Supplementary Materials

Table S1: Experimental design (Box-Behnken design) for each expression system (*E. coli* strain /expression vector).

a. M15 *E. coli* strain / pQE30 expression vector (M30) *

Sample number (Standard Order)	Randomized Order	Temperature (T) (°C)	Inducer Concentration (I) (mM)	Induction duration (D) (h)	Soluble activity ($U.L^{-1}$ of culture)
1	25	20	0.1	42	1978
2	13	30	0.1	42	1657
3	36	20	1	42	2086
4	8	30	1	42	1678
5	7	20	0.55	18	1517
6	30	30	0.55	18	1597
7	6	20	0.55	66	1941
8	12	30	0.55	66	1502
9	37	25	0.1	18	1795
10	21	25	1	18	1850
11	16	25	0.1	66	1943
12	23	25	1	66	2064
13	31	25	0.55	42	2098
14	3	25	0.55	42	2048
15	9	25	0.55	42	2059
16	17	20	0.1	42	1988
17	26	30	0.1	42	1652
18	43	20	1	42	2122
19	34	30	1	42	1681
20	20	20	0.55	18	1514
21	40	30	0.55	18	1561
22	41	20	0.55	66	1941
23	44	30	0.55	66	1484
24	35	25	0.1	18	1796
25	29	25	1	18	1842
26	2	25	0.1	66	1956
27	15	25	1	66	2061
28	18	25	0.55	42	2068
29	5	25	0.55	42	2098
30	39	25	0.55	42	2064
31	24	20	0.1	42	1991
32	33	30	0.1	42	1649
33	38	20	1	42	2085
34	22	30	1	42	1692
35	1	20	0.55	18	1532
36	14	30	0.55	18	1562
37	4	20	0.55	66	1924
38	19	30	0.55	66	1501
39	11	25	0.1	18	1767

40	42	25	1	18	1840
41	45	25	0.1	66	1943
42	27	25	1	66	2053
43	32	25	0.55	42	2063
44	10	25	0.55	42	2063
45	28	25	0.55	42	2101

***** All the activity measurements were carried out in triplicate.

b. IqExpress *E. coli* strain / pQE30 expression vector (I30) *

Sample number (Standard Order)	Randomized Order	Temperature (T) (°C)	Inducer Concentration (I) (mM)	Induction duration (D) (h)	Soluble activity ($U.L^{-1}$ of culture)
1	25	20	0.1	42	1976
2	13	30	0.1	42	1701
3	36	20	1	42	2128
4	8	30	1	42	1800
5	7	20	0.55	18	1778
6	30	30	0.55	18	1606
7	6	20	0.55	66	2031
8	12	30	0.55	66	1729
9	37	25	0.1	18	1957
10	21	25	1	18	2075
11	16	25	0.1	66	2042
12	23	25	1	66	2106
13	31	25	0.55	42	2162
14	3	25	0.55	42	2176
15	9	25	0.55	42	2174
16	17	20	0.1	42	1995
17	26	30	0.1	42	1667
18	43	20	1	42	2114
19	34	30	1	42	1848
20	20	20	0.55	18	1792
21	40	30	0.55	18	1593
22	41	20	0.55	66	2075
23	44	30	0.55	66	1729
24	35	25	0.1	18	1947
25	29	25	1	18	2080
26	2	25	0.1	66	2014
27	15	25	1	66	2121
28	18	25	0.55	42	2177
29	5	25	0.55	42	2180
30	39	25	0.55	42	2104
31	24	20	0.1	42	2027
32	33	30	0.1	42	1678
33	38	20	1	42	2136
34	22	30	1	42	1805
35	1	20	0.55	18	1778

36	14	30	0.55	18	1619
37	4	20	0.55	66	2048
38	19	30	0.55	66	1733
39	11	25	0.1	18	1942
40	42	25	1	18	2076
41	45	25	0.1	66	2049
42	27	25	1	66	2122
43	32	25	0.55	42	2214
44	10	25	0.55	42	2175
45	28	25	0.55	42	2179

