Cre-ACEMBLER Vers. 2.0
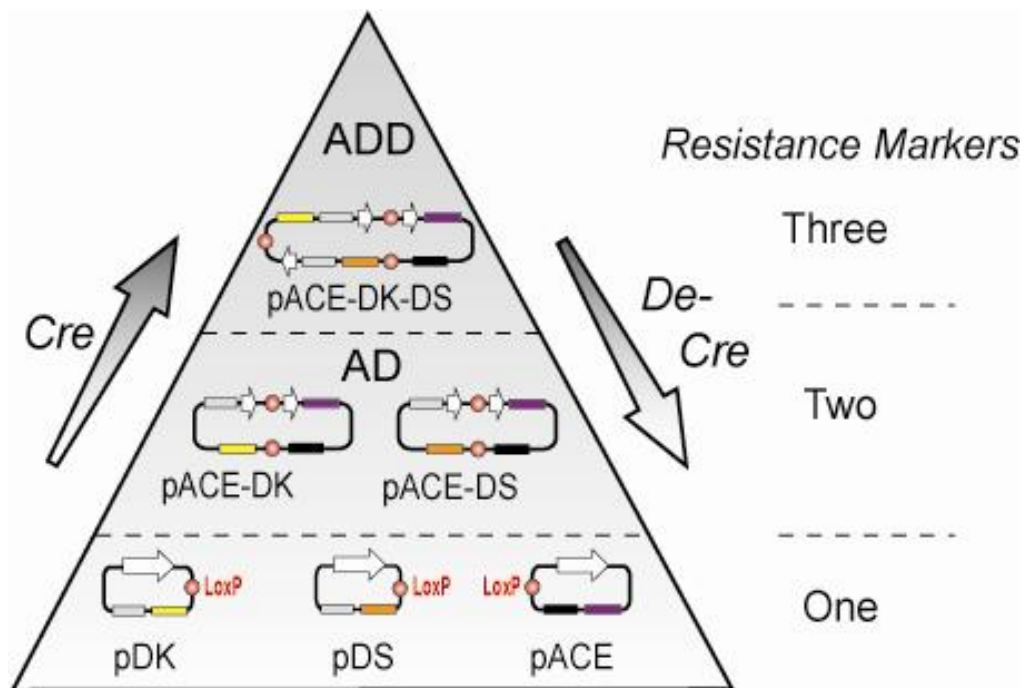
This application carries out *in silico* Cre-recombination of Donors and Acceptors to generate multigene expression plasmids (Donor-Acceoptor fusions). Likewise, a Cre-excision can be carried out of a fusion plasmid to re-generate the educts.

Inupt files can have FASTA or GenBank format.

Schematic of Cre-fusion and De-Cre reaction (excision) of

multigene fusion plasmids by Cre-ACEMBLER [1,2]

[1] Bieniossek, C., Nie, Y., Frey, D., Olieric, N., Schaffitzel, C., Collinson, I., Romier, C., Berger, P., Richmond, T.J., Steinmetz, M.O. & Berger, I. Nat Methods 6, 447-450. (2009)

[2] Nie, Y., Bieniossek, C., Frey, D., Olieric, N., Schaffitzel, C., Steinmetz, M.O. & Berger, I. Nat Protocols, doi: 10.1038/nprot.2009.104 (2009)
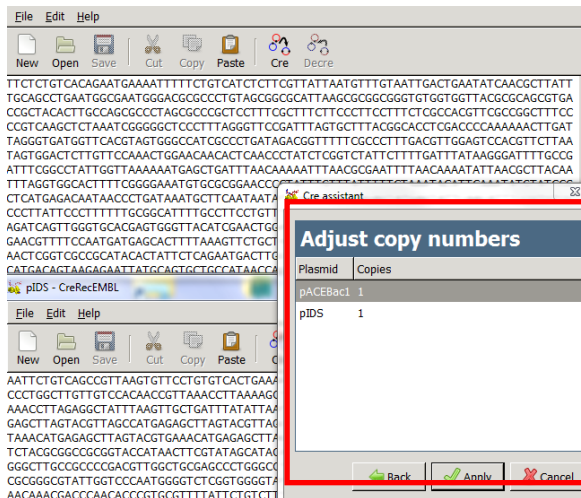
1. Cre-recombination by Cre-ACEMBLER

