invitrogen

Anza Restriction Enzyme Cloning System

Complete, one-buffer system—
for beautifully simple cloning

ThermoFisher SCIENTIFIC

Cloning has never been simpler

Finally, forget the frustrations of finding compatible buffers and sorting through protocols for your restriction enzyme digests—start getting reliable results for your downstream experiments.

The Invitrogen™ Anza™ Restriction Enzyme Cloning System is a complete system, comprised of:

128 restriction enzymes + 5 DNA modifying enzymes

All Anza™ restriction enzymes work together cohesively and are fully functional with the single Anza™ buffer.

The system offers:

- One buffer for all restriction enzymes
- One digestion protocol for all DNA types
- Complete digestion in 15 minutes
- Overnight digestion without star activity

DNA modifying enzymes

Anza T4 DNA Ligase Master Mix

Anza Thermosensitive Alkaline Phosphatase

Anza T4 Polynucleotide Kinase

Anza DNA Blunt End Kit

Anza DNA End Repair Kit

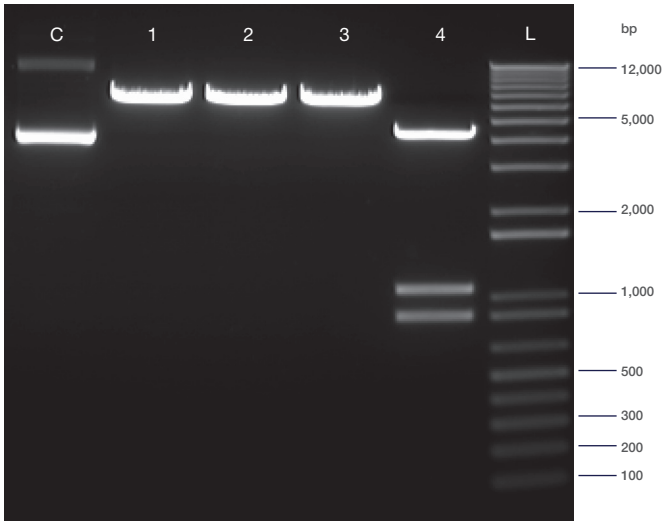
Convenient buffer formats

All Anza restriction enzymes come with an Anza 10X Buffer and an Anza 10X Red Buffer to give you the flexibility you require. The red buffer includes a density reagent containing red and yellow tracking dyes that migrate with 800 bp DNA fragments and faster than the 10 bp DNA fragments, respectively, in a 1% agarose gel. This eliminates tedious dye addition steps prior to gel loading and is compatible with downstream applications.*

*For applications that require analysis by fluorescence excitation, Anza 10X Buffer is recommended, as the Anza 10X Red Buffer may interfere with some fluorescence measurements.

A one-buffer system

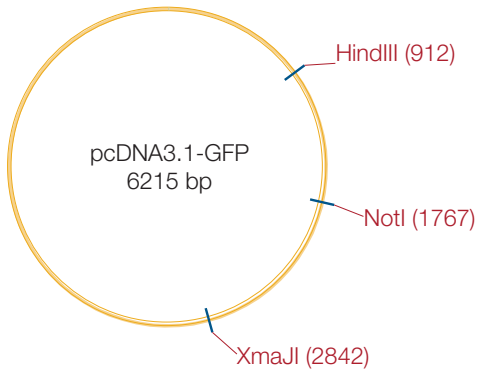
Eliminate the frustration of trying to find compatible buffers for all your enzymes. All Anza restriction enzymes allow for complete digestion using a single Anza buffer. Now you can save time and perform experiments with multiple enzymes in a single reaction without compromising your results.



Triple digestion
C – undigested plasmid DNA
1 – Anza 1 NotI
2 – Anza 16 HindIII
3 – Anza 15 XmaJI
4 – Anza 1 NotI + Anza 16 HindIII + Anza 15 XmaJI
L – 1 Kb Plus DNA Ladder

Figure 1. Anza restriction enzymes show complete digestion with three enzymes in a single buffer. Plasmid DNA (6215 bp) was digested using Anza 1 NotI, Anza 16 HindIII, and Anza 15 XmaJI. For single restriction enzyme digestions, reaction mixture included 1 µg of DNA and 1 µl of restriction enzyme to a total volume of 20 µL. Reaction mixture included 1 µg of DNA and 1 µL of each restriction enzyme to a total volume of 30 µL for triple digestion, as per recommended protocol. Incubation was done at 37°C for 15 minutes.

Mapping of restriction enzyme cut sites



A one-protocol system

Anza restriction enzymes utilize one simple two-step digestion protocol, regardless of the number of restriction enzymes in your reaction or the type of DNA you're working with—just prepare your reaction mixture and incubate at 37°C for 15 minutes.

