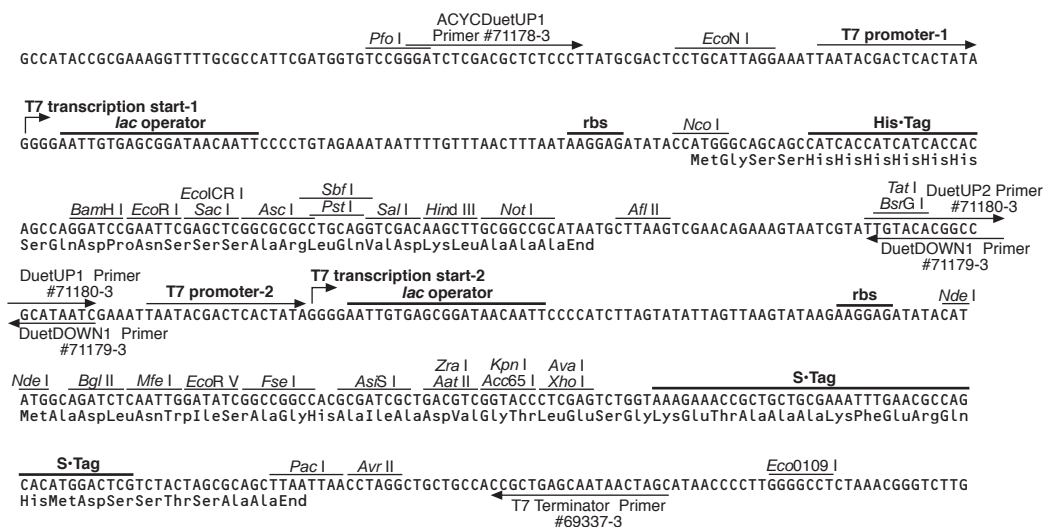
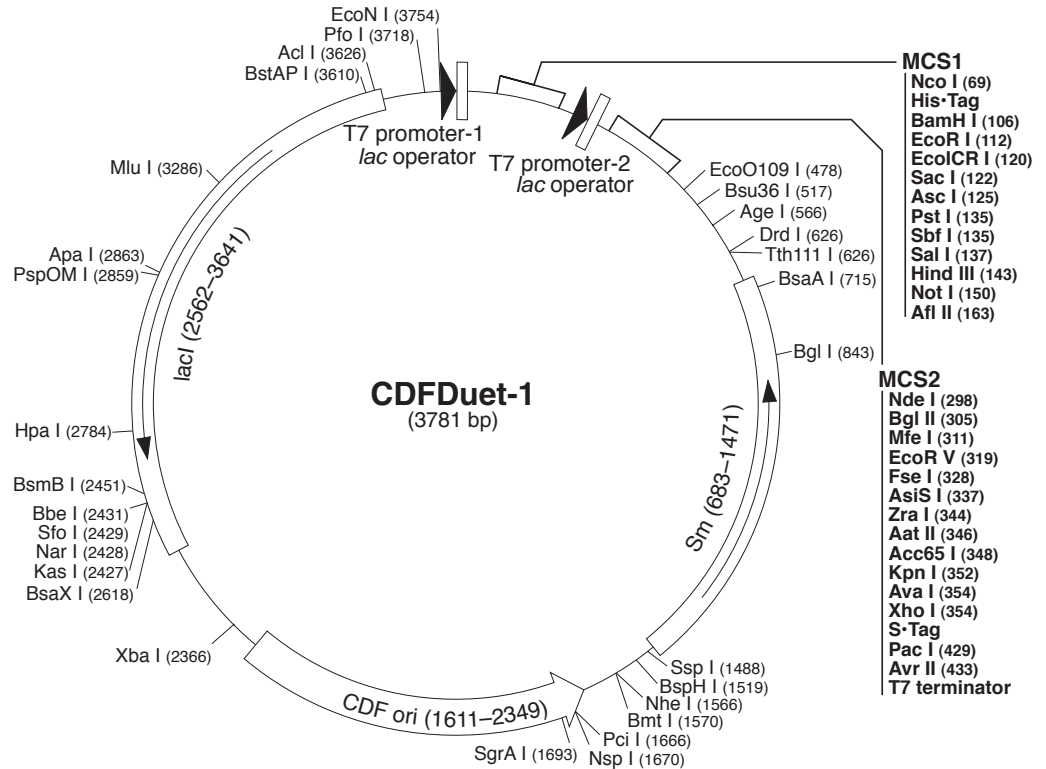


# pCDFDuet-1 Vector

	Cat. No.
pCDFDuet-1 DNA	71340-3
<b>pCDFDuet-1 sequence landmarks</b>	
T7 promoter-1	3765-3781
T7 transcription start-1	1
His•Tag® coding sequence	83-100
Multiple cloning sites-1	
( <i>Nco</i> I- <i>Afl</i> II)	69-168
T7 promoter-2	214-230
T7 transcription start-2	231
Multiple cloning sites-2	
( <i>Nde</i> I- <i>Avr</i> II)	297-438
S•Tag™ coding sequence	366-410
T7 terminator	462-509
<i>aadA</i> (SmR) coding sequence	683-1471
CDF origin	1611-2349
<i>lacI</i> coding sequence	2562-3641

pCDFDuet™-1 is designed for the coexpression of two target ORFs. The vector contains two multiple cloning sites (MCS), each of which is preceded by a T7lac promoter and ribosome binding site (rbs). The vector also carries the CloDF13-derived CDF replicon, *lacI* gene and streptomycin/spectinomycin resistance gene (Sm<sup>R</sup>). This vector can be used in combination with pACYCDuet™-1 (Cat. No. 71147-3), pRSFDuet™-1 (Cat. No. 71341-3), and pETDuet™-1 (Cat. No. 71146-3) in an appropriate host strain for the coexpression of 4 to 8 target proteins. ORFs inserted into MCS1 can be sequenced using the ACYCDuetUP1 Primer (Cat. No. 71178-3) and DuetDOWN1 Primer (Cat. No. 71179-3). ORFs inserted into MCS2 can be sequenced using the DuetUP2 Primer (Cat. No. 71180-3) and T7 Terminator Primer (Cat. No. 69337-3). Unique sites are shown on the circle map.



