2. MATERIALS

2.1 Chemicals, enzymes and kits

[γ-^{32}P]ATP	Amersham-Pharmacia
[^{35}S]Cysteine	Amersham-Pharmacia
β-Mercaptoethanol	Merck
Acrylamide	Serva
Agarose, ultra pure	Serva
Amicon Membranes	Millipore
Ammonium persulfate	Merck
Ampicillin	Boehringer-Mannheim
Bacto-Agar	Difco
Bacto-Tryptone	Difco
Benzamidine	Sigma
Bromophenol blue	Sigma
Cellulose Dialysis Membrane	Spectrum
Centriprep Devices	Millipore
Chloramphenicol	Serva
Comassie Brilliant Blue R-250	Sigma
Deoxyribonucleotides (dNTPs)	Boehringer-Mannheim
Dithioerythriol (DTE)	ICN
Edeine	Calbiochem
Ethidium bromide	Merck
Expand Long Template PCR System	Boehringer-Mannheim
Isopropyl-β-D-thiogalactopyranoside (IPTG)	Sigma
Jet Start Plasmid Kit	Genomed
L-Cysteine	Sigma
Lysozyme	Boehringer-Mannheim
N,N,N´,N´-Tetramethylethylenamide (TEMED)	Bio-Rad
NZCYM Broth	Fluka
Pfu DNA-polymerase	Stratagene
Phenol	Roth
Phenylmethylsulfonylfluoride (PMSF)	Boehringer-Mannheim
Proteinase K	Boehringer-Mannheim
PVDF Membranes	Millipore
Qiaquick Gel Extraction Kit	Qiagen
Qiaquick Nucleotide Removal Kit	Qiagen
Qiaquick PCR Purification Kit	Qiagen
Rapid Ligation Kit	Boehringer-Mannheim
Restriction endonucleases	New England Biolabs
RNase A	Boehringer-Mannheim
Sodiumdodecylsulfate (SDS)	Bio-Rad
Spermidine-hydrochloride	Serva

T4-Polynucleotide kinase	MBI Fermentas
Tetracycline	Serva
Tetrakis-acetoxy-mercuri-methane (TAMM)	Strem Chemicals
Yeast extract	Difco

All other chemicals and solution, if not differently specified, were from MERCK.

2.2 Bacterial strains

Escherichia coli:

XL1 Blue	*RecA1 endA1 gyrA96 thi-1 hsdR17 supE44 relA1 lac* [F' proAB *lacI*^{q}ZΔM15 Tn10 (Tet^r)
BL21(DE3)pLysS	*HsdS gal* F$^-$ *ompT* $r_B^-m_B^-$ (λcIts857 *ind*1 Sam7 *nin*5 *lac*UV5-T7 gene1)

Thermus thermophilus:

HB8

2.3 Plasmid

pET-11a	Novagen

2.4 Media

LB-Medium	10 g/l Bacto-Tryptone, 5 g/l Yeast extract, 10 g/l NaCl, pH 7.5
M9ZB-Medium	23 g/l NZCYM Broth, 1 g/l NH_4Cl, 3 g/l KH_2PO_4, 6 g/l $Na_2HPO_4 \cdot 2H_2O$, 4 g/l Glucose, pH 7,0

The media were prepared with bidestilled water and sterilised by autoclaving for 20 min at 120°C and 1.3 bar. Plates were prepared adding to the medium 15 g/l Bacto-Agar. Antibiotics were added after sterilisation.

2.5 Oligodeoxyribonucleotides

Oligonucleotides were synthesised by TIB Molbiol (Berlin).

2.6 Purification columns

Bio-Spin Chromatography Columns	Bio-Rad
DEAE Fast Flow	Pharmacia
NAP Column (Sephadex G-25)	Pharmacia
Phenyl Superose HR 5/5	Pharmacia
Resource S, 6ml	Pharmacia