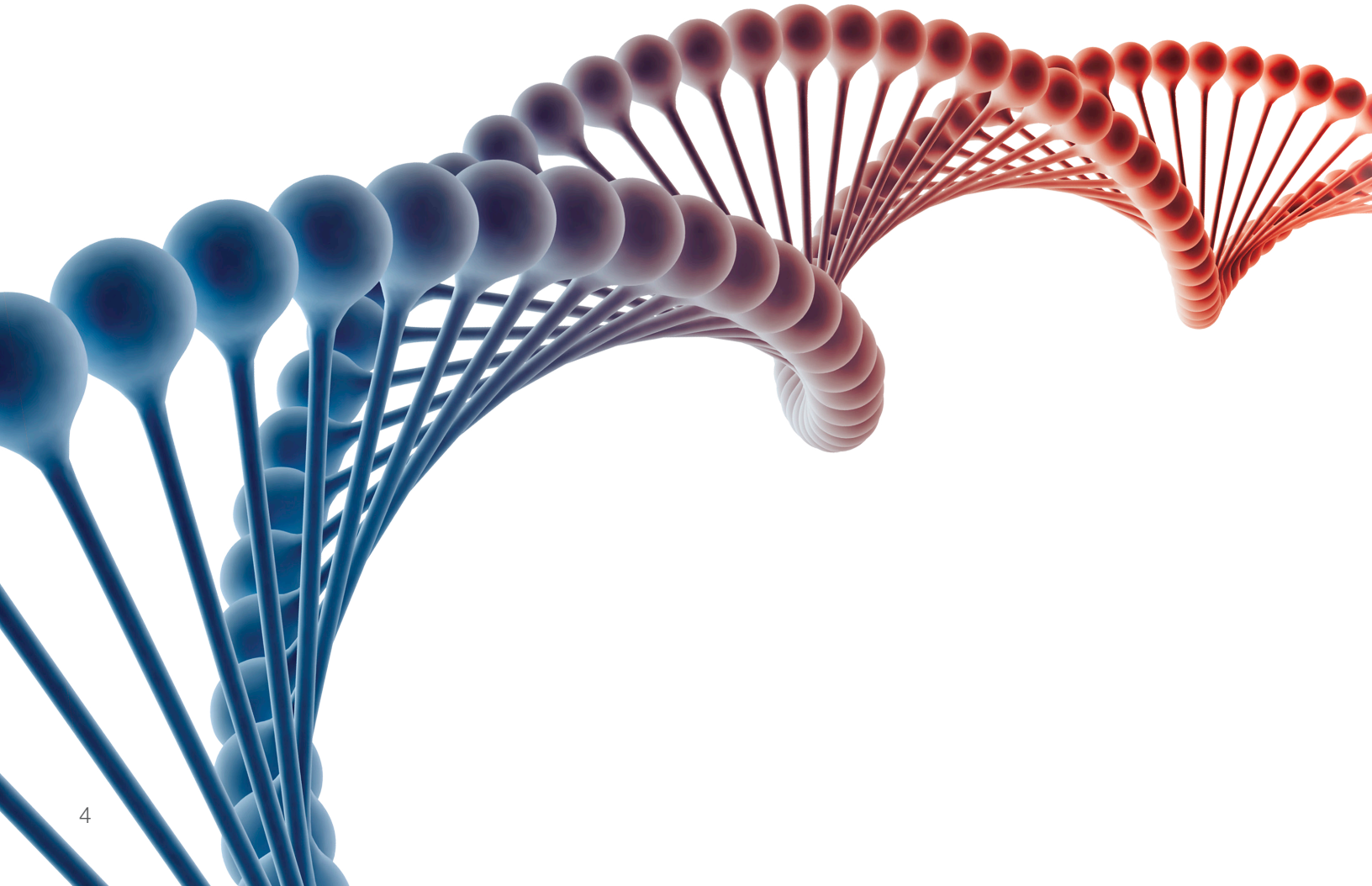
Protocol

1. Prepare a reaction mix by adding reagents in the order indicated in Table 1.
2. Incubate at 37°C for 15 minutes.

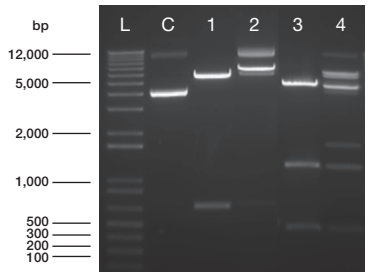
Table 1

Reagent	1-enzyme reaction	2-enzyme reaction	3-enzyme reaction
Nuclease-free water	As required to make up final reaction volume		
Anza 10X Buffer or Anza 10X Red Buffer	2 µL	2 µL	3 µL
DNA	0.2–1 µg	0.2–1 µg	0.2–1 µg
Anza restriction enzyme 1	1 µL	1 µL	1 µL
Anza restriction enzyme 2	—	1 µL	1 µL
Anza restriction enzyme 3	—	—	1 µL
Final reaction volume	20 µL	20 µL	30 µL

Volumes can be scaled up linearly to 5X.

