant forms of CSD1, CSD2 or CCS but is enhanced in the heat-tolerant *csd1*, *csd2* and *ccs* plants. Chromatin immunoprecipitation assays revealed that HSFA1b and HSFA7b are the two HSFs responsible for heat induction of miR398. Together, our results suggest that plants use a previously unrecognized strategy to achieve thermotolerance, especially for the protection of reproductive tissues. This strategy involves the down-regulation of CSD genes and their copper chaperone CCS through heat-inducible miR398.

Cell, **Volume 153, Issue 4, 896-909, 9 May 2013**

Sexually Dimorphic Neurons in the Ventromedial Hypothalamus Govern Mating in Both Sexes and Aggression in Males

[Cindy F. Yang](#), [Michael C. Chiang](#), [Daniel C. Gray](#), [Mahalakshmi Prabhakaran](#), [Maricruz Alvarado](#), [Scott A. Juntti](#), [Elizabeth K. Unger](#), [James A. Wells](#), [Nirao M. Shah](#)

Summary

Sexual dimorphisms in the brain underlie behavioral sex differences, but the function of individual sexually dimorphic neuronal populations is poorly understood. Neuronal sexual dimorphisms typically represent quantitative differences in cell number, gene expression, or other features, and it is unknown whether these dimorphisms control sex-typical behavior exclusively in one sex or in both sexes. The progesterone receptor (PR) controls female sexual behavior, and we find many sex differences in number, distribution, or projections of PR-expressing neurons in the adult mouse brain. Using a genetic strategy we developed, we have ablated one such dimorphic PR-expressing neuronal population located in the ventromedial hypothalamus (VMH). Ablation of these neurons in females greatly diminishes sexual receptivity. Strikingly, the corresponding ablation in males reduces mating and aggression. Our findings reveal the functions of a molecularly defined, sexually dimorphic neuronal population in the brain. Moreover, we show that sexually dimorphic neurons can control distinct sex-typical behaviors in both sexes.

Stephanie:

From Plant Molecular Biology:

The pEAQ vector series: the easy and quick way to produce recombinant proteins in plants

Hadrien Peyret • George P. Lomonosoff

Abstract: The pEAQ vectors are a series of plasmids designed to allow easy and quick production of recombinant proteins in plants. Their main feature is the use of the Cowpea Mosaic Virus hypertranslational "CPMV-HT"

expression system, which provides high yields of recombinant protein through extremely high translational efficiency without the need for viral replication. Since their creation, the pEAQ vectors have been used to produce a wide variety of proteins in plants. Viral proteins and Virus-Like Particles (VLPs) have been of particular interest, but other types of proteins including active enzymes have also been expressed. While the pEAQ vectors have mostly been used in a transient expression context, through agroinfiltration of leaves, they have also been shown to be suitable for the production of stably transformed lines of both cell cultures and whole plants. This paper looks back on the genesis of the pEAQ vectors and reviews their use so far.

Fionn:
Nature

Special on GMOs several interesting articles

Architecture and evolution of a minute plant genome

[Enrique Ibarra-Laclette, Eric Lyons, Gustavo Hernández-Guzmán, Claudia Anahí Pérez-Torres, Lorenzo Carretero-Paulet, Tien-Hao Chang, Tianying Lan, Andreanna J. Welch, María Jazmín Abraham Juárez, June Simpson, Araceli Fernández-Cortés, Mario Arteaga-Vázquez, Elsa Góngora-Castillo, Gustavo Acevedo-Hernández, Stephan C. Schuster, Heinz Himmelbauer, André E. Minoche, Sen Xu, Michael Lynch, Araceli Oropeza-Aburto, Sergio Alan Cervantes-Pérez, María de Jesús Ortega-Estrada, Jacob Israel Cervantes-Luevano, Todd P. Michael, Todd Mockler, Douglas Bryant, Alfredo Herrera-Estrella, Victor A. Albert & Luis Herrera-Estrella](#)

It has been argued that the evolution of plant genome size is principally unidirectional and increasing owing to the varied action of whole-genome duplications (WGDs) and mobile element proliferation¹. However, extreme genome size reductions have been reported in the angiosperm family tree. Here we report the sequence of the 82-megabase genome of the carnivorous bladderwort plant *Utricularia gibba*. Despite its tiny size, the *U. gibba* genome accommodates a typical number of genes for a plant, with the main difference from other plant genomes arising from a drastic reduction in non-genic DNA. Unexpectedly, we identified at least three rounds of WGD in *U. gibba* since common ancestry with tomato (*Solanum*) and grape (*Vitis*). The compressed architecture of the *U. gibba* genome indicates that a small fraction of intergenic DNA, with few or no active retrotransposons, is sufficient to regulate and integrate all the processes required for the development and reproduction of a complex organism.

PNAS

Heat shock protein (Hsp) 70 is an activator of the Hsp104 motor.