***** All the activity measurements were carried out in triplicate.

c. BL21 DE3 *E. coli* strain / pET19b expression vector (B19) *

Sample number (Standard Order)	Randomized Order	Temperature (T) (°C)	Inducer Concentration (I) (mM)	Induction duration (D) (h)	Soluble activity ($U.L^{-1}$ of culture)
1	25	20	0.1	42	2003
2	13	30	0.1	42	1805
3	36	20	1	42	2197
4	8	30	1	42	1897
5	7	20	0.55	18	1821
6	30	30	0.55	18	1705
7	6	20	0.55	66	2005
8	12	30	0.55	66	1652
9	37	25	0.1	18	1930
10	21	25	1	18	2028
11	16	25	0.1	66	2094
12	23	25	1	66	2190
13	31	25	0.55	42	2125
14	3	25	0.55	42	2156
15	9	25	0.55	42	2137
16	17	20	0.1	42	2001
17	26	30	0.1	42	1800
18	43	20	1	42	2192
19	34	30	1	42	1893
20	20	20	0.55	18	1822
21	40	30	0.55	18	1693
22	41	20	0.55	66	2018
23	44	30	0.55	66	1655
24	35	25	0.1	18	1902
25	29	25	1	18	2008
26	2	25	0.1	66	2101
27	15	25	1	66	2183
28	18	25	0.55	42	2135
29	5	25	0.55	42	2168
30	39	25	0.55	42	2131

31	24	20	0.1	42	1994
32	33	30	0.1	42	1790
33	38	20	1	42	2159
34	22	30	1	42	1893
35	1	20	0.55	18	1809
36	14	30	0.55	18	1698
37	4	20	0.55	66	2017
38	19	30	0.55	66	1657
39	11	25	0.1	18	1891
40	42	25	1	18	2021
41	45	25	0.1	66	2107
42	27	25	1	66	2170
43	32	25	0.55	42	2173
44	10	25	0.55	42	2152
45	28	25	0.55	42	2092

***** All the activity measurements were carried out in triplicate.

d. ArcticExpress DE3 *E. coli* strain / pET19b expression vector (A19) *

Sample number (Standard Order)	**Randomized Order**	**Temperature (T) (°C)**	**Inducer Concentration (I) (mM)**	**Induction duration (D) (h)**	**Soluble activity ($U.L^{-1}$ of culture)**
1	3	10	0.1	42	1624
2	16	20	0.1	42	1920
3	35	10	1	42	1688
4	7	20	1	42	2005
5	22	10	0.55	18	663
6	27	20	0.55	18	1833
7	13	10	0.55	66	1887
8	8	20	0.55	66	1742
9	45	15	0.1	18	1632
10	20	15	1	18	1649
11	39	15	0.1	66	1946
12	32	15	1	66	2007
13	9	15	0.55	42	1981
14	5	15	0.55	42	2052
15	12	15	0.55	42	2036
16	25	10	0.1	42	1642
17	19	20	0.1	42	1932
18	10	10	1	42	1702
19	2	20	1	42	2001
20	14	10	0,55	18	675
21	1	20	0,55	18	1832
22	21	10	0.55	66	1905
23	31	20	0.55	66	1736
24	4	15	0.1	18	1640

25	24	15	1	18	1647
26	38	15	0.1	66	1935
27	28	15	1	66	2009
28	41	15	0.55	42	2070
29	6	15	0.55	42	2019
30	11	15	0.55	42	2012
31	23	10	0.1	42	1634
32	34	20	0.1	42	1947
33	18	10	1	42	1710
34	29	20	1	42	2000
35	44	10	0.55	18	679
36	17	20	0.55	18	1843
37	15	10	0.55	66	1890
38	30	20	0.55	66	1759
39	42	15	0.1	18	1647
40	36	15	1	18	1645
41	33	15	0.1	66	1901
42	37	15	1	66	1979
43	40	15	0.55	42	2036
44	43	15	0.55	42	2050
45	26	15	0.55	42	2010

***** All the activity measurements were carried out in triplicate.

Table S2: Response surface quadratic model analysis of soluble enzyme production with M15 *E. coli* strain / pQE30 expression vector (M30), with IqExpress *E. coli* strain / pQE30 expression vector (I30), BL21 DE3 *E. coli* strain / pET19b expression vector (B19) and ArcticExpress DE3 *E. coli* strain / pET19b expression vector (A19)