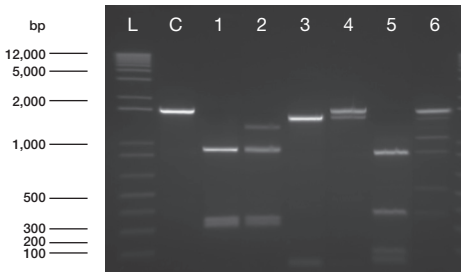


Anza restriction enzymes consistently digest DNA completely in 15 minutes and with varying DNA substrates.



Plasmid DNA

Digestion of plasmid DNA
L – 1 Kb Plus DNA Ladder
C – undigested plasmid DNA
1 – Anza 3 BcuI
2 – SpeI-HF (Competitor N isoschizomer to BcuI)
3 – Anza 47 Eco52I
4 – EagI-HF (Competitor N isoschizomer to Eco52I)

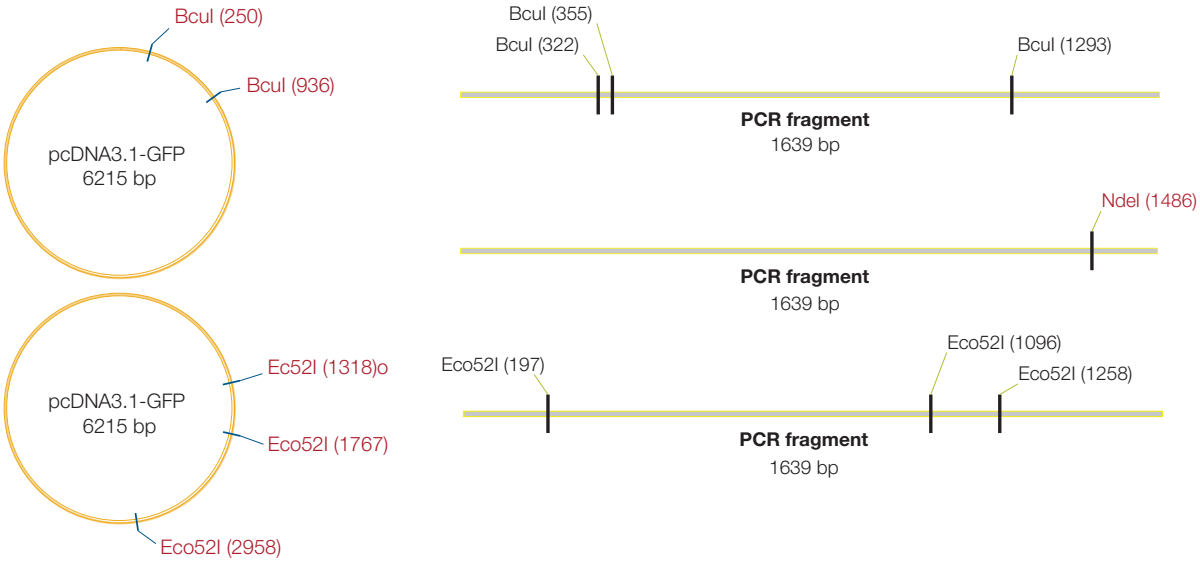


Purified PCR product

Digestion of PCR DNA
L – 1 Kb Plus DNA Ladder
C – undigested DNA
1 – Anza 3 BcuI
2 – SpeI-HF (Competitor N isoschizomer to BcuI)
3 – Anza 9 NdeI
4 – NdeI (Competitor N)
5 – Anza 47 Eco52I
6 – EagI-HF (Competitor N isoschizomer to Eco52I)

