Supplementary data

Functional interplay of DnaE polymerase, DnaG primase and DnaC helicase within a ternary complex, and primase to polymerase hand-off during lagging strand DNA replication in *Bacillus subtilis*

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Running title: *Functional insights into a ternary replication subcomplex*

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Key words; Lagging strand replication; protein-protein interactions; replisome; *Bacillus subtilis*

Table S1: Yeast two-hybrid matrix interaction assays [a]

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		BD- fusions	replicative DNA helicase								DNA polymerase III (alpha subunit)																	primase						helicase loading		DNA polymer		delta subunit, DNA PolIII			delta' subunit,		DNA polymerase III											SSB		BD-
			DnaC								DnaE																	DnaG						DnaI		DnaN	DnaX	HolA			HolB		PolC											SSB		-
			full 1-454				1-170		151-454		full 1-1115	1-272		1-826		248-946		286-1115		524-946			618-1115			922-1115		full 1-603	1-224	107-367	340-446	340-603	430-603	full 1-311		full 1-378	full 1-563	full 1-347			full 1-329		1-406		1-898		1-993	324-797		435-1437	792-1437			full 1-172		-
AD- fusions		autoactivation [3AT]	A12 [5]	A4 [5]	A1 [50]	A1 [50]	A2 H4 [50]	A1 H4 [50]	N	N	N	N	N	N	N	[5]	[3]	N	N	[10]	A8 [5]	A10 [10]	A5 [15]	A3 [15]	A3 [15]	[5]	[1.5]	[3]	N	[3]	N	N	N	N	N	N	N	A2 H10 [50]	A4 H10 [50]	A10 H10 [50]	N	N	N	N	N	N	N	N	N	N	[5]	[5]	A5 [10]	A1 H1 [50]	A1 H1 [50]	N
replicative DNA helicase	DnaC	full 1-454	A6 H4	A6 H4	A1 H1	A1 H1	-	-	A1 H1	A1 H2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A8 H2	-	-	-	A2 H2	A1 H1	A2 H2	A2 H2	-	A3 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			A6 H4	A6 H4	A1 H1	A1 H1	-	-	A1 H1	A1 H2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A8 H2	-	-	-	A2 H2	A1 H1	A2 H2	A2 H2	-	A3 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		1-170	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
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		85-234 ….	A3 H3	A1 H3	A1 H2	H2	-	-	A3 H2	A2 H2	-	-	-	-	-	-	-	-	-	-	-	-	A2 H13	A3 H19	A3 ? H5	-	-	-	-	-	-	-	-	A8 H2	A8 H2	-	A3 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			A3 H3	A1 H3	A1 H2	H2	-	-	A3 H2	A2 H2	-	-	-	-	-	-	-	-	-	-	-	-	A2 H13	A3 H15	A3 ? H5	-	-	-	-	-	-	-	-	A8 H2	A8 H2	-	A3 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		174-367 ….	A12 H3	H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A8 H2	A8 H2	-		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			A12 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A8 H2	A8 H2	-		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		151-454	A6 H4	-	A1 H1	A1 H1	-	-	A1 H1	A2 H2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A8 H2	A8 H2	-	A3 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			A6 H4	-	A1 H1	A1 H1	-	-	A1 H1	A2 H2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A8 H2	A8 H2	-	A3 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
DNA polymerase III (alpha subunit)	DnaE	full 1-1115	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H11	A13 H4	-	H10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
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		1-272	-	-	-	-	-	-	-	-	-	-	-	-	-	A2 H2	A1 H1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H17	-	-	-	-	-	-	-	-	-	-	-	-	-	-
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		1-826	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A19	-	-	A12	A3	A6	A4	A4	A3	-	-	-	-	-	-	-	-	-	-	-	-	A2	A2	A7	-	-	-	-	-	-	-	-	-	-	-	-	A5	-	-	-
		248-946	-	-	-	-	-	-	-	-	-	A21 H2	A19 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H4	-	H15	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
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		618-1115	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H11	A13 H4	-	H10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
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		804-988 ….	A12	-	-	-	-	-	-	-	-	-	-	A13 H2	A15 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H15	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A12	-	-	-	-	-
		805-990 ….	A12	-	-	-	-	-	-	-	-	-	-	A13 H2	A19 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A12	-	-	-	-	-
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DNA primase	DnaG	full 1-603	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
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		340-603	-	-	H4	H4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			-	-	H4	H4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		430-603	-	-	H4	H4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			-	-	H4	H4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
helicase loading	DnaI	full 1-311	A2 H2	A1 H1	A1 H1	A1 H1	-	-	A1 H1	A1 H1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			A2 H2	A1 H1	A1 H1	A1 H1	-	-	A1 H1	A1 H1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
DNA polymerase III (beta subunit)	DnaN	full 1-378	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H32	-	-	A6 H14	A11 H7	-	-	-	-	-	-	-	-	-	-	A3 H2	A3 H2	A3 H3	-	-	-
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DNA polymerase III (gamma and tau	DnaX	full 1-563	A2 H2	A2 H2	A1 H1	A1 H1	-	-	A2 H1	A2 H2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H4	-	-	-	H4	H9	-	-	-	A25	A1 H2	A1 H1	A2 H2	-	-	H3	H2	H6	H6	H4	-	-	-	-	-	-	-	-	-
			A2 H2	A2 H2	A1 H1	A1 H1	-	-	A2 H1	A2 H2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H4	-	-	-	H4	H9	-	-	-	A25	A1 H2	A1 H1	A2 H2	-	-	H3	H2	H6	H6	H4	-	-	-	-	-	-	-	-	-
DNA polymerase III (delta subunit)	HolA	full 1-347	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H7	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
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DNA polymerase III (delta' subunit)	HolB	full 1-329	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
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DNA polymerase III	PolC	1-406	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
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		435-1437	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		792-1437	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	H25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		875-1437	-	-	-	-	-	-	-	-	-	-	-	-	-	A18	11 H11	-	-	-	-	-	A2 H6	A3 H15	A3 ? H5	-	-	-	-	-	-	-	-	-	-	A2 H3	-	A2 H6	A3 H6	A4 H7	-	-	-	-	-	-	-	-	-	-	A12	A7	-	-	-	-
			-	-	-	-	-	-	-	-	-	-	-	-	-	A18	11 H11	-	-	-	-	-	A2 H6	A3 H15	A3 ? H1	-	-	-	-	-	-	-	-	-	-	A2 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A12	A7	-	-	-	-
		1052-1437	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A19	-	-	-	-	-	A4 H13	A3 H15	A3 ? H1	-	-	-	-	-	-	-	-	-	-	A3 H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
single-strand DNA-binding protein	SSB	full 1-172	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A2 H4	A2 H3	A3 H3	H2	H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A1	-
			-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A2 H4	A2 H3	A3 H3	H2	H3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	A1	-
AD-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

