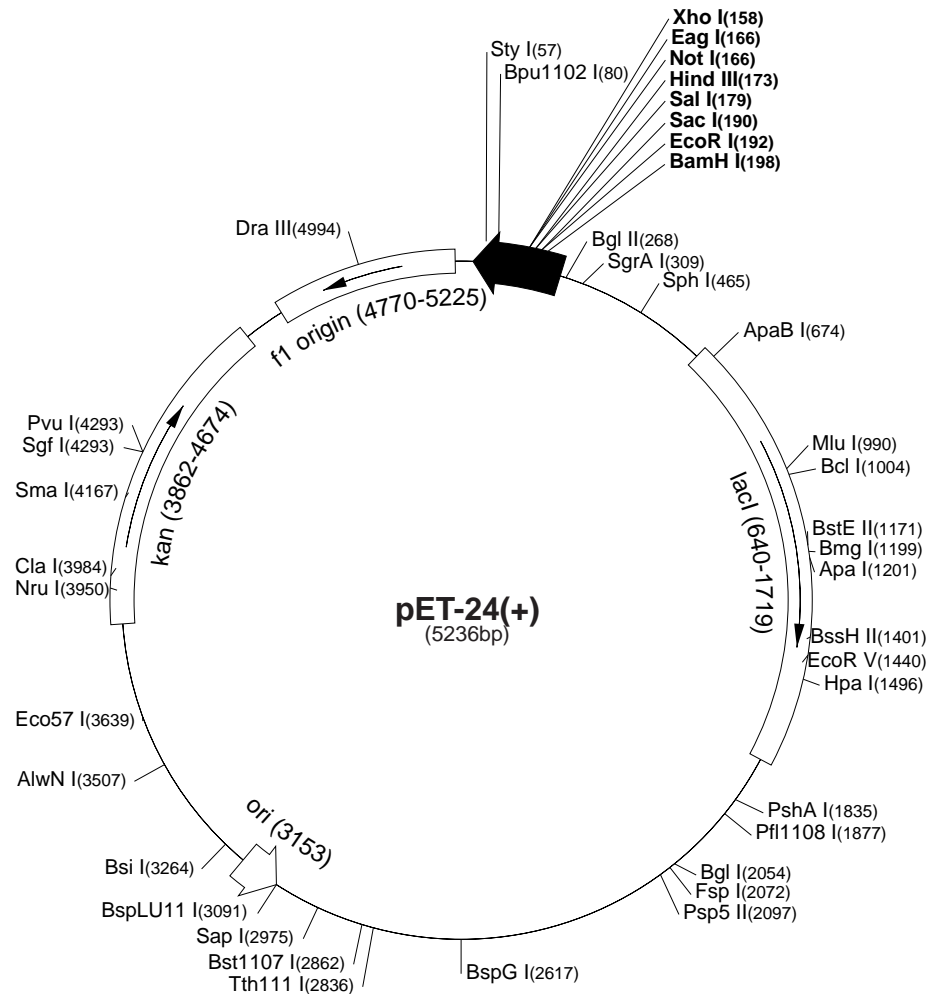


pET-24(+)⁺ Vector

pET-24(+)⁺ (Cat. No. 69772-3) is a transcription vector designed for expression from bacterial translation signals carried within a cloned insert. It therefore lacks the ribosome binding site and ATG start codon present on the pET translation vectors. A C-terminal His•Tag[®] sequence is available. Unique sites are shown on the circle map. Note that the sequence is numbered by the pBR322 convention, so the T7 expression region is reversed on the circular map. The cloning/expression region of the coding strand transcribed by T7 RNA polymerase is shown below. The f1 origin is oriented so that infection with helper phage will produce virions containing single-stranded DNA that corresponds to the coding strand. Therefore, single-stranded sequencing should be performed using the T7 terminator primer (Cat. No. 69337-3).