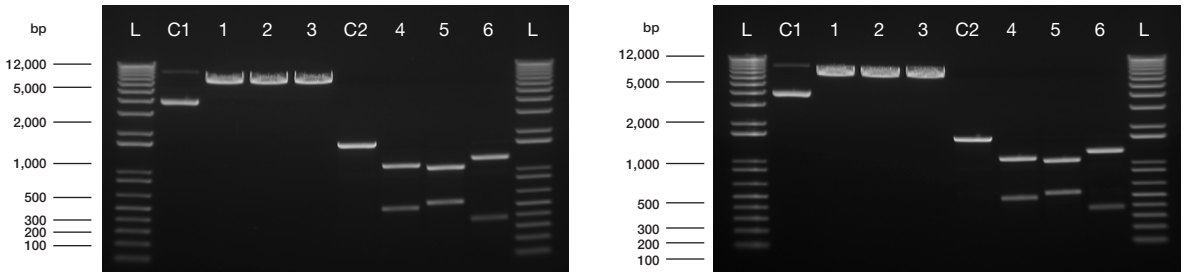
Figure 2. Anza restriction enzymes utilize one digestion protocol to provide superior performance. Plasmid DNA (6,215 bp) was digested using Anza 3 BcuI and Anza 47 Eco521, as well as Competitor N SpeI-HF (isoschizomer to BcuI) and Competitor N EagI-HF (isoschizomer to Eco52I). Purified PCR product (1.6 kb) was digested using Anza 3 BcuI, Anza 9 NdeI, and Anza 47 Eco521, as well as Competitor N SpeI-HF (isoschizomer to BcuI), Competitor N NdeI and Competitor N EagI-HF (isoschizomer to Eco52I). Incubation was done at 37°C and per protocol for each enzyme, respectively.

Mapping of restriction enzyme cut sites



A flexible system

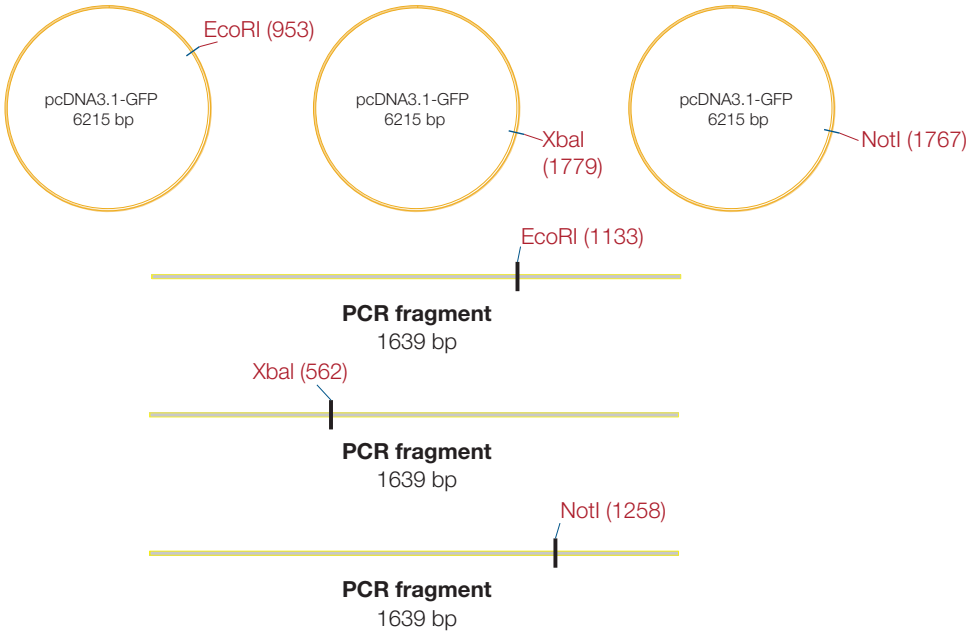
Some restriction enzymes can exhibit star activity, or a decrease in specificity to their DNA recognition site, with prolonged digestions. Star activity results in nonspecific cleavage of DNA and can occur when reaction conditions are not optimal, such as high glycerol content or presence of Mg^{2+}. Anza restriction enzymes, in conjunction with the Anza buffer, have been optimized to allow for flexibility in digestion times—from complete digestion in 15 minutes to overnight digestions—without the worry of star activity.



L – 1 Kb Plus DNA Ladder
C1 – undigested plasmid DNA
1 – Anza 11 EcoRI
2 – Anza 12 XbaI
3 – Anza 1 NotI
C2 – Undigested PCR fragment
4 – Anza 11 EcoRI
5 – Anza 12 XbaI
6 – Anza 1 NotI