[a] The proteins (full size or fragment derived thereof) fused to the binding (first lines) or activating (first column) domain of Gal4 are indicated. The fusions constructed here are indicated in red (details are available upon request). Remaining fusions were from the P. Noirot's collection (16 in the main text). AD- and BD- indicate empty pGBDU and pGAD vectors used to detect self-activation and as negative controls for interaction. The coordinates of the fragments cloned are indicated (in amino acids relative to the protein sequence). The line "autoactivation [3AT]" indicate behavior of pGBDU derived diploids onto plates selecting for the expression of the ADE2 and HIS3 interaction reporters: N, absence of autoactivation on either plates; Ax, autoactivation onto ADE selecting plates observed from day x; Hy, autoactivation onto HIS (or HIS + 3-aminotriazole) selecting plates observed from day y; [z]; concentration of 3-aminotriazole added to HIS selecting plates to reduce or eliminate autoactivation observed on this medium. Results are indicated as follow: -, no interaction; Ax Hy, interaction observed on ADE or HIS selecting plates at day x and y, respectively. Blue cells highlight interactions previously described (16 in the main text). Green ones indicate new interactions identified in this work. Red highlights previously described interactions not seen in this study. Yellow indicates inconsistent data and grey autoactivation or interaction presumably masked by early autoactivation.

Table S2. RNA, DNA, and RNA:DNA standards used in this study with the 5'-d(CTA)-containing 23-mer template 5'-d(CAGACACACACACACTACACACA)-C3. Ribonucleotides are italicized and underlined and the initial dinucleotide synthesized by DnaG primase is in bold.

Oligonucleotide Standard	Length (bp)	Sequence (5' to 3')
RNA primer	16-mer	***AG****UGUGUGUGUGUCUG*
RNA primer	14-mer	***AG****UGUGUGUGUGUC*
RNA primer	12-mer	***AG****UGUGUGUGUG*
RNA primer	10-mer	***AG****UGUGUGUG*
RNA primer	7-mer	***AG****UGUGU*
RNA primer	5-mer	***AG****UGU*
DNA primer	16-mer	AGTGTGTGTGTGTCTG
DNA primer	15-mer	GTGTGTGTGTGTCTG
DNA primer	14-mer	TGTGTGTGTGTCTG
DNA primer	13-mer	GTGTGTGTGTCTG
DNA primer	12-mer	TGTGTGTGTCTG
DNA primer	11-mer	GTGTGTGTCTG
DNA primer	10-mer	TGTGTGTCTG
DNA primer	6-mer	TGTCTG
RNA:DNA hybrid primer	16-mer	***AG***TGTGTGTGTGTCTG
RNA:DNA hybrid primer	16-mer	***AG****UG*TGTGTGTGTCTG

Table S3. Molecular modelling metrics

SWISS-MODEL Workspace

Workunit: P000002 Title:DnaG

Model Summary:

Model information:

Modelled residue range:	112 to 426
Based on template:	3b39A (2.35 A)
	Remark: No search for template was performed. Only user specified template was used for modelling.
Sequence Identity [%]:	32.075
Evalue:	0

Quaternary structure information:

Template (3b39): MONOMER
Model built: SINGLE CHAIN

Ligand information:

Ligands in the template: DG: 1.
Ligands in the model: none.

Quality information:

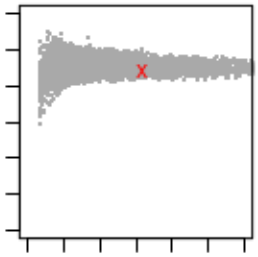
QMEAN Z-Score: -1.02

Global Model Quality Estimation:

QMEAN4 global scores:

QMEANscore4: 0.715

Estimated absolute model quality:

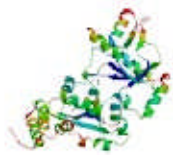


Z-Score: -1.02

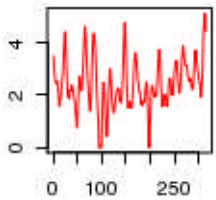
Score components:

Local scores:

Coloring by residue error:



Residue error plot:



QMEAN4 global scores:

The QMEAN4 score is a composite score consisting of a linear combination of 4 statistical potential terms (estimated model reliability between 0-1). The pseudo-energies of the contributing terms are given below together with their Z-scores with respect to scores obtained for high-resolution experimental structures of similar size solved by X-ray crystallography:

Scoring function term	Raw score	Z-score
C_beta interaction energy	-129.83	0.10
All-atom pairwise energy	-9529.60	0.21
Solvation energy	-45.93	1.38
Torsion angle energy	-49.56	-1.96
QMEAN4 score	0.715	-1.02

If you publish results from QMEAN, please cite the following paper:
Benkert P, Biasini M, Schwede T. (2011). "Toward the estimation of the absolute quality of individual protein structure models." Bioinformatics, 27(3):343-50.

Local Model Quality Estimation:

Alignment:

```
TARGET    1          SGEQKM AEAHELLKKF YHHLLINTKE GQEALDYLLS RGFTKELINE
3b39A     109    na--hqrqtl yqlmdglntf yqqslq-qpv atsarqylek rglsheviar

TARGET               hhhhh hhhhhhhhhh hhhh        hhhhhhhhh h   hhhhhh
3b39A                hhhhhh hhhhhhhhhh hhhhh       hhhhhhhhh     hhhhhh


TARGET    47     FQIGYALDSW DFITKFLVKR GFSEAQMEKA GLLIRREDGS GYFDRFRNRV
3b39A     156    faigfappgw dnvlkrfggn penrqslida gmlvtndrs- --ydrfrerv

TARGET             ssss     hhhhh     hhhhhhhhh   sss         sss  sss
3b39A           h  ssss     hhhhh     hhhhhhhhh   sssss ss    sss  sss


TARGET    97     MFPIHDHHGA VVAFSGRALG SQQPKYMNSP ETPLFHKSKL LYNFYKARLH
3b39A     205    mfpirdkrgr vigfggrvlg ndtpkylnsp etdifhkgrq lyglyeaqqd

TARGET           ssssss   s ssssssss       sssss               hhhhhhh
3b39A            ssssss   s ssssssss       sssss                hhh   h


TARGET    147    IRKQERAVLF EGFADVYTAV SSDVKESIAT MGTSLTDDHV KILRRNVEEI
3b39A     255    naepnrllvv egymdvvala qyginyavas lgtsttadhi qllfratnnv

TARGET                 ssss    hhhhhhh      sss         hhhh hhhhhh sss
3b39A                  ssss   hhhhhhhh      sss         hhhh hhhhhh sss


TARGET    197    ILCYDSDKAG YEATLKASEL L---QKKGCK VRVAMIPDGL DPDDYIKKFG
3b39A     305    iccydgdrag rdaawcalet alpymtdgrq lrfmflpdge dpdtlvrkeg

TARGET           ssssss hhh hhhhhhhhhh          s ssssss      hhh
3b39A            ssssss hhh hhhhhhhhhh hhh      s ssssss      hhh


TARGET    244    GEKFKNDIID ASVTVMAFKM QYFRKGKNLS DEGDRLAYIK DVLKEISTLS
3b39A     355    keafea-rme qamplsaflf nslmpqvdls tpdgrarlst lalplisqvp

TARGET           hhhhh        ssshhhhh hhhhh       hhhhhhhhh hhhhhhh
3b39A            hhhhhh hhh   ssshhhhh hhhhh       hhhhhhhhh hhhhhhh


TARGET    294    GSLEQEVYVK QLASEFSLSQ ES -
3b39A     404    getlriylrq elgnklgild dsql

TARGET            hhhhhhhhh hhhhhh
3b39A             hhhhhhhhh hhhhhh
```

Modeling Log:

```
3.70 (SP3)
Loading Template: 3b39A.pdb
Loading Raw Sequence
Loading Alignment: ./NXXX.align.submit.fasta
Removing HET groups from template structure
Refining Raw Sequence Alignment
```

```
ProModII: doing simple assignment of backbone
ProModII: adding blocking groups
Adding Missing Sidechains
AddPolar H
BuildDeletetedLoopsModel
connectivity problem (C-N > 3.0A) at residue: 85
Trying Ligating with anchor residues ARG 82 and GLY 85
connectivity problem --> including residue GLY 86
Trying Ligating with anchor residues ARG 82 and SER 86
Trying Ligating with anchor residues ARG 82 and GLY 87
Trying Ligating with anchor residues ARG 82 and TYR 88
Trying Ligating with anchor residues ARG 82 and PHE 89
Number of Ligations found: 500
ACCEPTING loop  344: clash=   0 FF=       -213.9 PP= -2.00
Trying Ligating with anchor residues LEU 217 and LYS 220
Trying Ligating with anchor residues LEU 217 and GLY 221
Trying Ligating with anchor residues LEU 217 and CYS 222
Number of Ligations found: 500
ACCEPTING loop  144: clash=   0 FF=       -207.2 PP= -2.00
Building CSP loop with anchor residues ILE 22 and LYS 25
Number of Ligations found: 8
ACCEPTING loop    0: clash=   0 FF=       -225.0 PP= -2.00
Building CSP loop with anchor residues ASN 249 and ILE 252
Number of Ligations found: 3
ACCEPTING loop    2: clash=   0 FF=        869.8 PP=  0.00
Optimizing Sidechains
Adding Hydrogens
Optimizing loops and OXT (nb = 19)
Final Total Energy:       -7260.481 KJ/mol
Dumping Sequence Alignment
```

Template Selection Log:

```
 - Start SMR-Pipeline in automated mode on BC2-cluster at Mon Dec  3 16:26:20 2012


 - User specified template structure by a PDB identifier(3b39A), entering user template mode

 - Aligning sequence of the user template structure with the target sequence using BLAST
 - Alignment quality between target and specified template is too low

 - Aligning sequence of the user template structure with the target sequence using HHSearch
 - Send 1 target-template alignments for modeling

 - @@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@@
 -               ******************************************
 - building model based on 3b39A (112-426) was successfull
 - Workspace Pipeline parameter

   Cut-off parameters to model the target based on a BLAST target-template alignment
    Evalue :                                     0.0001
    Minimum Template size (aa) for ranking :     25
    Minimum Sequence identity :                  60

   Cut-off parameters to model the target based on a HHSearch target-template alignment
    Evalue :                                     0.0001
    Probability :                                50
    MAC :                                        0.3

   Parameters for model selection
    Minimal number of uncovered target
     residues after BLAST to run HHSEARCH :      50
    Minimal number of uncovered target
     residues to model an additional template :  25

 - Finish SMR-Pipeline in automated mode on BC2-cluster at Mon Dec  3 17:16:48 2012
```

Quaternary Structure Annotation of the Template
3b39 is annotated as MONOMER
The oligomeric state of the structure was assigned by the authors of the corresponding PDB entry
The following biological unit was used to build the template structure: 3b39.pdb1.gz

Quaternary Structure Modelling of the Target Protein
The target and template sequences are too diverse (seqid: 32.075) to infer a conservation of the oligomeric state
Please use the advancend features of the SwissModel Project Mode

The final model was calculated as single chain

Ligand Modeling Log: Template's ligands section
Ligands in the template: DG: 1.
The template contains ligands that are not yet part of the pipeline. Ligands which are currently assessed are listed in the help page.
No ligands were included in the model.

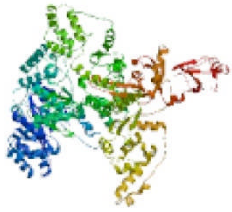
References: If you publish results using SWISS-MODEL, please cite the following papers:

- Arnold K., Bordoli L., Kopp J., and Schwede T. (2006). The SWISS-MODEL Workspace: A web-based environment for protein structure homology modeling. Bioinformatics, 22,195-201.
- Schwede T, Kopp J, Guex N, and Peitsch MC (2003) SWISS-MODEL: an automated protein homology-modeling server. Nucleic Acids Research 31: 3381-3385.
- Guex, N. and Peitsch, M. C. (1997) SWISS-MODEL and the Swiss-PdbViewer: An environment for comparative protein modeling. Electrophoresis 18: 2714-2723.

