Aureobasidin A: Selectable Drug Resistance for Yeast

A novel reporter gene, *AUR1-C,* confers resistance to the potent yeast antibiotic, Aureobasidin A

- **Aureobasidin A resistance is a definitive selectable marker for yeast**
- **Effectively reduces backgrounds in Matchmaker™ Gold yeast two-hybrid screens**
- **Identify genuine positives more easily, with fewer false positives**

Aureobasidin A (AbA) is a potent and unique yeast antibiotic that kills *S. cerevisiae* at low concentrations (1). The drug is a cyclic multipeptide (Figure 1) that acts by inhibiting an essential yeast enzyme, inositol phosphorylceramide synthase. A mutant enzyme, encoded by the *AUR1-C* gene, confers resistance to AbA and can be used as a very effective selectable marker that requires little or no optimization. The ability of resistant yeast to grow in the presence of AbA depends on the level of *AUR1-C* expression (see page 3, Figure 5).

Aureobasidin A and Matchmaker Gold

Clontech's **Matchmaker Gold Yeast Two-Hybrid System** (see pages 1–3), is a highly optimized yeast two-hybrid (Y2H) screening system that employs the definitive selectivity of AbA resistance as a reporter for interacting Y2H protein pairs. The *AUR1-C* gene is stably integrated in the Matchmaker Gold reporter strain, **Y2HGold,** and is used for primary and secondary colony screens of Y2H libraries.

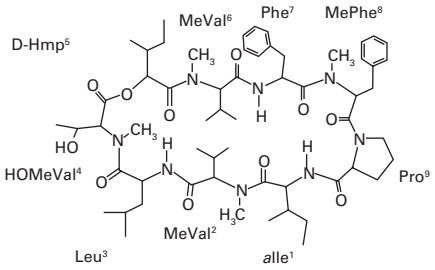

Figure 1. Structure of Aureobasidin A. Aureobasidin A (AbA; MW 1,100) is a cyclic depsipeptide antibiotic isolated from the fungus, *Aureobasidium pullulans* R106. AbA inhibits the product of the yeast AUR1 gene (inositol phosphorylceramide synthase) and is toxic to *S. cerevisiae* at low concentrations (0.1 µg/ml). The gene product of a dominant mutant allele, *AUR1-C,* confers resistance to AbA, and its expression can be used as a selectable marker.

Exceptionally Low Background

AbA selection virtually eliminates the high numbers of background colonies that often plague low stringency primary screens that use nutritional markers alone (e.g. *HIS3*). Because AbA actually kills sensitive cells, rather than merely retarding their growth, AbA-based selection greatly favors the growth and identification of genuinely positive clones. In general practice, a high percentage of clones that emerge from low stringency primary screens using AbA selection, are subsequently verified on high stringency secondary screens that select for all four Matchmaker Gold reporters (*AUR1-C, HIS3, ADE2* and *MEL1).*

Product	Size	Cat. No.
Aureobasidin A	1 mg	630466
Matchmaker Gold Yeast Two-Hybrid System	each	630489

NEW!

Notice to Purchaser

Please see the Aureobasidin Drug, Aureobasidin Resistance Gene, Matchmaker™ Two-Hybrid System, and Reverse Two-Hybrid Technology licensing statements on page 40.

Like Ampicillin... for Yeast

Many researchers have yearned for a yeast selection system akin to those used for *E. coli* or mammalian cells. In fact, AbA is used for yeast essentially as ampicillin and kanamycin are used for cloning in *E. coli,* or as G418 is used to select stably transfected clones of mammalian cells. AbA resistance is far easier to use in Y2H library screening than are auxotrophic reporters, which often require optimization to achieve selective growth conditions.

If you wish to take your search for protein-protein interactions to the next level, look no further than Matchmaker Gold, and take advantage of the powerful and definitive selection of Aureobasidin A.

Reference

1. Takesako, K. *et. al* (1993) *J. Antibiot.* (Tokyo) **46**(9):1414–1420.