## pCDFDuet-1 cloning/expression regions

# pCDFDuet-1 Restriction Sites

Enzyme	# Sites	Locations	Enzyme	# Sites	Locations
AatII	1	346	EciI	4	1010 1706 1893 3501
Acc65I	1	348	Eco57I	2	1328 2426
AccI	2	138 411	Eco57MI	5	1222 1328 2426 2965 3454
AcII	1	3626	EcoICRI	1	120
AfIII	1	163	EcoNI	1	3754
AfIII	2	1666 3286	EcoO109I	1	478
Agel	1	566	EcoRI	1	112
Apal	1	3083	EcoRV	1	319
ApaLI	2	1193 3306	FseI	1	328
AscI	1	125	HaeII	8	830 979 1409 1614 1934
Asel	4	213 2544 2603 3764			2651 2894 3675
AsiSI	1	337	HincII	2	139 2784
AvaI	1	354	HindIII	1	143
AvrII	1	433	HpaI	1	2784
BaeI	2	365 1972	KasI	1	2647
BamHI	1	106	KpnI	1	352
BanI	4	348 2517 2647 3366	MfeI	1	311
BanII	2	122 3083	MluI	1	3286
Bbel	1	2651	MslI	4	1200 2920 2950 3238
BbsI	2	2801 3140	NaeI	2	326 821
BceAI	7	211 1352 1396 1757 1868	NarI	1	2648
		2802 3429	NcoI	1	69
Bcgl	2	162 2966	NdeI	1	298
BciVI	3	1521 1887 2834	NgoMIV	2	324 819
BclI	2	1270 3272	NheI	1	1566
BglI	1	843	NotI	1	150
BglIII	1	305	NspI	1	1670
BlpI	2	451 1160	PacI	1	429
Brne1580I	3	1197 3083 3310	PciI	1	1666
Bmri	7	824 833 929 2118 2488	PfiMI	2	401 3711
		3128 3365	PfoI	1	3718
BmtI	1	1570	PspOMI	1	3079
BpmI	3	1222 2965 3454	PstI	1	135
BpuEI	4	515 2074 2315 2478	PvuI	2	337 1093
BsaAI	1	715	PvuII	2	2597 2690
BsaHI	4	343 1416 2648 3331	SacI	1	122
BsaWI	8	551 566 1002 1754 1890	SalI	1	137
		2038 2464 2967	SbfI	1	135
BsaXI	1	2618	SfiI	5	29 131 226 1213 3777
BseYI	3	1989 2752 2887	SfoI	1	2649
BsgI	2	3241 3441	SgrAI	1	1693
BsiEI	8	153 199 325 337 625	SmII	6	163 354 494 2053 2330
		1093 1580 2507			2493
BsiHKA1	4	122 1197 2209 3310	Sspl	1	1488
BsmAI	6	1521 1592 2671 3058 3184	StyI	5	69 433 473 694 1036
		3589	TaqII	2	1566 2493
BsmBI	1	2671	TatI	1	190
BsmFI	2	912 1673	TspGWI	2	1192 1634
Bsp1286I	5	122 1197 2209 3083 3310	Tth111I	1	626
BspCNI	3	443 530 2707	XbaI	1	2366
BspHI	1	1519	XcmI	4	1643 2901 2919 3435
BspMI	2	124 1121	XhoI	1	354
BsrBI	4	13 243 1525 2364	ZraI	1	344
BsrDI	4	832 1125 2879 3245			
BsrFI	6	324 566 819 872 1693			
		3600			
BsrGI	1	190			
BssHII	3	125 744 2875			
BstAPI	1	3610			
BstEII	2	1307 3104			
BstXI	3	3240 3363 3492			
BstYI	9	106 305 641 987 1827			
		2326 2337 2510 3722			
Bsu36I	1	517			
BtgI	4	69 1175 1645 2226			
BtsI	4	543 1357 2559 2927			
DrallI	2	718 1338			
DrdI	1	626			
EaeI	7	150 196 322 326 636			
		1381 2612			
EagI	3	150 196 322			
EarI	3	1479 2394 3669			

Enzymes that do not cut pCDFDuet-1:				
AarI	AfeI	AhdI	AleI	Alol
AlwNI	BbvCI	BfrBI	BmgBI	BpII
Bpu10I	BsaBI	Bsal	BseRI	BsiWI
BsmI	BspEI	BssSI	BstBI	BstZ17I
Clal	DraI	FalI	FspAI	FspI
MscI	NruI	Nsil	PmeI	PmlI
Ppil	PpuMI	PshAI	Psil	Psrl
RsrII	SacII	SanDI	SapI	Scal
SexAI	SfiI	SmaI	SnaBI	SpeI
SphI	SrfI	StuI	Swal	XmaI
XmnI				