SWISS-MODEL Workspace

Workunit: P000029 Title: DnaE

Model Summary:



Model information:

Modelled residue range: 1 to 1111

Based on template:

Quaternary structure information:

Quality information:

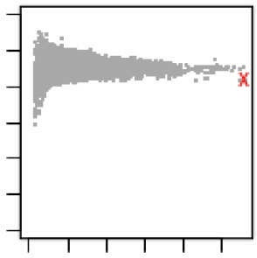
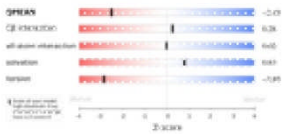
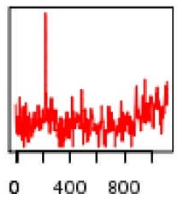
QMEAN Z-Score: -2.493

Ligand information:

Global Model Quality Estimation:

QMEAN4 global scores:

Local scores:

QMEANscore4:	Estimated absolute model quality:	Score components:	Coloring by residue error:	Residue error plot:
0.612				

Z-Score: -2.493

QMEAN4 global scores:

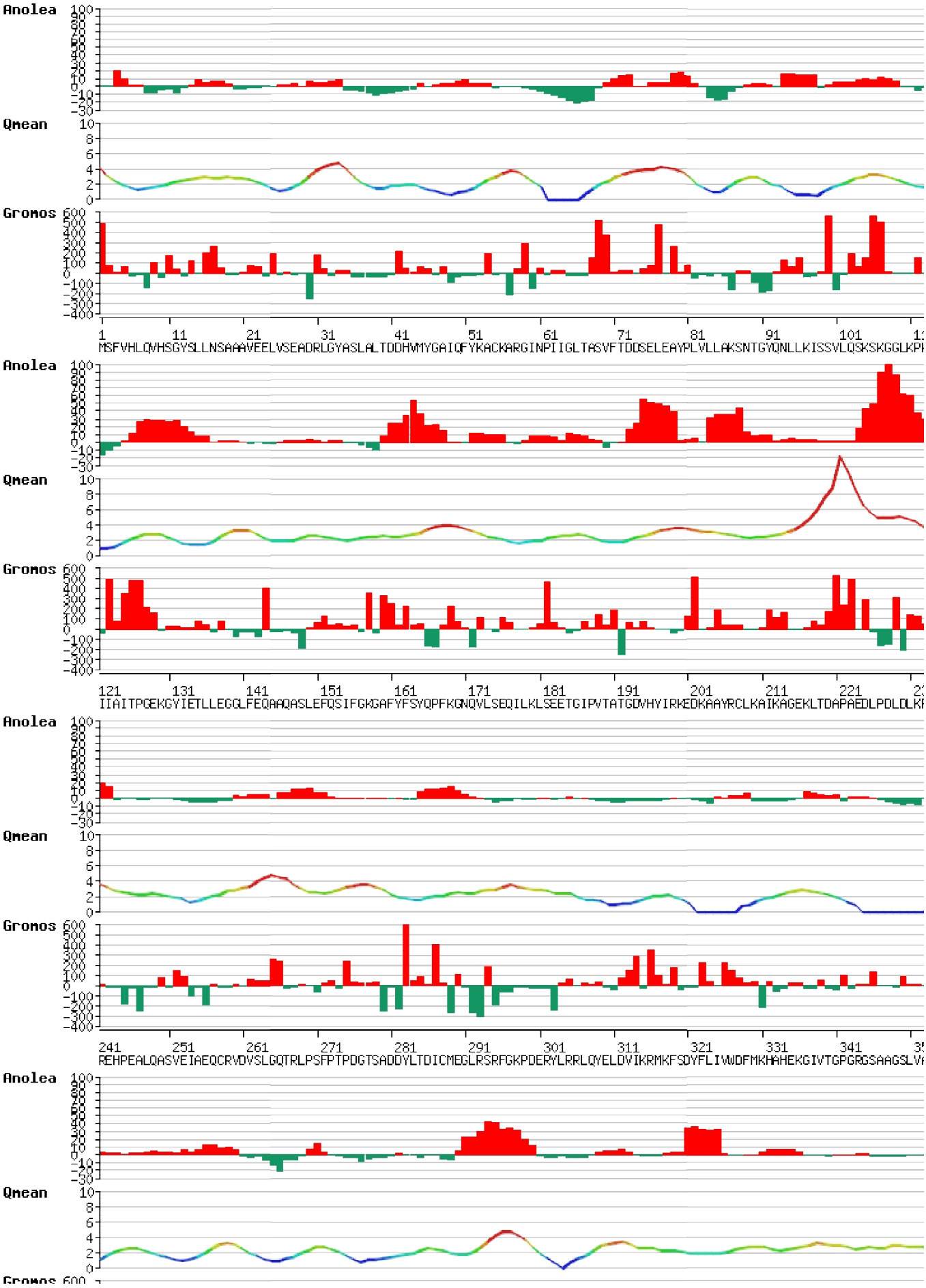
The QMEAN4 score is a composite score consisting of a linear combination of 4 statistical potential terms (estimated model reliability between 0-1). The pseudo-energies of the contributing terms are given below together with their Z-scores with respect to scores obtained for high-resolution experimental structures of similar size solved by X-ray crystallography:

Scoring function term	Raw score	Z-score
C_beta interaction energy	-443.11	0.28
All-atom pairwise energy	-29032.21	0.02
Solvation energy	-128.21	0.83
Torsion angle energy	-105.42	-2.85
QMEAN4 score	0.612	-2.49

If you publish results from QMEAN, please cite the following paper:

Benkert P, Biasini M, Schwede T. (2011). "Toward the estimation of the absolute quality of individual protein structure models." Bioinformatics, 27(3):343-50.

