Protein Production in Avidity Atum Vectors

The yield of target proteins with different promoters is difficult to predict and varies with the individual proteins being produced. Avidity offers the choice of the T5 promoter (IPTG inducible) and Rha_{BAD} (rhamnose inducible) both of which work well in Avidity's BirA-overproducing *E. coli* strains (AVB100, AVB101).

T5 Induction Protocol

The IPTG-inducible T5 promoter is flanked by dual lac operators for efficient repression of the un-induced promoter. Glucose is preferentially used by the E. coli and presence of glucose will suppress expression of the T5 promoter.

1) Grow the overnight culture in LB or rich media plus kanamycin (50 micrograms /ml).
2) Dilute 1/100 into fresh LB or rich media (no glucose) with kanamycin (50 micrograms/ml) and grow to an OD_{600} of 0.4-0.6 (LB) or OD_{600} 0.6-0.8 (rich media minus glucose).
3) Induce by adding IPTG to 1mM and grow for 4-8 additional hours.

A literature search reveals and interesting twist on the T5 induction with the addition of ethanol to 3% (see "An efficient protocol to enhance recombinant protein expression using ethanol in Escherichia coli",Gaurav Chhetri, Parismita Kalita, Timir Tripathi; MethodsX 2 (2015) 385–391.)