1.1. Methods M3

RNA Isolation: Briefly, cultures were rinsed with sterile tris-buffered saline then collected with Buffer RLT (Qiagen) with 10 mL/L β-2-mercaptoethanol in nuclease free water. Three replicates were performed for all treatments, where each replicate was prepared by pooling cell lysates from four cultures. Cell lysates were homogenized using the QIA shredder followed by purification using the RNeasy Minikit (Qiagen). Isolated RNA was measured for its RNA content using the Nano Drop (ND-1000 spectrophotometer) and Qubit (Qubit 2.0 Fluorometer with RNA assay kit) analyzers (Supplemental data D1). Qubit quantification was used for further dilution calculations. RNA integrity number (RIN) was measured using a Bioanalyzer (Aligent 2100 Bioanalyzer with Aligent RNA 6000 pico kit) and confirmed to be above 8.8 (except one NF sample which had a RIN of 7.7) (Table D1)

Table D1: Nanodrop and Qubit estimates of RNA (ribonucleic acid) content, and Bioanalyzer analysis of RNA Integrity (RIN (RNA integrity number), where level 10 RNA is completely intact). All RIN values were within an acceptable range. Qubit RNA concentrations were used for further dilutions and analysis since Qubit measurements are more specific as they are based on a fluorescence-based dyes that bind specifically to RNA.

Sample #	Sample Name	Nanodrop (ng/ul)	Qubit RNA (ng/ul)	% Differences in Concentration	Bio Analyzer(RIN)
1	SC 1	42.4	41.8	1.5	9.0
2	SC 2	64.7	59.0	8.8	9.0
3	SC 3	68.6	84.2	22.8	9.2
4	NF 1	52.6	50.2	4.6	7.7
5	NF 2	42.2	41.8	1.0	9.1
6	NF 3	36.0	34.2	4.9	9.2
7	TCPS 1	25.5	22.6	11.2	9.5
8	TCPS 2	55.0	57.8	5.1	9.0
9	TCPS 3	44.5	44.2	0.7	9.2
10	TCPS+OS 1	36.2	35.4	2.1	8.9
11	TCPS+OS 2	31.7	27.4	13.5	9.4
12	TCPS+OS 3	56.4	54.2	3.9	9.4
13	MOD SC 1	63.5	67.6	6.5	9.2
14	MOD SC 2	67.7	63.8	5.7	9.1
15	MOD SC 3	61.3	57.2	6.6	9.4
16	MOD NF 1	30.0	28.8	4.1	9.0
17	MOD NF 2	26.6	25.5	4.1	8.8
18	MOD NF 3	43.4	42.2	2.9	9.4