Local Model Quality Estimation:


Anolea
Qmean
Gromos
1 11 21 31 41 51 61 71 81 91 101
MSFVHLQVHSGYSLLNSAAAVEELVSEADRLGYASLALTDDHMYGAIQFYKACKARGINPIIGLTASVFTDDSELEAYPLVLLAKSNTGYQNLLKISSVLQSKSKGGLKP
Anolea
Qmean
Gromos
121 131 141 151 161 171 181 191 201 211 221
IIAITPGEKGYIETLLEGGLFEQAAQASLEFQSIFGKGAFYFSYQPFKGNQVLSEQILKLSEETGIPVTATGDVHYIRKEDKAAYRCLKAIKAGEKLTDAPAEDLPDLDLKP
Anolea
Qmean
Gromos
241 251 261 271 281 291 301 311 321 331 341
REHPEALQASVEIAEQCRVDVSLGQTRLPSFPTPDGTSADDYLTDICMEGLRSRFGKPDERYLRRLQYELDVIKRMKFSDYFLIVWDFMKHAHEKGIVTGPGRGSAAGSLVA
Anolea
Qmean
Gromos

Alignment:

```
TARGET    1       MSFVHLQV  HSGYSLLNSA  AAVEELVSEA  DRLGY--ASL  ALTDDHVMYG
3e0dA     5       lkfahlhq  htqfsllnga  aklqdllkwv  kettpedpal  amtdhgnlfg

TARGET                                  hhhhhhhh  hhh      s  ssss
3e0dA                                   hhhhhhhh  hhh    sss  ssss    h


TARGET    47    AIQFYKACKA  RGINPIIGLT  ASVFTDDSEL  E--------A  YPLVLLAKSN
3e0dA     53    avefykkata  mgvkpiigye  ayvaaesrfd  rkrgg----y  fhltllakdf
                                                        ****
TARGET           hhhhhhh      sssssss s                     ssssss h
3e0dA           hhhhhhhh      sssssss sssss           s    s  sssssss h


TARGET    89    TGYQNLLKIS SVLQSKSK--  -GGLKPKWLH  SYREGIIAIT  PGEKGYIETL
3e0dA     104   tgyqnlvrla sraylegfye  kpridreilr  ehaqglials  gclgaeipqf

TARGET           hhhhhhhhhh hhhhh           hhhhh  h    ssssss      hhhhh
3e0dA            hhhhhhhhhh hhhhh  sss sss hhhhh  h    ssssss      hhhhh


TARGET    136   LEGGLFEQAA QASLEFQSIF  GKGAFYFSYQ  P-FKGNQVLS  EQILKLSEET
3e0dA     154   ilqdrldlae arlnedlsif  gdrffieiqn  hglpeqkkvn  qvlkefarky

TARGET           h   hhhhhh hhhhhhhhhh    ssssss      hhhhhh  hhhhhhhhhh
3e0dA            hh    hhhhh hhhhhhhhhh    ssssss      hhhhhh  hhhhhhhhh


TARGET    185   GIPVTATGDV HYIRKEDKAA  YRCLKAIKAG  EKLTDAPAED  LPD--LDLKP
3e0dA     204   glgmvatnng hyvrkedara  hevllaiqsk  ttlddperwr  fpcdefyvkt

TARGET              sss            hh  hhh    hhh
3e0dA             sssss            hh  hhhhhhhhh


TARGET    233   LEEMQNIYRE HPE---ALQA  SVEIAEQCRV  DVSL---GQT  RLPSFPTPDG
3e0dA     254   peemramlpe aewgdepfdn  tveiarmcdv  dlpigdkmvy  riprfplg--

TARGET           hhhhhhh             h  hhhhhh
3e0dA            hhhhhhh         hhhhh  hhhhhh


TARGET    277   TSADDYLTDI  CMEGLRSRFG  KP--------  ----------  ----------
3e0dA     304   rteaqylrel  tflgllrryp  driteafyre  vlrllderal  aealarveek

TARGET             hhhhhhhh  hhhhhhhh
3e0dA              hhhhhhhh  hhhhhhhh       hhhhhh  hhh   hhh  hhhhh   hh


TARGET    299   ----------  -DERYLRRLQ  YELDVIKRMK  FSDYFLIVWD  FMKHAHEKGI
3e0dA     361   aweelrkrew  taeailhral  yelsviermg  fpgyflivqd  yinwarghgv

TARGET                        hhhhhhhh  hhhhhhhh    hhhhhhhhh  hhhhhhh
3e0dA            h           hhhhhhhhh  hhhhhhhh    hhhhhhhhh  hhhhhhh


TARGET    338   VTGPGRGSAA GSLVAYVLYI  TDVDPIKHHL  LFERFLNPER  VSMPDIDIDF
3e0dA     419   svgpgrgsaa gslvayavgi  tnidplrfgl  lferflnper  vsmpdidtdf
```

```
TARGET          sss         hhhhhhh               hhh             sss
3e0dA           sss         hhhhhhh               hhh             sss


TARGET     388  PDTRRDEVIQ  YVQQKYGAMH VAQIITFGTL  AAKAALRDVG RVFGVSPKEA
3e0dA      469  sdrerdrviq  yvrerygedk vaqigtfgsl  askaalkdva rvygiphkka

TARGET          sh   hhhhh  hhhhhh   s ss      sss  hhhhhhhhhh hh      hhh
3e0dA           shhhhhhhhh  hhhhhh   s ss  sss sss  hhhhhhhhhh hh      hhh


TARGET     438  DQLAKLIPS-  -RPGMTLDEA RQQSPQLDKR  LRESSLLQQV YSIARKIEGL
3e0dA      519  eelaklipvq  fgkpkplqea feaepelrae  mekderirqv ievamrlegl

TARGET          hhhhh                           hh    hhhhh hhhhhh   s
3e0dA           hhhhh                           hh    hhhhh hhhhhhh  s