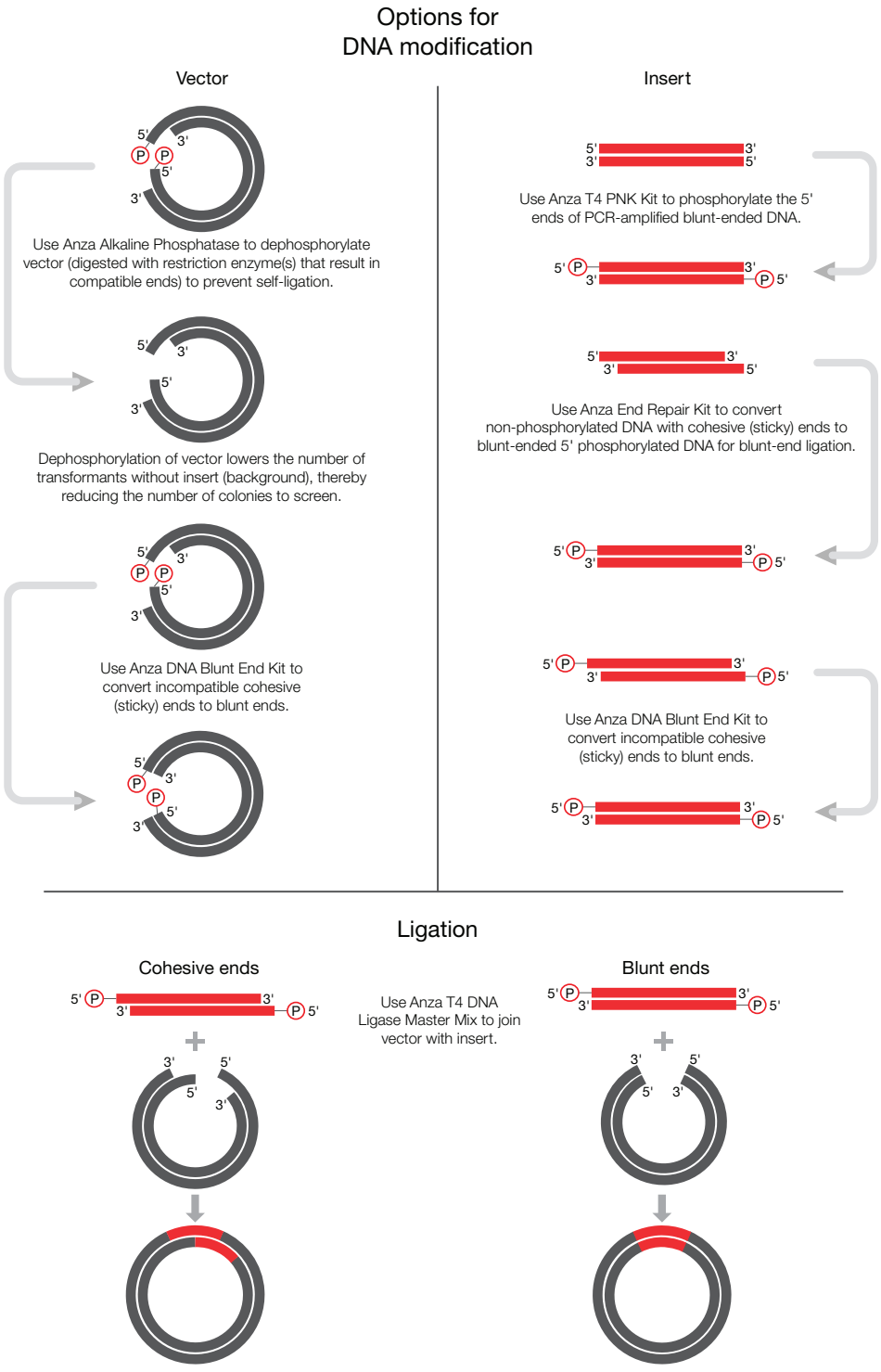
Figure 3. Anza restriction enzymes show complete digestion in 15 minutes with no star activity after overnight digestion. Plasmid DNA (6,215 bp) and purified PCR product (1.6 kb) were digested using Anza 11 EcoRI, Anza 12 XbaI, and Anza 1 NotI. Reaction mixture included 1 µg of DNA and 1 µL of restriction enzyme to a total volume of 20 µL, following the recommended protocol. Incubation was done at 37°C for 15 minutes on the left and 37°C for 16 hours on the right.

Mapping of restriction enzyme cut sites



A complete system

The Anza restriction enzyme cloning xystem is a complete system, inclusive of additional Anza DNA modifying enzymes that are fully functional with the Anza buffer. Now you can complete your additional cloning steps with ease.



DNA modifying enzymes

Anza T4 DNA Ligase Master Mix

Anza Thermosensitive Alkaline Phosphatase

Anza T4 Polynucleotide Kinase

Anza DNA Blunt End Kit

Anza DNA End Repair Kit

Get started

The Anza Starter Kits include 5 or 10 of the most common Anza restriction enzymes, Anza 10X buffer, and selected Anza DNA modifying enzymes, so you can be on your way to simpler cloning.