a. Polynomial function regression coefficients

Coded factors [a]	Coded regression coefficients				Standard error				*p*-value [b]			
	M30	**I30**	**B19**	**A19**	**M30**	**I30**	**B19**	**A19**	**M30**	**I30**	**B19**	**A19**
Constant	2073.32	2171.14	2141.13	2029.44	13.31	12.15	9.10	22.89	0.000	0.000	0.000	0.000
T	-141.86	-140.45	-120.87	202.06	8.15	7.44	5.57	14.02	0.000	0.000	0.000	0.000
I	38.98	58.98	58.93	26.83	8.15	7.44	5.57	14.02	0.000	0.000	0.000	0.064
D	89.09	64.88	63.42	221.30	8.15	7.44	5.57	14.02	0.000	0.000	0.000	0.000
T^2	-248.23	-258.41	-214.33	-239.16	12.00	10.95	8.20	20.64	0.000	0.000	0.000	0.000
I^2	29.71	-6.53	41.92	26.79	12.00	10.95	8.20	20.64	0.018	0.555	0.000	0.203
D^2	-193.72	-120.41	-131.03	-253.35	12.00	10.95	8.20	20.64	0.000	0.000	0.000	0.000
T × I	-20.22	2.42	-21.83	0.61	11.53	10.52	7.88	19.83	0.088	0.819	0.009	0.976
T × D	-123.00	-36.07	-59.93	-328.04	11.53	10.52	7.88	19.83	0.000	0.002	0.000	0.000
I × D	13.69	-11.78	-7.82	16.08	11.53	10.52	7.88	19.83	0.243	0.271	0.328	0.423

[a] Factors coded as following : temperature (T), Inducer concentration (I) and culture duration (D).

[b] *p*-value < 0.05 indicates a statistical significance of the factor.

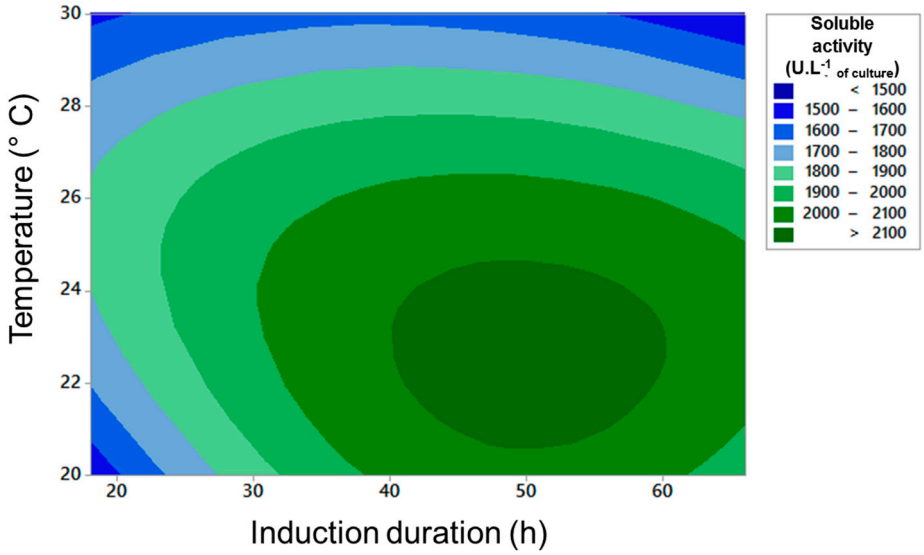
b. Analysis of variance (ANOVA)

Source [a]	Sum of squares				Degree of freedom				*F* value				*p*-value [b]			
	M30	**I30**	**B19**	**A19**	**M30**	**I30**	**B19**	**A19**	**M30**	**I30**	**B19**	**A19**	**M30**	**I30**	**B19**	**A19**
Model	1963026	1532237	1284812	4758653	9	9	9	9	136.81	128.21	191.51	112.09	0.000	0.000	0.000	0.000
T	482971	473401	350658	979862	1	1	1	1	302.93	356.50	470.40	207.73	0.000	0.000	0.000	0.000
I	36465	83497	83355	17275	1	1	1	1	22.87	62.88	111.82	3.66	0.000	0.000	0.000	0.064
D	190478	101023	96520	1175413	1	1	1	1	119.47	76.08	129.48	249.19	0.000	0.000	0.000	0.000
T^2	682550	739644	508860	633549	1	1	1	1	428.12	557.00	682.63	134.31	0.000	0.000	0.000	0.000
I^2	9777	472	19462	7948	1	1	1	1	6.13	0.36	26.11	1.68	0.018	0.555	0.000	0.203
D^2	415704	160588	190188	710970	1	1	1	1	260.74	120.93	255.13	150.73	0.000	0.000	0.000	0.000
T × I	4905	71	5720	4	1	1	1	1	3.08	0.05	7.67	0.00	0.088	0.819	0.009	0.976
T × D	181548	15610	43104	1291336	1	1	1	1	113.87	11.76	57.82	273.77	0.000	0.002	0.000	0.000
I × D	2250	1664	733	3104	1	1	1	1	1.41	1.25	0.98	0.66	0.243	0.271	0.328	0.423
Lack of fit	49275	33761	18529	156098	3	3	3	3	80.54	28.32	26.14	185.12	0.000	0.000	0.000	0.000
Pure error	6526	12715	7561	8994	32	32	32	32								
Total	2018827	1578714	1310903	4923745	44	44	44	44								