TARGET     486  PRHASTHAAG  VVLSEEPLTD VVPLQEGHEG  IYLTQYAMDH LEDLGLLKMD
3e0dA      569  nrhasvhaag  vviaaepltd lvplmrdqeg  rpvtqydmga vealgllkmd

TARGET          ss      ss  ssss        sssss    sssss   hhh hhh   ssss
3e0dA           ss   sss  ss  ssss        sssss    sssss   hhh hhh   ssss


TARGET     536  FLGLRNLTLI  ESITSMIEKE ENIKIDLSSI  SYSDDKTFSL LSKGDTTGIF
3e0dA      619  flglrtltfl  dearrivkes kgveldydrl  plddpktfel lsrgetkgvf

TARGET          ssss  hhhhh  hhhhhhhhhh                 hhhhh hhh
3e0dA           ssss  hhhhh  hhhhhhhhhh                 hhhhh hhh


TARGET     586  QLESAGMRSV  LKRLKPSGLE DIVAVNALYR  PGPMENIPLF IDRKHGRAPV
3e0dA      669  qlesggmtat  vrglkprrle diialvslyr  pgpmehipty irrhhgqepv

TARGET              hhhhhh  hhhh     h hhhhhhhh       hhhhhhh hhhh
3e0dA               hhhhhh  hhhh     h hhhhhhhh     h    hhh hhhh


TARGET     636  HY---PHED-  -LRSILEDTY GVIVYQEQIM  MIASRMAGFS LGEADLLRRA
3e0dA      719  syaefphaek  ylrpildety gipvyqeqim  qiasqvagys lgeadllrra

TARGET                      h             hhhh hhhhhhh    hhhhhhhhhh
3e0dA               hhhhh  hhhh             hhhh hhhhhh     hhhhhhhhhh


TARGET     681  VSKKKKEILD  RERSHFVEGC LKKEYSVDTA  NEVYDLIVKF ANYGFNRSHA
3e0dA      769  mgkkrveemq  khrerfvrga kergvpeeea  nrlfdmleaf anygfnksha

TARGET          hhh     hh  hhhhhhhhhh hhh     hhh  hhhhhhhhhh h      hhhh
3e0dA           hhh     hh  hhhhhhhhhh hhh     hhh  hhhhhhhhhh h      hhhh


TARGET     731  VAYSMIGCQL  AYLKAHYPLY FMCGLLTSVI  GNEDKISQYL YEAKGSGIRI
3e0dA      819  aaysllsyqt  ayvkahypve fmaallsver  hdsdkvaeyi rdaralgipv

TARGET          hhhhhhhhhh  hhhhh   hh hhhhhhh       hhhhhhhh hhhh    ss
3e0dA           hhhhhhhhhh  hhhhh   hh hhhhhhh       hhhhhhhh hhhh    ss


TARGET     781  LPPSVNKSSF  PFTVENGSVR YSLRAIKSVG  VSAVKDIYKA -RKEKPFEDL
3e0dA      869  lppdvnrsgf  dfkvvgeeil fglsavknvg  emaarailee rerggpfksl
```

```
TARGET          s            sss  sss  s           hhhhhhhh          h
3e0dA           s            sss  sss  s          hhhhhhhhh hhh      h


TARGET    830   FDFCFRVPSK SVNRKMLEAL  IFSGAMDEFG QNRATLLASI DVALEHAELF
3e0dA     919   gdflkrlpeq vvnkralesl  vkagaldafg -drarllasl epllrwaaet

TARGET          hhhhhh       hhhhhhh  hh             hhhhhhh hhhhhhhhhh
3e0dA           hhhhhh       hhhhhhh  hh            hhhhhhhh hhhhhhhhhh


TARGET    880   AAD--DDQMG  LFLDESFSIK  PKYVETEELP LVDLLAFEKE TLGIYFSNHP
3e0dA     968   rergrsglvg  lfaeve---e  pplveaspld eitmlryeke algiyvsghp

TARGET          hh                                hhhhhhhhhh h
3e0dA           hhhh                              hhhhhhhhhh h


TARGET    928   LSAFR----K  QLTAQGAVSI  LQAQRAVKRQ LSLGVLLSKI KTIRTKTGQN
3e0dA     1015  vlrypglrev  asctieelse  fvrelpgkpk vllsgmveev rf--------

TARGET                          hhhh  hh      ss sssssssss ss
3e0dA                hhhhh      hhhh  hh      ss ssssssss    s


TARGET    974   MAFLTLSDET GEMEAVVFPE  QFRQLSPVLR EGALLFTAGK  CEVRQDKIQF
3e0dA     1069  ----tlsdet galevvk---  ---------- edipllvlae  verv-----l

TARGET          sssssssss   sss  hhhhhh  hhh          ssssss  s       s
3e0dA               ssss   sssss                     ssssss  s       s


TARGET    1024  IMSRAELLED MDAEKAPSVY  IKIESSQHSQ EILAKIKRIL  LEHKGETGVY
3e0dA     1115  aqavwtleev leapka--le  vevdhallde kga-rlksll  dehpgslpvy

TARGET          sssss   hhh        sss  sss            hhhhh  h  sss  ss
3e0dA           sssss   hh    sss  ss  sss           hhhhhh  h  sss sss


TARGET    1074  LYYERQKQ-T IKLPESFHIN  ADHQVLY--- -RLKELLGQK  NVV
3e0dA     1163  lrvlgpfgea lfalrevrvg  eealglleae gyraylvpdr  evf-

TARGET          sss       s ssss   sss   hhhh         sss
3e0dA           sssss   sss ssss   sss   hhhhhh      sssssss
```