Anza 5-pk Starter Kit

Anza 1 NotI

Anza 5 BamHI

Anza 11 EcoRI

Anza 12 XbaI

Anza 16 HindIII

Anza T4 DNA Ligase Master Mix

Anza Alkaline Phosphatase

Anza 10X Buffer

Anza 10X Red Buffer

Anza 10-pk Starter Kit

Anza 1 NotI

Anza 3 BcuI*

Anza 5 BamHI

Anza 8 XhoI

Anza 10 DpnI

Anza 11 EcoRI

Anza 12 XbaI

Anza 16 HindIII

Anza 19 BglII

Anza 26 Eco32I**

Anza T4 DNA Ligase Master Mix

Anza Alkaline Phosphatase

Anza 10X Buffer

Anza 10X Red Buffer

*Isoschizomer to prototype SpeI

**Isoschizomer to prototype EcoRV

An isoschizomer is a protein that recognizes the same recognition sequence and digests the DNA in the same location and pattern.

Ordering information

Product	Units (U)	Volume (µL)	Conc. (U/µL)	Cat. No.
Restriction enzymes				
Anza 1 NotI	500	25	20	IVGN001-4
Anza 1 NotI	2,000	100	20	IVGN001-6
Anza 2 NcoI	800	40	20	IVGN002-6
Anza 2 NcoI	4,000	200	20	IVGN002-8
Anza 3 BcuI	400	40	10	IVGN003-4
Anza 3 BcuI	2,000	200	10	IVGN003-6
Anza 4 BpiI	250	25	10	IVGN004-4
Anza 4 BpiI	1,200	120	10	IVGN004-6
Anza 5 BamHI	8,000	800	10	IVGN005-6
Anza 5 BamHI	24,000	2,400	10	IVGN005-8
Anza 6 NheI	800	80	10	IVGN006-6
Anza 6 NheI	4,000	400	10	IVGN006-8
Anza 7 BshTI	250	25	10	IVGN007-4
Anza 7 BshTI	1,200	120	10	IVGN007-6
Anza 8 XhoI	4,000	200	20	IVGN008-6
Anza 8 XhoI	20,000	1,000	20	IVGN008-8
Anza 9 NdeI	3,000	150	20	IVGN009-6
Anza 9 NdeI	15,000	750	20	IVGN009-8
Anza 10 DpnI	800	40	20	IVGN010-6
Anza 10 DpnI	4,000	200	20	IVGN010-8
Anza 11 EcoRI	8,000	400	20	IVGN011-6
Anza 11 EcoRI	40,000	2,000	20	IVGN011-8
Anza 12 XbaI	2,000	200	10	IVGN012-6
Anza 12 XbaI	12,000	1,200	10	IVGN012-8
Anza 13 Esp3I	800	160	5	IVGN013-6
Anza 14 SalI	1,500	75	20	IVGN014-6
Anza 14 SalI	8,000	400	20	IVGN014-8
Anza 15 XmaJI	100	20	5	IVGN015-4
Anza 15 XmaJI	400	80	5	IVGN015-6
Anza 16 HindIII	8,000	400	20	IVGN016-6
Anza 16 HindIII	40,000	2,000	20	IVGN016-8
Anza 17 KpnI	3,000	300	10	IVGN017-6
Anza 17 KpnI	15,000	1,500	10	IVGN017-8
Anza 18 PacI	200	100	2	IVGN018-4
Anza 18 PacI	1,000	500	2	IVGN018-6
Anza 19 BglII	1,600	160	10	IVGN019-6
Anza 19 BglII	8,000	800	10	IVGN019-8
Anza 20 SacI	1,600	160	10	IVGN020-6
Anza 20 SacI	8,000	800	10	IVGN020-8

