Takara Bio provides yeast media in three different formats:

I. **Yeast Media Sets** (Table 1) are specifically designed for Matchmaker® Gold Systems.

II. **Yeast Media Pouches** (Table 2) are ready-to-go preformulated pouches; just add water and autoclave. No measuring, mixing, or pH adjustments are required. Each pouch makes 0.5 L of media, with or without agar.

III. **Bulk media** (Table 3), which require measuring, mixing, and pH adjustment, are also available.

I. Matchmaker Gold Yeast Media Sets:

Matchmaker Gold Media Sets contain the media and supplements you need for yeast two-hybrid screening. The ready-to-go pouches are supplied in the quantities sufficient for the screening protocols described in the Matchmaker Gold Yeast Two-Hybrid System User Manual (PT4084-1).

Table 1 lists the components supplied in the **Yeast Media Set 2** (Cat. No. 630494). The **Yeast Media Set 2 Plus** (Cat. No. 630495) contains X-alpha-Gal and Aureobasidin A in addition to the media pouches of Yeast Media Set 2.

- For preparing media using the pouches, see the directions in Section II below.
- The Matchmaker Gold User Manual (PT4084-1) contains the directions for using **Aureobasidin A** (Cat. Nos. 630466 & 630499) and **X-alpha-Gal** (Cat. Nos. 630462 & 630463).

Table 1. Components of Yeast Media Set 2 & Yeast Media Set 2 Plus

Yeast Media Set 2 Pouches (Cat. No. 630494)	Quantity	Volume of Media
YPDA Broth	2	0.5 L
YPDA with Agar	1	0.5 L
SD/–Leu Broth	1	0.5 L
SD/–Leu with Agar	1	0.5 L
SD/–Trp Broth	1	0.5 L
SD/–Trp with Agar	1	0.5 L
SD/–Leu/–Trp with Agar	10	0.5 L
SD/–Ade/–His/–Leu/–Trp with Agar	1	0.5 L
Additional Components in Yeast Media Set 2 Plus (Cat. No. 630495)		
X-alpha-Gal	250 mg	—
Aureobasidin A	1 mg	—

II. Preparing Ready-To-Go Yeast Media Pouches:

See Table 2 below for a list of available yeast media pouches for Matchmaker Gold Protocols, as well as **Aureobasidin A** (Cat. Nos. 630466 & 630499) and **X-alpha-Gal** (Cat. Nos. 630462 & 630463).

- Yeast media pouches are supplied as packs of 10.
- YPDA pouches contain YPD supplemented with adenine hemisulfate.
- SD pouches contain premixed yeast nitrogen base (YNB), ammonium sulfate, glucose, and the indicated amino acid dropout supplement(s).

1. Simply add the pouch contents to 500 ml ddH_20 and autoclave 15 min at 121°C. Broth media that does not contain agar may also be filter sterilized.
2. pH adjustment is not usually required, but if the water used is particularly acidic, adjust pH to 5.8 before autoclaving.

Table 2. Yeast Media Pouches for Matchmaker Gold Protocols

Yeast Media Pouches	Cat. No.	Quantity (each)	Volume (Liter/Pouch)
Rich Media (for routine culturing of untransformed yeast)			
YPDA Broth	630306	10	0.5 L
YPDA with Agar	630307	10	0.5 L
Minimal Media Single Dropouts (SDO)			
SD–Trp Broth	630308	10	0.5 L
SD–Trp with Agar	630309	10	0.5 L
SD–Leu Broth	630310	10	0.5 L
SD–Leu with Agar	630311	10	0.5 L
Minimal Media Double Dropouts (DDO)			
SD–Leu/–Trp Broth	630316	10	0.5 L
SD–Leu/–Trp with Agar	630317	10	0.5 L
Minimal Media Triple Dropouts (TDO)			
SD–His/–Leu/–Trp Broth	630318	10	0.5 L
SD–His/–Leu/–Trp with Agar	630319	10	0.5 L
Minimal Media Quadruple Dropouts (QDO)			
SD–Ade/–His/–Leu/–Trp Broth	630322	10	0.5 L
SD–Ade/–His/–Leu/–Trp with Agar	630323	10	0.5 L
X-alpha-Gal	630462	100 mg	—
	630463	250 mg	—
Aureobasidin A	630466	1 mg	—
	630499	10 mg	—

III. Preparing Rich and Minimal Bulk Media:

See Table 3 below for a list of available bulk yeast media and supplements for two-hybrid screening.

