

## ETHANOL RED® ACTIVE DRY YEAST PRODUCT GUIDELINES

Ingredients	Yeast ( <i>saccharomyces cerevisiae</i> )
Properties	% dry weight = 94 – 96.5% Living cells at packaging > 20 x 10 <sup>9</sup> /gram
Package	10 kg (22.05 pounds) vacuum packed sachets contained in a cardboard protective box
Shelf Life	18 months (< 70° F). Higher storage temperatures will result in increased loss of activity. Product should never be stored above 80° F (27° C). Partially used packages should be tightly sealed, removing as much air as possible, stored at refrigerated temperatures (40° F or 4° C) and used within 1 week of opening. To maintain highest activity and to assure best results, Phibro recommends that purchase quantities be limited to a 3-4 month supply.
Kosher	Pareve certified

### Dosage

The goal when determining the inoculation rate is to allow for maximum biomass production during propagation and to insure that yeast is in its exponential growth phase at time of pitch. The general rule of thumb is to dose at approximately 1 box (10 kg) per 200,000 gallons (~ 757,000 liters) of fermentor working volume. Usage of this rule of thumb will yield a dose rate of 3 boxes of Ethanol Red for a fermentor with the working volume of 650,000 to 700,000 gallons (~ 2,460,500 to 2,650,000 liters).

- Typical dose for 50 mgpy = 3 boxes
- Typical dose for 100 mgpy = 4 boxes

Other process conditions may warrant increasing the doses recommended in the guidelines above. Such conditions would include:

1. Mash dry solids concentrations approaching 34 to 35%.
2. Bacterial infections resulting in elevated lactic and/or acetic acid levels. Lactic acid levels in excess of 0.4% w/v and acetic acid levels approaching 0.15% w/v will be strongly inhibitory to yeast and will require additional yeast dosing during bacterial excursions.
3. Reduced propagation times below 5 hours.

### Rehydration

- Optimum mix tank temperature = 92° F to 98° F (33-37° C), setting yeast tank temperature at approximately 90° F (32° C) will ensure that optimum yeast temperature is not exceeded. **NOTE:** Reaching > 105° F is likely to result in yeast cell death.
- Time = Not less than 15 minutes and not more than 30 minutes. (Longer time periods will initiate thermal kill of some yeast population).
- Nutrient requirements = 2% w/v sugar or 10:1 water to mash ratio mix. The addition of a 5 gallon bucket of mash is usually sufficient. This prevents yeast stress by minimizing osmotic stress across the yeast cell membrane.
- Agitation = Light agitation from a mixing blade or paddle is best to prevent the yeast from clumping and thus not hydrating properly. Ideally, the mixing tank addition pump and the propagator recirculation line should not be used to achieve homogenous mixing due to excessive shear.

### Propagation

Propagation and yeast conditioning time will vary based on plant design and environmental factors. Typical propagation is complete within 8 to 9 hours. Dry solids concentration at the start of propagation should be approximately 20 to 24% while free amino nitrogen (FAN) concentration should be in excess of 600 ppm to encourage biomass production. This can be adjusted depending on environmental yeast stress conditions.



## Typical Profile

A typical profile at the time a propagator is sent to a fermentor is summarized below:

Cell Count	250 to 450 (x10 <sup>6</sup> ) cells per ml
Budding	> 25%
Viability	> 90%
Ethanol	< 2.5% w/v
Glucose	1 – 2% w/v
pH	4.8 to 5.8 depending on rate of acid addition

## Fermentation

Fermenter additions can occur shortly after ferm fill begins. The optimum addition sequence to the fermentor is given below:

1. Antimicrobial
2. Gluco amylase
3. Urea (to yield a FAN of greater than 300 ppm at completion of ferm fill. In a 700,000 gallon or 2,650,000 liter fermentor a urea of dose of 3,000 pounds or 1,361 kg would be typical)
4. Yeast prop  
**Special note:** Mix urea and gluco amylase (GA) separately to prevent denaturing GA with the urea.

The optimum temperature at which to begin fermentation is 93° F (34° C). Numerous environmental factors will impact the rate of fermentation. Primary factors include rate of alpha and gluco amylase addition, yeast inoculation rate, yeast growth phase, bacterial presence, organic acid concentrations, dry solids concentration, temperature and cooling availability, free amino nitrogen availability, and pH. A typical profile that might be seen throughout fermentation is summarized in the table below:

Age	pH	Brix	Temp (°F)	Temp (°C)	Cell Count	Budding	Viability	DP4	DP3	DP2	Glucose	Lactic	Glycerol	Acetic	Ethanol	Total Sugar
YP Drop	4.96	8.0	91.0	32.70	364	36.5	97	3.127	0.066	1.544	1.511	0.098	0.559	0.028	2.077	6.248
6 Hours	5.30	13.7	93.3	34.05	—	—	—	12.661	0.350	4.286	11.363	0.132	0.724	0.056	1.138	28.232
12 Hours	4.80	25.6	93.6	34.22	221.5	30.5	93	8.860	0.255	2.245	11.812	0.154	1.050	0.057	3.260	23.172
18 Hours	4.59	21.4	92.9	33.83	253.5	25	97	6.019	0.239	0.581	9.913	0.152	1.315	0.060	6.467	16.752
24 Hours	4.55	18.0	92.4	33.55	273	22	95	2.956	0.202	0.465	8.778	0.154	1.395	0.060	8.833	12.401
30 Hours	4.58	15.9	90.3	32.38	306.5	13.5	96	2.203	0.163	0.439	5.746	0.156	1.427	0.069	10.625	8.551
36 Hours	4.55	14.2	88.1	31.16	277.5	13	93	1.671	0.133	0.457	3.746	0.159	1.524	0.069	12.439	6.007
42 Hours	4.54	12.6	88.4	31.33	—	—	—	1.305	0.110	0.444	1.826	0.156	1.523	0.075	13.963	3.685
48 Hours	4.57	11.2	88.3	31.27	—	—	—	0.954	0.065	0.415	0.259	0.155	1.541	0.075	14.417	1.623
Drop	4.65	10.6	88.2	31.22	—	—	—	0.646	0.082	0.441	0.050	0.160	1.595	0.081	14.849	1.019

As fermentation progresses, the yeast will become susceptible to increasing levels of stress induced by temperature and increasing ethanol concentration. If it is desired to offset these factors then consideration can be given to temperature staging. This technique will lower the temperature to reduce yeast stress while ethanol concentration is on the rise. This offsetting technique will allow the yeast to remain vital longer and thus provide optimum conditions for maximizing sugar consumption. Temperature staging guidelines would be to start fermentation at the optimum 93° F (34° C) and then reduce it in steps to as low as 88° F (31° C) but will depend on specific plant conditions.



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