Product	Units (U)	Volume (µL)	Conc. (U/µL)	Cat. No.
Restriction enzymes				
Anza 21 SgsI	400	40	10	IVGN021-4
Anza 21 SgsI	2,000	200	10	IVGN021-6
Anza 22 SmaI	2,000	400	5	IVGN022-6
Anza 22 SmaI	8,000	1,600	5	IVGN022-8
Anza 23 PstI	8,000	800	10	IVGN023-6
Anza 23 PstI	40,000	4,000	10	IVGN023-8
Anza 24 MssI	400	40	10	IVGN024-4
Anza 24 MssI	2,000	200	10	IVGN024-6
Anza 25 PaeI	400	40	10	IVGN025-4
Anza 25 PaeI	2,000	200	10	IVGN025-6
Anza 26 Eco32I	3,000	150	20	IVGN026-6
Anza 26 Eco32I	15,000	750	20	IVGN026-8
Anza 27 PvuI	400	40	10	IVGN027-4
Anza 28 MluI	800	80	10	IVGN028-6
Anza 29 KflI	100	50	2	IVGN029-4
Anza 30 Bsu15I	1,500	75	20	IVGN030-6
Anza 31 MunI	400	40	10	IVGN031-4
Anza 32 ApaI	4,000	2,000	2	IVGN032-6
Anza 33 LguI	400	40	10	IVGN033-4
Anza 34 Pfl23II	250	125	2	IVGN034-4
Anza 35 Eco47III	200	20	10	IVGN035-4
Anza 36 Eco31I	800	80	10	IVGN036-6
Anza 37 Mph1103I	800	160	5	IVGN037-6
Anza 38 ScaI	800	80	10	IVGN038-6
Anza 39 Bsp1407I	800	160	5	IVGN039-6
Anza 40 SfaAI	400	40	10	IVGN040-4
Anza 41 HpyF3I	800	160	5	IVGN041-6
Anza 42 RsaI	800	160	5	IVGN042-6
Anza 43 Eco105I	400	80	5	IVGN043-4
Anza 44 AluI	800	40	20	IVGN044-6
Anza 45 PteI	400	80	5	IVGN045-4
Anza 46 AatII	400	80	5	IVGN046-4
Anza 47 Eco52I	400	20	20	IVGN047-4
Anza 48 MnlI	400	80	5	IVGN048-4
Anza 49 SmiI	1,500	300	5	IVGN049-6
Anza 50 KspAI	400	40	10	IVGN050-4
Anza 51 BspTI	1,500	300	5	IVGN051-6
Anza 52 PvuII	4,000	800	5	IVGN052-6
Anza 53 AanI	150	75	2	IVGN053-4

Ordering information (continued)

Product	Units (U)	Volume (µL)	Conc. (U/µL)	Cat. No.
Restriction enzymes				
Anza 54 Eco147I	800	80	10	IVGN054-6
Anza 55 MboI	400	80	5	IVGN055-4
Anza 56 Hin1II	1,000	100	10	IVGN056-6
Anza 57 Bpu1102I	400	40	10	IVGN057-4
Anza 58 PagI	400	80	5	IVGN058-4
Anza 59 HhaI	1,500	150	10	IVGN059-6
Anza 60 Kpn2I	800	40	20	IVGN060-6
Anza 61 PfoI	40	20	2	IVGN061-4
Anza 62 MlsI	400	20	20	IVGN062-4
Anza 63 CpoI	400	200	2	IVGN063-4
Anza 64 SaqAI	400	40	10	IVGN064-4
Anza 65 MspI	4,000	200	20	IVGN065-6
Anza 66 BstXI	800	80	10	IVGN066-6
Anza 67 RruI	800	400	2	IVGN067-6
Anza 68 BsuRI	2,500	250	10	IVGN068-6
Anza 69 BglI	1,500	150	10	IVGN069-6
Anza 70 NsbI	400	80	5	IVGN070-4
Anza 71 HinfI	4,000	200	20	IVGN071-6
Anza 72 HincII	800	80	10	IVGN072-6
Anza 73 BclI	2,500	250	10	IVGN073-6
Anza 74 CsiI	200	20	10	IVGN074-4
Anza 75 Alw44I	2,000	100	20	IVGN075-6
Anza 76 VspI	1,500	150	10	IVGN076-6
Anza 77 DraI	1,500	150	10	IVGN077-6
Anza 78 AdeI	800	40	20	IVGN078-6
Anza 79 PdiI	400	40	10	IVGN079-4
Anza 80 FspBI	400	80	5	IVGN080-4
Anza 81 Eco91I	1,500	150	10	IVGN081-6
Anza 82 Eco72I	1,500	750	2	IVGN082-6
Anza 83 Eco81I	800	80	10	IVGN083-6
Anza 84 FspAI	200	20	10	IVGN084-4
Anza 85 MreI	100	20	5	IVGN085-4
Anza 86 PdmI	800	160	5	IVGN086-6
Anza 87 Eco47I	1,500	75	20	IVGN087-6
Anza 88 Bsp119I	2,000	200	10	IVGN088-6
Anza 89 Mva1269I	400	40	10	IVGN089-4
Anza 90 Eco88I	1,500	300	5	IVGN090-6
Anza 91 Acc65I	1,500	300	5	IVGN091-6
Anza 92 EheI	500	25	20	IVGN092-4