A. Rich Liquid Media (Broth):

1. Add 50 g of **YPD** or **YPDA Medium** to 1 L of deionized H_2O. Mix to dissolve.
2. Important: verify that the medium has a pH of 6.5; adjust if necessary.
3. **Autoclave at 121°C for 15 min.**
4. Store autoclaved medium at room temperature in subdued light.

B. Rich Plating Media with Agar:

1. Add 70 g of **YPD** or **YPDA Agar Medium** to 1 L of deionized H_2O. Mix to dissolve. The agar will not dissolve until it is autoclaved.
2. Important: verify that the medium has a pH of 6.5; adjust if necessary.
3. **Autoclave at 121°C for 15 min.** Cool to ~50°C before pouring plates.
4. Allow plated medium to harden at room temperature. Store plates inverted, in a plastic sleeve at 4°C.

C. Minimal Liquid Media (Broth):

1. Add a **Minimal SD Base Medium** and the appropriate dropout supplement in the amounts shown in Table I to
   1 L of deionized H_2O and stir to dissolve.
2. Important: verify that the medium has a pH of 5.8; adjust if necessary.
3. **Autoclave at 121°C for 15 min.**
4. Store autoclaved, liquid SD medium at 4° in subdued light.

D. Minimal Plating Media with Agar:

1. Add a **Minimal SD Agar Base** and the appropriate dropout supplement(s) in the amounts shown in Table I to
   1 L of deionized H_2O. Mix to dissolve. The agar will not dissolve until it is autoclaved.
2. Important: verify that the medium has a pH of 5.8; adjust if necessary.
3. **Autoclave at 121°C for 15 min.** Cool to ~50°C before pouring plates.
4. Allow plated medium to harden at room temperature. Store plates inverted, in a plastic sleeve at 4°C.

Table 3. Bulk Yeast Media and Supplements for Two-Hybrid Screening

Yeast Media	Cat. No.	Final Concentration (g/L)
Rich Media (for routine culturing of untransformed yeast)		
YPD Medium (500 g)	630409	50
YPD Agar Medium (700 g)	630410	70
YPDA Medium (500 g)	630464	50
YPDA Agar (700 g)	630465	70
Minimal Media (for preparing specific DO media)		
Minimal SD Base Medium (267 g)	630411	26.7
Minimal SD Agar Base (467 g)	630412	46.7
Minimal SD Base/Gal/Raf (185 g)	630420	37
Minimal SD Agar Base/Gal/Raf (270 g)	630421	54
Single Dropout Supplements (SDO)		
-Trp DO Supplement (10 g)	630413	0.74
-Leu DO Supplement (10 g)	630414	0.69
-His DO Supplement (10 g)	630415	0.77
-Ura DO Supplement (10 g)	630416	0.77
Double Dropout Supplements (DDO)		
-Leu/-Trp DO Supplement (10 g)	630417	0.64
-His/-Leu DO Supplement (10 g)	630418	0.67
-His/-Ura DO Supplement (10 g)	630422	0.75
-Trp/-Ura DO Supplement (10 g)	630427	0.72
-Met/-Trp DO Supplement (10 g)	630431	0.64
Triple Dropout Supplements (TDO)		
-His/-Leu/-Trp DO Supplement (10 g)	630419	0.62
-His/-Leu/-Ura DO Supplement (10 g)	630423	0.65
-His/-Trp/-Ura DO Supplement (10 g)	630424	0.70
-Leu/-Trp/-Ura DO Supplement (10 g)	630426	0.62
-Leu/-Met/-Trp DO Supplement (10 g)	630430	0.62
Quadruple Dropout Supplements (QDO)		
-His/-Leu/-Trp/-Ura DO Supplement (10 g)	630425	0.60
-Ade/-His/-Leu/-Trp DO Supplement (10 g)	630428	0.60
-His/-Leu/-Met/-Trp DO Supplement (10 g)	630429	0.60

*Freezing Medium is YPD Medium plus 25% glycerol.

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