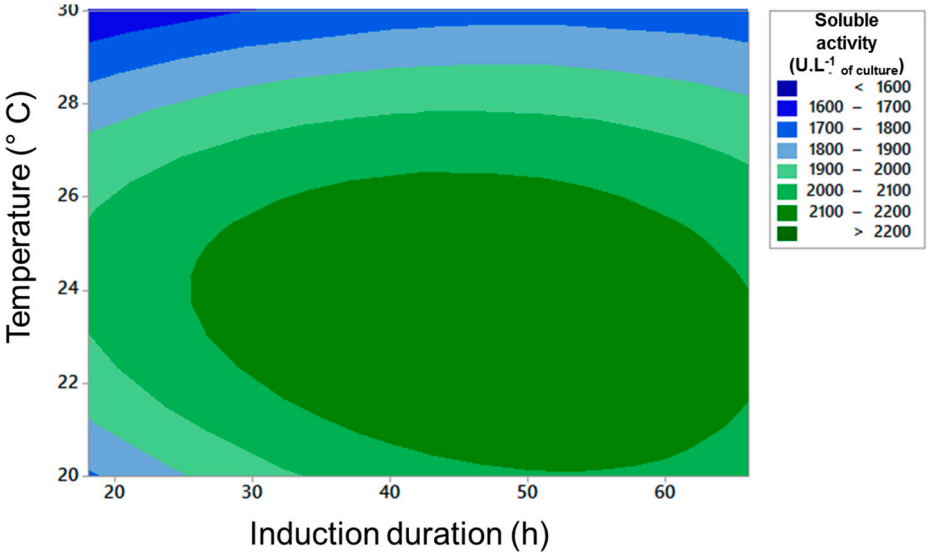
[a] Factors coded as following: temperature (T), Inducer concentration (I) and culture duration (D).
[b] *p*-value < 0.05 indicates a statistical significance of the factor.

Figure S1: Contour plots showing the effects of cross term product factors on the soluble HPL activity level. (Only significant interactions were kept).

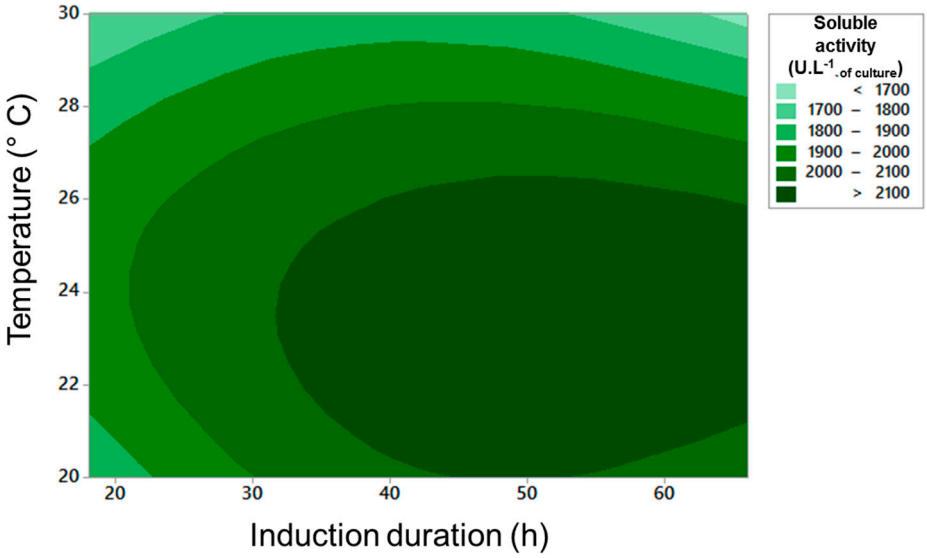
a. Contour plot showing the effects of temperature and induction duration on soluble HPL activity for M30 expression system