pET-24(+)⁺ sequence landmarks

| | |
|--|-----------|
| T7 promoter | 237-253 |
| T7 transcription start | 236 |
| Multiple cloning sites (<i>Bam</i> H I - <i>Xho</i> I) | 158-203 |
| His•Tag coding sequence | 140-157 |
| T7 terminator | 26-72 |
| <i>lac</i> I coding sequence | 640-1719 |
| pBR322 origin | 3153 |
| Kan coding sequence | 3862-4674 |
| f1 origin | 4770-5225 |



pET-24(+)⁺ cloning/expression region

pET-24(+) Restriction Sites

| Enzyme | # Sites | Locations |
|----------|---------|--|
| AccI | 2 | 180 2861 |
| AccIII | 7 | 757 1485 1816 2600 2741 3043 4834 |
| Acil | 73 | |
| AflIII | 2 | 990 3091 |
| AluI | 22 | |
| AlwI | 13 | |
| Alw21I | 7 | 159 190 490 974 2085 2909 3409 |
| Alw44I | 3 | 970 2905 3405 |
| AlwNI | 1 | 3507 |
| ApaI | 1 | 1201 |
| ApaBI | 1 | 674 |
| ApoI | 6 | 192 1265 3906 4090 4796 4807 |
| AvaI | 2 | 158 4165 |
| AvaII | 5 | 1542 1918 2006 2097 2376 |
| BamHI | 1 | 198 |
| BanI | 8 | 312 333 447 910 1629 1759 1885 5031 |
| BanII | 6 | 190 374 388 1201 3948 5069 |
| BbsI | 4 | 1136 1475 1849 2209 |
| BbvI | 23 | |
| BccI | 13 | |
| Bce83I | 6 | 21 1804 1974 3182 3480 3721 |
| BceII | 6 | 509 850 1477 3593 4612 5020 |
| BcgI | 8 | 160 194 1282 1316 1816 1850 2668 2702 |
| BclI | 1 | 1004 |
| BfaI | 6 | 70 203 2105 3586 3893 5145 |
| BglI | 1 | 2054 |
| BglII | 1 | 268 |
| BmgI | 1 | 1199 |
| BpmI | 4 | 828 1317 1951 2618 |
| Bpu10I | 2 | 2197 4310 |
| Bpu1102I | 1 | 80 |
| BsaAI | 2 | 2843 4994 |
| BsaBI | 3 | 267 273 2288 |
| BsaHI | 5 | 313 334 448 947 1630 |
| BsaJI | 9 | 57 427 433 1625 2063 3251 4164 4165 4566 |
| BsaWI | 7 | 2 1309 1812 2280 3297 3444 4428 |
| BsaXI | 2 | 1649 4942 |
| Bsbl | 2 | 2807 4901 |
| BscGI | 11 | |
| Bsgl | 3 | 841 1041 2251 |
| Bsil | 1 | 3264 |
| BsiEI | 5 | 169 1775 3007 3431 4293 |
| BsII | 23 | |
| BsmI | 2 | 4177 4254 |
| BsmAI | 6 | 687 1092 1218 1605 2732 4309 |
| BsmBI | 3 | 1605 2732 4309 |
| BsmFI | 4 | 451 1992 2362 5209 |
| BsoFI | 41 | |
| Bsp24I | 10 | 280 312 831 863 1133 1165 3584 3616 3762 3794 |
| Bsp1286I | 12 | |
| BspEI | 2 | 2 2280 |
| BspGI | 1 | 2617 |
| BspLU11I | 1 | 3091 |
| BsrI | 21 | |
| BsrBI | 4 | 223 3024 4692 5138 |
| BsrDI | 2 | 1037 1403 |
| BsrFI | 7 | 300 309 676 1888 2048 4247 5095 |
| BssHII | 1 | 1401 |