Lee J, Kim JH, Biter AB, Sielaff B, Lee S, Tsai FT

Abstract

Heat shock protein (Hsp) 104 is a ring-forming, protein-remodeling machine that harnesses the energy of ATP binding and hydrolysis to drive protein disaggregation. Although Hsp104 is an active ATPase, the recovery of functional protein requires the species-specific cooperation of the Hsp70 system. However, like Hsp104, Hsp70 is an active ATPase, which recognizes aggregated and aggregation-prone proteins, making it difficult to differentiate the mechanistic roles of Hsp104 and Hsp70 during protein disaggregation. Mapping the Hsp70-binding sites in yeast Hsp104 using peptide array technology and photo-cross-linking revealed a striking conservation of the primary Hsp70-binding motifs on the Hsp104 middle-domain across species, despite lack of sequence identity. Remarkably, inserting a Strep-Tactin binding motif at the spatially conserved Hsp70-binding site elicits the Hsp104 protein disaggregating activity that now depends on Strep-Tactin but no longer requires Hsp70/40. Consistent with a Strep-Tactin-dependent activation step, we found that full-length Hsp70 on its own could activate the Hsp104 hexamer by promoting intersubunit coordination, suggesting that Hsp70 is an activator of the Hsp104 motor.

Keith:

Nature of Structural and Molecular Biology – April 2013

Conformational selection of translation initiation factor 3 signals proper substrate selection

Nature Structural & Molecular Biology **20**, 628–633 (2013)

Margaret M Elvekrog & Ruben L Gonzalez Jr

Department of Chemistry, Columbia University, New York, New York, USA.

During translation, initiation factor 3 (IF3) binds to the small (30S) ribosomal subunit and regulates the fidelity with which the initiator tRNA and mRNA start codon substrates are selected into the 30S initiation complex (30S IC). The molecular mechanism through which IF3 promotes the recognition and signaling of correct substrate selection, however, remains poorly defined. Using single-molecule fluorescence resonance energy transfer, we show that 30S IC-bound *Escherichia coli* IF3 exists in a dynamic equilibrium between at least three conformations. We found that recognition of a proper anticodon-codon interaction between initiator tRNA and the start codon within a completely assembled

30S IC selectively shifts this equilibrium toward a single conformation of IF3. Our results strongly support a conformational selection model in which the conformation of IF3 that is selectively stabilized within a completely and correctly assembled 30S IC facilitates further progress along the initiation pathway

Journal of Biological Chemistry – May 2013

Mechanism of an ATP-independent Protein Disaggregase

I. STRUCTURE OF A MEMBRANE PROTEIN AGGREGATE REVEALS A MECHANISM OF RECOGNITION BY ITS CHAPERONE

May 10, 2013 The Journal of Biological Chemistry, 288, 13420-13430.

Thang X. Nguyen, Peera Jaru-Ampornpan, Vinh Q. Lam, Peigen Cao, Samantha Piskiewicz, Sonja Hess and Shu-ou Shan

Division of Chemistry and Chemical Engineering and the Proteome Exploration Laboratory, Division of Biology, Beckman Institute, California Institute of Technology, Pasadena, California 91125

Protein aggregation is detrimental to the maintenance of proper protein homeostasis in all cells. To overcome this problem, cells have evolved a network of molecular chaperones to prevent protein aggregation and even reverse existing protein aggregates. The most extensively studied disaggregase systems are ATP-driven macromolecular machines. Recently, we reported an alternative disaggregase system in which the 38-kDa subunit of chloroplast signal recognition particle (cpSRP43) efficiently reverses the aggregation of its substrates, the light-harvesting chlorophyll a/b-binding (LHC) proteins, in the absence of external energy input. To understand the molecular mechanism of this novel activity, here we used biophysical and biochemical methods to characterize the structure and nature of LHC protein aggregates. We show that LHC proteins form micellar, disc-shaped aggregates that are kinetically stable and detergent-resistant. Despite the nonamyloid nature, the LHC aggregates have a defined global organization, displaying the chaperone recognition motif on its solvent-accessible surface. These findings suggest an attractive mechanism for recognition of the LHC aggregate by cpSRP43 and provide important constraints

Mechanism of an ATP-independent Protein Disaggregase

II. DISTINCT MOLECULAR INTERACTIONS DRIVE MULTIPLE STEPS DURING AGGREGATE DISASSEMBLY

May 10, 2013 The Journal of Biological Chemistry, 288, 13431-13445.

Peera Jaru-Ampornpan, Fu-Cheng Liang, Alex Nisthal, Thang X. Nguyen, Pengcheng Wang, Kuang Shen, Steven L. Mayo and Shu-ou Shan

Division of Chemistry and Chemical Engineering and the Proteome Exploration Laboratory, Division of Biology, Beckman Institute, California Institute of Technology, Pasadena, California 91125

The ability of molecular chaperones to overcome the misfolding and aggregation of proteins is essential for the maintenance of proper protein homeostasis in all cells. Thus far, the best studied disaggregase systems are the Clp/Hsp100 family of “ATPases associated with various cellular activities” (AAA⁺) ATPases, which use mechanical forces powered by ATP hydrolysis to remodel protein aggregates. An alternative system to disassemble large protein aggregates is provided by the 38-kDa subunit of the chloroplast signal recognition particle (cpSRP43), which uses binding energy with its substrate proteins to drive disaggregation. The mechanism of this novel chaperone remains unclear. Here, molecular genetics and structure-activity analyses show that the action of cpSRP43 can be dissected into two steps with distinct molecular requirements: (i) initial recognition, during which cpSRP43 binds specifically to a recognition motif displayed on the surface of the aggregate; and (ii) aggregate remodeling, during which highly adaptable binding interactions of cpSRP43 with hydrophobic transmembrane domains of the substrate protein compete with the packing interactions within the aggregate. This establishes a useful framework to understand the molecular mechanism by which binding interactions from a molecular chaperone can be used to overcome