Modeling Log:

```
3.70  (SP3)
Loading Template: 3e0dA.pdb
Loading Raw Sequence
Loading Alignment: ./user.align.submit.fasta.FF
Removing  HET  groups  from  template  structure
Refining Raw Sequence Alignment
ProModII:  doing  simple  assignment  of  backbone
C-terminal overhang trimmed for chain ' '. End at residue: 1111
ProModII: adding blocking groups
Adding            Missing
Sidechains AddPolar H
```

```
BuildDeletetedLoopsModel
Trying Ligating with anchor residues GLY 32 and SER 35
Number of Ligations found: 16
ACCEPTING loop 2: clash= 0 FF= -53.9 PP= 0.00
Trying Ligating with anchor residues LEU 76 and TYR 79
Trying Ligating with anchor residues GLU 75 and TYR 79
Trying Ligating with anchor residues SER 74 and TYR 79
Number of Ligations found: 16
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues ASP 73 and TYR 79
Number of Ligations found: 63
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues ASP 72 and TYR 79
Number of Ligations found: 210
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues ASP 72 and PRO 80
Number of Ligations found: 500
ACCEPTING loop 326: clash= 0 FF= -19.6 PP= 1.00
Trying Ligating with anchor residues SER 105 and GLY 108
Number of Ligations found: 11
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues LYS 104 and GLY 108
Number of Ligations found: 50
ACCEPTING loop 0: clash= 0 FF= 839.1 PP= -3.00
Small Ligation (C-N <3.0A) ignored;
GROMOS will repair it at residue PHE 168
Trying Ligating with anchor residues PRO 226 and ASP 229
Trying Ligating with anchor residues LEU 225 and ASP 229
Number of Ligations found: 30
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues LEU 225 and LEU 230
Number of Ligations found: 80
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues ASP 224 and LEU 230
Number of Ligations found: 500
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues ASP 224 and LYS 231
Number of Ligations found: 500
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues GLU 223 and LYS 231
Number of Ligations found: 500
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues GLU 223 and PRO 232
Number of Ligations found: 500
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues ALA 222 and PRO 232
Number of Ligations found: 500
all loops are bad; continuing CSP with larger segment
+++ Warning: Ligation Failed, SparePart will be inserted later
+++          It is usually the sign that the region is misaligned.
Trying Ligating with anchor residues PRO 244 and LEU 247
Trying Ligating with anchor residues HIS 243 and LEU 247
Trying Ligating with anchor residues GLU 242 and LEU 247
Number of Ligations found: 88
ACCEPTING loop 74: clash= 0 FF= 474.7 PP= -2.00
Trying Ligating with anchor residues SER 262 and GLN 265
Number of Ligations found: 15
ACCEPTING loop 14: clash= 0 FF= 450.0 PP= 0.00
connectivity problem (C-N > 3.0A) at residue: 275
Trying Ligating with anchor residues PRO 272 and ASP 275
Trying Ligating with anchor residues PRO 272 and GLY 276
Trying Ligating with anchor residues PRO 272 and THR 277
Number of Ligations found: 500
ACCEPTING loop 188: clash= 0 FF= -52.9 PP= -2.00
Trying Ligating with anchor residues LYS 297 and GLU 300
Trying Ligating with anchor residues GLY 296 and GLU 300
Trying Ligating with anchor residues PHE 295 and GLU 300
Number of Ligations found: 287
```

```
ACCEPTING loop   24: clash=   0  FF=        267.3 PP=  -2.00
Trying Ligating with anchor residues PRO  445  and PRO  448
Number of Ligations found: 5
ACCEPTING loop    1: clash=   0  FF=         20.3 PP=  0.00
connectivity problem (C-N >3.0A) at residue:  494
Trying Ligating with anchor residues THR  491  and ALA  494
Number of Ligations found: 10
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues SER  490  and ALA  494
Number of Ligations found: 65
ACCEPTING loop   37: clash=   0  FF=       -160.4 PP=  -1.00
Trying Ligating with anchor residues HIS  636  and HIS  639
Trying Ligating with anchor residues VAL  635  and HIS  639
Trying Ligating with anchor residues VAL  635  and GLU  640
Number of Ligations found: 422
ACCEPTING loop   54: clash=   0  FF=        134.2 PP=  0.00
Trying Ligating with anchor residues GLU  640  and ARG  643
Number of Ligations found: 7
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues HIS  639  and ARG  643
Trying Ligating with anchor residues PRO  638  and ARG  643
Number of Ligations found: 362
ACCEPTING loop  282: clash=   0  FF=       -105.6 PP=  0.00
Trying Ligating with anchor residues ALA  820  and GLU  823
Trying Ligating with anchor residues ALA  820  and LYS  824
Number of Ligations found: 100
ACCEPTING loop   99: clash=   0  FF=       -379.6 PP=  -2.00
Trying Ligating with anchor residues ASP  882  and GLN  885
Number of Ligations found: 10
ACCEPTING loop    0: clash=   0  FF=       1880.1 PP=  -4.00
Trying Ligating with anchor residues PHE  931  and GLN  934
Trying Ligating with anchor residues ALA  930  and GLN  934
Trying Ligating with anchor residues ALA  930  and LEU  935
Trying Ligating with anchor residues SER  929  and LEU  935
Number of Ligations found: 180
ACCEPTING loop   60: clash=   0  FF=       -139.2 PP=  -2.00
connectivity problem (C-N >3.0A) at residue:  965
Trying Ligating with anchor residues LYS  962  and THR  965
connectivity problem --> including residue THR  966
Trying Ligating with anchor residues LYS  962  and ILE  966
Trying Ligating with anchor residues LYS  962  and ARG  967
Trying Ligating with anchor residues LYS  962  and THR  968
Trying Ligating with anchor residues LYS  962  and LYS  969
Trying Ligating with anchor residues LYS  962  and THR  970
Trying Ligating with anchor residues LYS  962  and GLY  971
Trying Ligating with anchor residues LYS  962  and GLN  972
+++ Warning: Ligation Failed, SparePart will be inserted later
+++          It is usually the sign that the region is misaligned.
connectivity problem (C-N > 3.0A) at residue: 991
Trying Ligating with anchor residues ALA 988 and PHE 991
Trying Ligating with anchor residues ALA 988 and PRO 992
Trying Ligating with anchor residues ALA 988 and GLU 993
Trying Ligating with anchor residues ALA 988 and GLN 994
Trying Ligating with anchor residues ALA 988 and PHE 995
Trying Ligating with anchor residues ALA 988 and ARG 996
Trying Ligating with anchor residues ALA 988 and GLN 997
Trying Ligating with anchor residues ALA 988 and LEU 998
+++ Warning: Ligation Failed, SparePart will be inserted later
+++          It is usually the sign that the region is misaligned.
connectivity problem (C-N > 3.0A) at residue: 1017
Trying Ligating with anchor residues GLU 1015 and GLN 1018
Trying Ligating with anchor residues GLU 1015 and ASP 1019
Trying Ligating with anchor residues GLU 1015 and LYS 1020
Trying Ligating with anchor residues GLU 1015 and ILE 1021
Trying Ligating with anchor residues GLU 1015 and GLN 1022
Trying Ligating with anchor residues GLU 1015 and PHE 1023
Number of Ligations found: 500
ACCEPTING loop  424: clash=     0  FF=       1384.0  PP=  -3.00
```

```
connectivity problem (C-N >3.0A) at residue: 1057
Trying Ligating with anchor residues GLU 1054 and ALA 1057
Trying Ligating with anchor residues GLU 1054 and LYS 1058
Number of Ligations found: 1
ACCEPTING loop 0: clash= 0 FF= 65.0 PP= -2.00
Trying Ligating with anchor residues GLN 1079 and THR 1082
Trying Ligating with anchor residues ARG 1078 and THR 1082
Number of Ligations found: 2
all loops are bad; continuing CSP with larger segment
Trying Ligating with anchor residues GLU 1077 and THR 1082
Number of Ligations found: 61
ACCEPTING loop 50: clash= 0 FF= 75.5 PP= 0.00
Trying Ligating with anchor residues LEU 1098 and LEU 1101
Trying Ligating with anchor residues VAL 1097 and LEU 1101
Trying Ligating with anchor residues VAL 1097 and LYS 1102
Number of Ligations found: 130
ACCEPTING loop 58: clash= 0 FF= 1607.8 PP= 1.00
Building CSP loop with anchor residues GLY 859 and ARG 862
Number of Ligations found: 4
ACCEPTING loop 2: clash= 0 FF= -155.6 PP= -1.00
Building CSP loop with anchor residues SER 893 and LYS 897
Building CSP loop with anchor residues GLU 892 and LYS 897
Number of Ligations found: 36
ACCEPTING loop 26: clash= 0 FF= 1153.3 PP= 0.00
Building CSP loop with anchor residues ALA 1039 and VAL 1042
Building CSP loop with anchor residues LYS 1038 and VAL 1042
Building CSP loop with anchor residues LYS 1038 and TYR 1043
Number of Ligations found: 46
all loops are bad; continuing CSP with larger segment
Building CSP loop with anchor residues GLU 1037 and TYR 1043
Number of Ligations found: 81
ACCEPTING loop 25: clash= 0 FF= 856.6 PP= 2.00
Finding Spare-Part loop with anchor residues ALA 220 and LYS 231
all loops are bad; continuing scan with larger segment
Finding Spare-Part loop with anchor residues ASP 219 and LYS 231
ACCEPTING loop 4 from 2BBKH Clash= 4 FF= 505.1 PP=2027.12
BadPhi= 2 BadGX= 0 BadXP= 1 weakXP= 1 Score=8.75 rms= 0.00
Finding Spare-Part loop with anchor residues SER 961 and THR 978
ACCEPTING loop 39 from 3DFR_ Clash= 4 FF= 255.3 PP=2009.60
BadPhi= 0 BadGX= 0 BadXP= 0 weakXP= 0 Score=6.00 rms= 0.00
Finding Spare-Part loop with anchor residues GLU 987 and GLU 1004
ACCEPTING loop 81 from 3SDHA Clash= 7 FF= 219.8 PP=2107.56
BadPhi= 0 BadGX= 0 BadXP= 0 weakXP= 0 Score=10.50 rms= 0.00
Optimizing Sidechains
Adding Hydrogens
Optimizing loops and OXT (nb = 123)
Final Total Energy: 87564.758 KJ/mol
Dumping Sequence Alignment
```