Product	Units (U)	Volume (µL)	Conc. (U/µL)	Cat. No.
Restriction enzymes				
Anza 93 HpaII	1,500	150	10	IVGN093-6
Anza 94 BfmI	200	20	10	IVGN094-4
Anza 95 MauBI	100	20	5	IVGN095-4
Anza 96 XmiI	800	160	5	IVGN096-6
Anza 97 Bsp143I	800	80	10	IVGN097-6
Anza 98 XceI	200	40	5	IVGN098-4
Anza 99 XagI	800	80	10	IVGN099-6
Anza 100 Bsh1236I	800	80	10	IVGN100-6
Anza 101 BoxI	800	160	5	IVGN101-6
Anza 102 CaiI	400	40	10	IVGN102-4
Anza 103 Psp1406I	250	125	2	IVGN103-4
Anza 104 MboII	250	25	10	IVGN104-4
Anza 105 Hin1I	1,500	150	10	IVGN105-6
Anza 106 Van91I	800	40	20	IVGN106-6
Anza 107 BspLI	200	20	10	IVGN107-4
Anza 108 SatI	150	30	5	IVGN108-4
Anza 109 Alw26I	800	40	20	IVGN109-6
Anza 111 XapI	800	40	20	IVGN111-6
Anza 112 BseGI	800	160	5	IVGN112-6
Anza 113 BcnI	1,500	150	10	IVGN113-6
Anza 114 Hpy8I	800	80	10	IVGN114-6
Anza 115 MbiI	800	80	10	IVGN115-6
Anza 116 Cfr13I	800	160	5	IVGN116-6
Anza 117 EcoO109I	1,500	150	10	IVGN117-6
Anza 118 BseDI	800	160	5	IVGN118-6
Anza 119 BmsI	250	25	10	IVGN119-4
Anza 120 NmuCI	400	20	20	IVGN120-4
Anza 121 Bsp120I	1,200	120	10	IVGN121-6
Anza 122 Csp6I	1,500	300	5	IVGN122-6
Anza 123 Hin6I	1,500	150	10	IVGN123-6
Anza 124 PfeI	400	80	5	IVGN124-4
Anza 125 HpyF10VI	400	40	10	IVGN125-4
Anza 126 Alw21I	800	160	5	IVGN126-6
Anza 127 RseI	400	40	10	IVGN127-4
Anza 128 PspFI	200	20	10	IVGN128-4
Anza 129 BshNI	400	20	20	IVGN129-4

Ordering information (continued)

Product	No. of reactions	Volume (µL)	Conc. (U/µL)	Cat. No.
DNA modifying enzymes				
Anza 10X Buffer Set	2,000	4,000 (4 x 1,000)	—	IVGN200-8
Anza T4 DNA Ligase Master Mix	200	800 (4 x 200)	4X	IVGN210-8
Anza T4 DNA Ligase Master Mix	50	250	4X	IVGN210-4
Anza Alkaline Phosphatase	2,000	2,000 (4 x 500)	1 U/µL	IVGN220-8
Anza Alkaline Phosphatase	500	500	1 U/µL	IVGN220-4
Anza T4 PNK Kit	50	50	10 U/µL	IVGN230-4
Anza DNA Blunt End Kit	100	100	—	IVGN240-4
Anza DNA End Repair Kit	20	20	—	IVGN250-4

Product	Units (U)	Volume (µL)	Conc. (U/µL)	Cat. No.
Starter packs				
Anza 5-pk Starter Kit				IVGN300-4
Anza 1 NotI	400	20	20	
Anza 5 BamHI	200	20	10	
Anza 11 EcoRI	400	20	20	
Anza 12 XbaI	200	20	10	
Anza 16 HindIII	400	20	20	
Anza T4 DNA Ligase Master Mix	20 rxns	100	4X	
Anza Alkaline Phosphatase	20 rxns	20	1	
Anza 10X Buffer	—	500	10X	
Anza 10X Red Buffer	—	500	10X	
Anza 10-pk Starter Kit				IVGN300-6
Anza 1 NotI	400	20	20	
Anza 3 BcuI	200	20	10	
Anza 5 BamHI	200	20	10	
Anza 8 XhoI	400	20	20	
Anza 10 DpnI	400	20	20	
Anza 11 EcoRI	400	20	20	
Anza 12 XbaI	200	20	10	
Anza 16 HindIII	400	20	20	
Anza 19 BglII	200	20	10	
Anza 26 Eco32I	400	20	20	
Anza T4 DNA Ligase Master Mix	20 rxns	100	4X	
Anza Alkaline Phosphatase	20 rxns	20	1	
Anza 10X Buffer	—	500	10X	
Anza 10X Red Buffer	—	500	10X	

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