1.) Open all Acceptor and Donor plasmid sequence files (prepared for example in VectorNTI) which are to be recombined *in silico*. (It is important to specify the file format e.g. FASTA or GenBank by selection from the drop down menu)
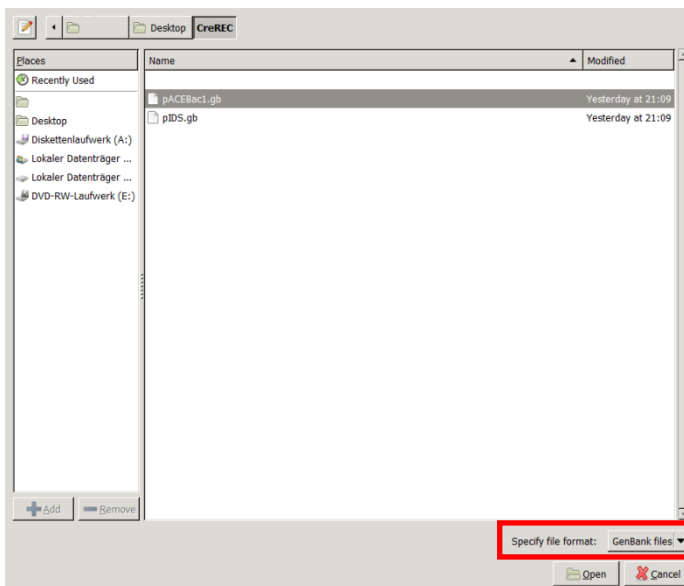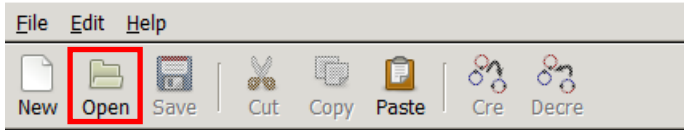
2.) Press the "Cre" button and specify the Acceptor respectively Donor plasmids.
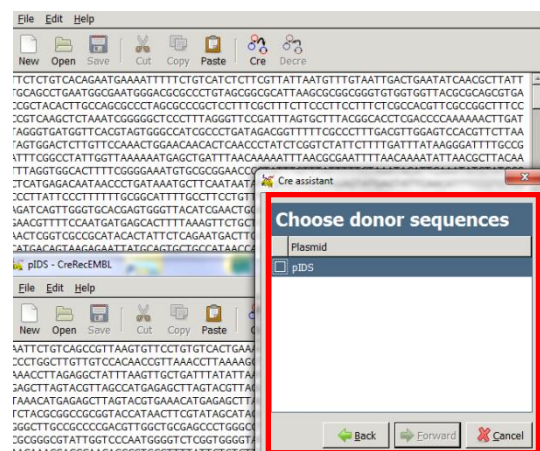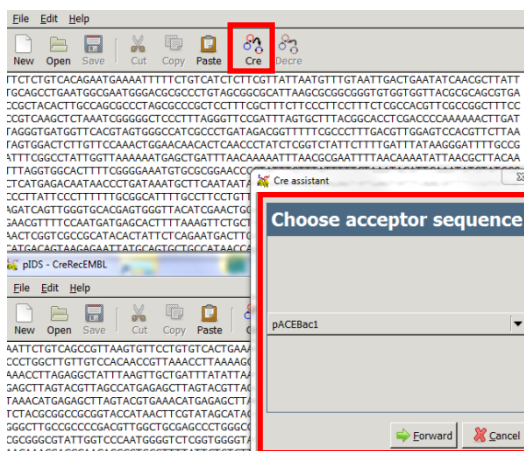
3.) Enter the number of copies of each type of plasmid to be fused. Note that Cre-recombination of multiple Acceptor and Donor plasmids results in numerous variants of fusion plasmids. These variants will open as a new window.

4.) Save the *in silico* Cre-recombined multigene Acceptor-Donor fusion sequences as FASTA or GenBank files.

De-Cre reaction (decatenation) by Cre-ACEMBLER

1.) Open a multigene fusion plasmid sequence (Acceptor-Donor fusions) file *in silico*. (It is important to specify the file format e.g. FASTA or GenBank by selection from the drop down menu)

2.) Press the "Decre" button.
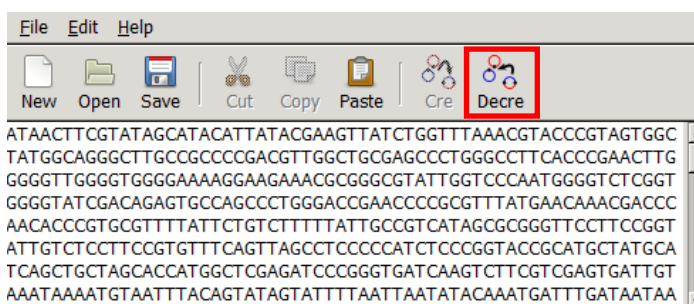
3.) Sequence files containing the Acceptor and Donor plasmid sequences will be opened in new windows.

4.) Save the *in silico* Cre-decatenated Acceptor/Donor plasmid sequence files as FASTA or GenBank files.