Template Selection Log:

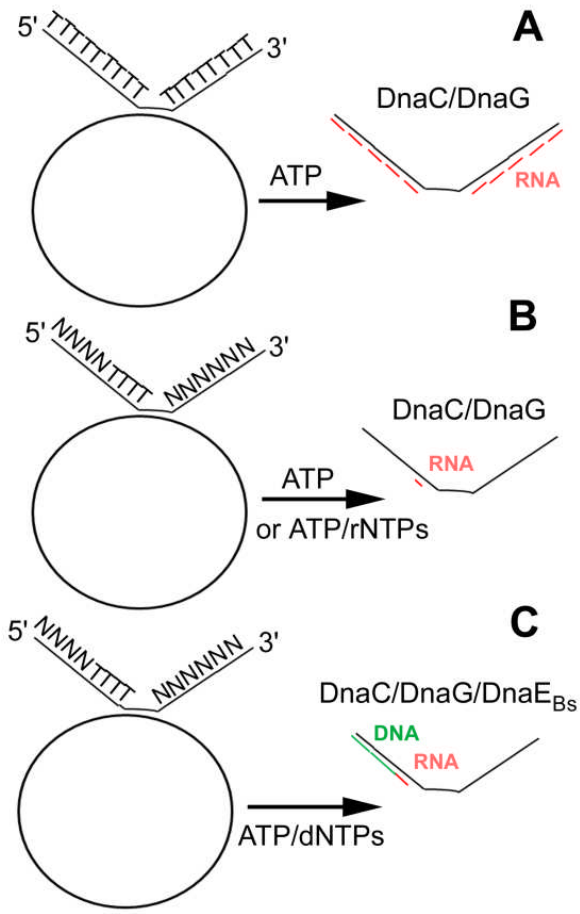
Ligand Modeling Log:

References: If you publish results using SWISS-MODEL, please cite the following papers:

- Arnold K., Bordoli L., Kopp J., and Schwede T. (2006). The SWISS-MODEL Workspace: A web-based environment for protein structure homology modeling. Bioinformatics, 22,195-201.

- Schwede T, Kopp J, Guex N, and Peitsch MC (2003) SWISS-MODEL: an automated protein homology-modeling server. Nucleic Acids Research 31: 3381-3385.
- Guex, N. and Peitsch, M. C. (1997) SWISS-MODEL and the Swiss-PdbViewer: An environment for comparative protein modeling. Electrophoresis 18: 2714-2723.

Supplementary Figure 1S



Supplementary Figure 1s. Coupled helicase-primase and helicase-primase-polymerase assays.

A. A schematic diagram explaining the basis of the coupled helicase-primase assay using a forked DNA substrate with polyT tails. Since 5'-d(TTT) is a starting prime site for the B. subtilis DnaG multiple RNA primers (shown in red) will be formed along both 5'- and 3'-tails and the displaced oligonucleotide will be shifted in non-denaturing polyacrylamide gels. ATP will provide the energy fuel for the translocating helicase and the substrate for the primase to synthesize poly(A) RNA primers.

B. The same assay was carried out with a forked DNA substrate with random sequences in the tails but possessing a single 5'-d(TTTT) site along the 5'-tail (see EXPERIMENTAL PROCEDURES in the main text). In the presence of ATP or ATP+rNTPs only a small amount of small RNA primers will be synthesized which are not sufficient to cause a shift of the displaced oligonucleotide.

C. A schematic diagram explaining the basis of the coupled helicase-primase-polymerase assay The same assay as that described in panel B was carried out in the presence of ATP+dNTPs and $DnaE_{Bs}$. This time a clear shift of the displaced oligonucleotide was observed as the small RNA primers were extended further by $DnaE_{Bs}$ to produce larger RNA-DNA hybrids (Red-Green) that annealed onto the displaced oligonucleotide.