| Enzyme | # Sites | Locations |
|----------|---------|---|
| Bst1107I | 1 | 2862 |
| BstEII | 1 | 1171 |
| BstXI | 3 | 792 921 1044 |
| BstYI | 9 | 132 198 268 554 1766 2283 3732 3743 4542 |
| Cac8I | 39 | |
| CjeI | 24 | |
| CjePI | 18 | |
| Clal | 1 | 3984 |
| CviJI | 82 | |
| CviRI | 22 | |
| Ddel | 11 | |
| Dpnl | 21 | |
| DrallI | 1 | 4994 |
| DrdI | 3 | 2784 3199 4949 |
| DrdII | 2 | 713 4999 |
| Dsal | 2 | 427 2063 |
| EaeI | 4 | 166 298 430 1664 |
| EagI | 1 | 166 |
| EarI | 3 | 608 2975 4106 |
| Ecil | 3 | 767 3165 3311 |
| Eco47III | 3 | 395 1896 2345 |
| Eco57I | 1 | 3639 |
| EcoNI | 2 | 525 4205 |
| EcoO109I | 3 | 53 423 2097 |
| EcoRI | 1 | 192 |
| EcoRII | 9 | 713 1028 1568 1625 3117 3238 3251 4181 4538 |
| EcoRV | 1 | 1440 |
| FauI | 17 | |
| FokI | 9 | 1036 1045 2310 2372 2450 2636 2777 3931 4537 |
| Fspl | 1 | 2072 |
| GdIII | 4 | 166 298 430 1664 |
| HaeI | 6 | 718 2039 3106 3117 3569 4380 |
| HaeII | 14 | |
| HaeIII | 23 | |
| HgaI | 11 | |
| HgiEII | 2 | 588 3677 |
| HhaI | 46 | |
| Hin4I | 3 | 889 3979 4521 |
| HincII | 2 | 181 1496 |
| HindIII | 1 | 173 |
| Hinfl | 18 | |
| HpaI | 1 | 1496 |
| HphI | 16 | |
| Maell | 14 | |
| MaellI | 16 | |
| MbolI | 12 | |
| MluI | 1 | 990 |
| MmeI | 7 | 3306 3490 3935 4129 4491 4500 4971 |
| MnlI | 25 | |
| MseI | 23 | |
| MslI | 6 | 1042 1330 1360 2078 2273 2664 |
| MspI | 29 | |
| MspA1I | 8 | 84 1020 1590 1683 2682 2801 3433 3678 |
| MwoI | 39 | |
| NarI | 4 | 313 334 448 1630 |
| NciI | 12 | |
| NgoAIV | 4 | 300 1888 2048 5095 |
| NlaIII | 24 | |
| NlaIV | 21 | |
| Nott | 1 | 166 |
| NruI | 1 | 3950 |
| Nsil | 2 | 4143 4409 |
| NspI | 4 | 465 2436 2728 3095 |
| Pfl1108I | 1 | 1877 |
| PfIMI | 2 | 572 4556 |

| Enzyme | # Sites | Locations |
|----------|---------|---|
| PleI | 9 | 251 539 626 1422 2985 3470 4525 4929 4937 |
| PshAI | 1 | 1835 |
| Psp5II | 1 | 2097 |
| Psp1406I | 4 | 652 2020 2416 4779 |
| PvuI | 1 | 4293 |
| PvuII | 3 | 1590 1683 2682 |
| RcaI | 3 | 388 3811 4686 |
| RsaI | 3 | 1137 2897 4128 |
| SacI | 1 | 190 |
| Sall | 1 | 179 |
| SapI | 1 | 2975 |
| Sau96I | 14 | |
| Sau3AI | 21 | |
| ScrFI | 21 | |
| SfaNI | 23 | |
| Sfcl | 4 | 236 3356 3547 5213 |
| SgfI | 1 | 4293 |
| SgrAI | 1 | 309 |
| Smal | 1 | 4167 |
| SphI | 1 | 465 |
| Sspl | 2 | 4218 4786 |
| StyI | 1 | 57 |
| TaqI | 15 | |
| TaqII | 6 | 898 1116 1789 2993 4547 4898 |
| TfiI | 9 | 1669 1971 2141 2645 3066 4204 4260 4432 4523 |
| Thal | 35 | |
| TseI | 23 | |
| Tsp45I | 7 | 1171 1999 2530 2743 2838 4440 5167 |
| Tsp509I | 19 | |
| Tth111I | 1 | 2836 |
| Tth111II | 8 | 829 1522 2552 3681 3688 3720 4129 4256 |
| UbaII | 18 | |
| VspI | 5 | 251 1675 1734 4492 4681 |
| XcmI | 3 | 846 1362 1380 |
| XhoI | 1 | 158 |
| XmnI | 2 | 2649 4682 |

Enzymes that do not cut pET-24(+):

| | | | | |
|--------|-------|----------|-------|----------|
| AatII | AflII | AgeI | AscI | AvrII |
| BaeI | BsaI | BseRI | BspMI | BsrGI |
| Bsu36I | DraI | Eam1105I | FseI | KpnI |
| MscI | MunI | NcoI | NdeI | NheI |
| NspV | Pacl | PmeI | PmlI | PstI |
| RleAI | RsrII | SacII | Scal | SexAI |
| Sfil | SnaBI | SpeI | SrfI | Sse8387I |
| StuI | SunI | Swal | XbaI | |