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Matchmaker gold yeast two hybrid system user manual

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YUMPU effortlessly converts print PDFs into web-optimized ePapers that Google loves. Additionally, Copy Matchmakerâ„¢ Gold Yeast Two Hybrid System User Manual Extended embed settings describe how to transform your publications into a flipbook using our unique Google-optimized e-Paper software. The Matchmaker Gold system is distinctive because it employs the AUR1-C gene as a novel reporter conferring resistance to AbA, a potent antibiotic toxic to S. cerevisiae. This property enables straightforward Y2H library screening without optimization required for nutritional selection alone. Four reporters identify genuine positives The stringency of Matchmaker Gold relies on the use of four selective reporter genes: AUR1-C, HIS3, ADE2, and MEL1 (α-galactosidase), whose expression is driven by three different GAL4-binding promoters. All reporter genes are stably integrated into the genome of the Y2HGold reporter strain. Superior to nutritional selection alone The use of auxotrophic reporters for Y2H screening often requires optimization steps, especially with HIS3-based selection. The Matchmaker Gold system eliminates this need by using AbA selection and two nutritional markers. This makes 3-AT unnecessary, which is typically required to suppress growth of background colonies. Mate & Plate libraries and screening Another significant advantage of Matchmaker Gold is the easy Mate & Plate strategy that replaces cumbersome library-scale yeast transformation. This involves combining haploid yeast strains expressing bait and prey fusion proteins (Figure 3). We offer pretransformed Mate and Plate libraries or allow users to create their own using the Make Your Own “Mate & Plate” Library System. The Y187 strain is combined with a culture expressing bait proteins from the Y2HGold strain, and then cultured overnight before being plated on selective agar medium. This process utilizes the Matchmaker Gold Yeast Two-Hybrid System, which offers advanced capabilities due to its new yeast strain, stringent reporters, and easy-to-use libraries. A key aspect of this system is its use of four independent reporter genes, which helps to minimize false positives by requiring prey proteins to interact with all three bait recognition sequences and activate all four reporters. This approach also eliminates the possibility of background growth, as Aureobasidin A, a novel yeast antibiotic, kills non-resistant cells. The Matchmaker Gold Yeast Two-Hybrid System provides a robust platform for investigating protein-protein interactions, making it an ideal tool for researchers seeking to identify novel interactions, confirm suspected ones, and define interacting domains.