Supplementary Material

MATERIALS AND METHODS

Standardization of PJ

PJ was standardized by estimation of total phenolics using Folin-Ciocalteu (FC) reagent with gallic acid standard [1]. Total acids in PJ was estimated by titration with 0.1MNaOH [2]. Total iron in the PJ was estimated by carbonizing and acid digestion of the juice followed by BPS based colorimetric estimation [3, 4]**.**

RESULTS

Standardization of PJ

Total phenolic content of PJ was estimated to be 147.66 ± 19.74 mg gallic acid equivalent/100ml. PJ had total acid content of about 3.2% and total iron content of PJ was 0.903 ± 0.09 mg/100ml.

Generation of ID Cells

The observations from the present study (Table **1**) indicate that culturing *S. cerevisiae* cells in iron deficient medium with 100µm BPS did not alter the cell viability and generation time. We observed a 2% reduction in the cell size (Table **1**) however, this was not statistically significant. About 18% reduction in the dry weight was observed in the ID cells (Table **1**). The iron content in ID cells was 1.041 ± 0.13 ng/mg dry weight, which was 89%lower ($p<0.0001$) than that that in IN cells. It was also observed that the ID cells mainly had ferrous (Fe^{2+}) form of iron, the storage or ferric (Fe^{3+}) form was not detected. Further, the heme content in the ID cells was 2.44 ± 0.08 ng/mg DW, which was 39% lower than that of IN cells (Table **1**). RNA content in the ID cells was estimated to be 86.25 ± 5.90 µg/mg DW, which was 32% less than that in the IN cells ($p<0.0001$). However, reduction in the RNA content did not alter the total protein content significantly in the IN and ID cells, but led to 25% reduction ($p<0.01$) in the lipid content in the ID cells.

Supplementary Table 1: Physiological and Biochemical Parameters in IN and ID *S. cerevisiae* Cells

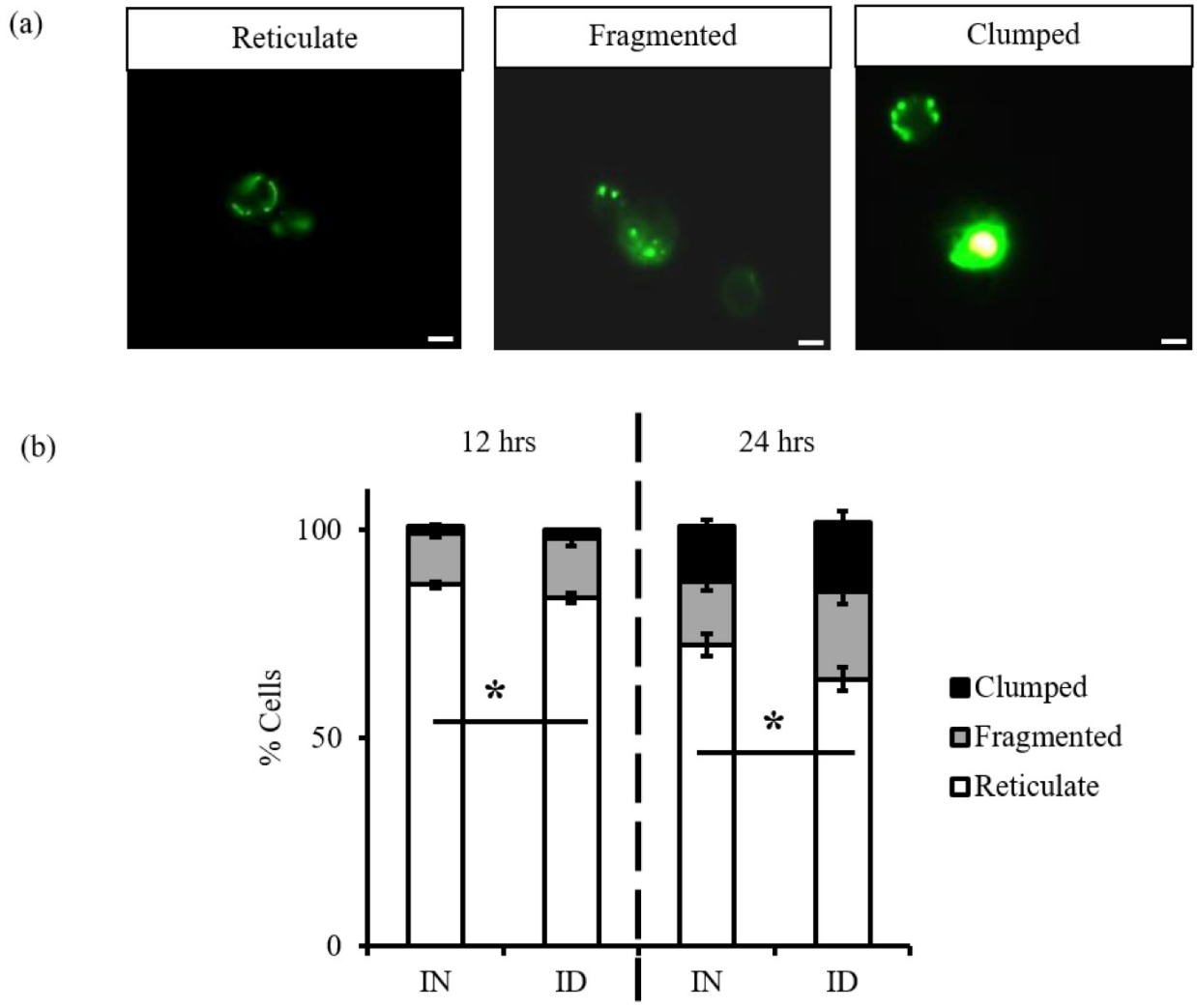
Parameter	IN	ID	% Reduction
Viability (CFU/ml)	1×10^6	1×10^6	0
Generation time (min)	97.18	97.18	0
Cell size (µm)	1.49 ± 0.08	1.46 ± 0.14	2
Dry weight (mg/10^6)	1.75 ± 0.09	1.44 ± 0.03	18*
RNA (µg/mg DW)	126.57 ± 6.69	86.25 ± 5.90	32*
Protein (mg/gm DW)	401 ± 10	398 ± 10	0.75
Lipid (mg/gm DW)	137 ± 5	103 ± 6	25*
Total iron (ng/mg DW)	9.71 ± 0.22	1.041 ± 0.13	89*
Ferrous iron (ng/mg DW)	4.8 ± 0.11	1.04 ± 0.13	78*
Heme (ng/mg DW)	3.99 ± 0.55	2.44 ± 0.08	39*
ATP (ng/mg DW)	117.95 ± 27.24	83.12 ± 33.10	30

DW – Dry weight; * - Significant ($p<0.01$).
IN – Iron normal; ID – Iron deficient.

Iron Deficiency Altered Mitochondrial Structure and Function

The percentage of cells with reticulate or healthy mitochondria was significantly ($p<0.001$) lower in the ID cells when compared to IN cells (Figure **1**). It was also observed that the proportion of cells with fragmented and clumped

(degenerating) mitochondria was higher in the ID cells than in the IN cells (Figure **1b**). This trend was observed in both 12 h and 24 h cultures even though the difference was much pronounced after 24 h. ATP levels in the ID cells, was 30% lower than that detected in IN cells (Table **1**).



Supplementary Figure 1: Mitochondrial structural forms in IN and ID cells. (**a**) Representative images of reticulate, fragmented and clumped mitochondria of *S. cerevisiae* cells (**b**) Percentage of IN and ID cells with each form of mitochondria at 12 h and 24 h (* Statistically significant).

REFERENCES

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