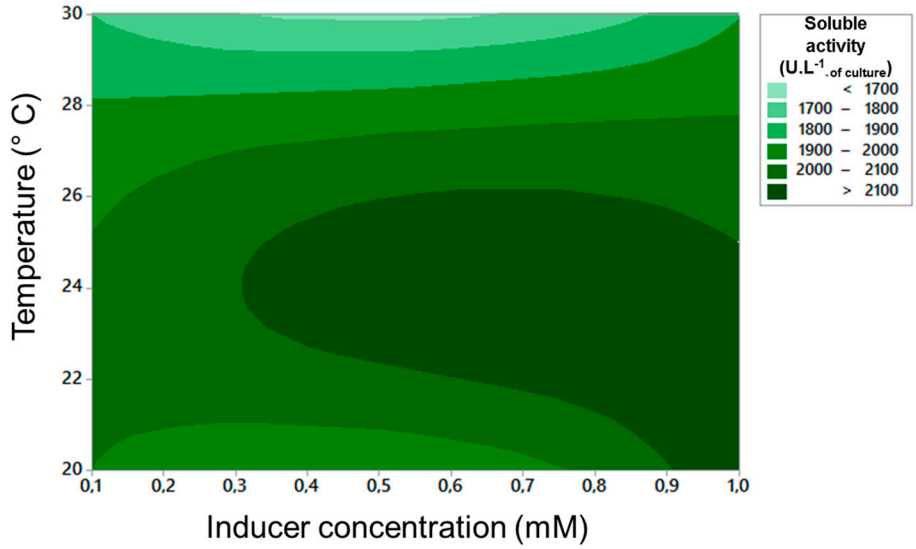
b. Contour plot showing the effects of temperature and induction duration on soluble HPL activity for I30 expression system



c. Contour plot showing the effects of temperature and induction duration on soluble HPL activity for B19 expression system



d. Contour plot showing the effects of temperature and inducer concentration on soluble HPL activity for B19 expression system



e. Contour plot showing the effects of temperature and induction duration on soluble HPL activity for A19 expression system

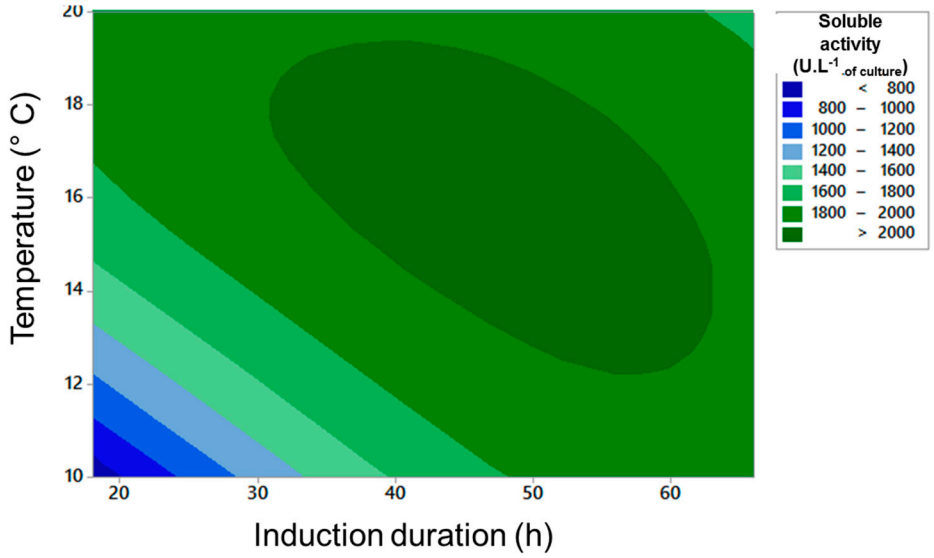


Table S3: *E. coli* strain genotypes used in this study for recombinant protein expression and for cloning experiments.

***E. coli* expression strains**	**Genotype**
M15	*F-, Φ80ΔlacM15, thi, lac-, mtl-, recA+, KmR*
IqExpress	*MiniF lacIq (CamR) / fhuA2 [lon] ompT gal sulA11 R(mcr-73::miniTn10--TetS)2 [dcm] R(zgb-210::Tn10--TetS) endA Δ(mcrCmrr)114::IS10*
BL21 DE3	*fhuA2 [lon] ompT gal (λ DE3) [dcm] ΔhsdS λ DE3 = λ sBamHIo ΔEcoRI-B int::(lacI::PlacUV5::T7 gene1) i21Δnin5*
Arctic Express DE3	*E. coli B, F-, ompT, hsdS, (rB- mB-), dcm+, TetR, gal λ(DE3) endA Hte [cpn10 cpn60 GentR].*