Ginseng extracts modulate mitochondrial bioenergetics of live cardiomyoblasts: a functional comparison of different polar extracts

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Running title: Ginseng extracts modulate mitochondrial bioenergetics of cardiomyoblasts

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Table S1

Mass spectra properties of marker chemicals in ginseng extracts.

Chemical	Formula	Calculated mass [M]	Precursor ion [M-H][1)]	Fragmentor energy	Collison energy	Product ion[2)]
Ginsenoside Rb1	$C_{54}H_{92}O_{23}$	1108.6	1107.6	250	41	945.5
					49	783.5
Ginsenoside Rd	$C_{48}H_{82}O_{18}$	946.5	945.5	250	33	783.5
					45	621.6
Ginsenoside Re	$C_{48}H_{82}O_{18}$	946.5	945.5	250	41	637.5
					53	475.5
Ginsenoside Rg1	$C_{42}H_{72}O_{24}$	800.5	799.5	250	21	637.3
					37	475.4
Astragaloside IV	$C_{41}H_{68}O_{14}$	784.9	829.5[3)]	190	5	829.5
					25	783.2

[1)]The detected chemicals had better responses under the negative mode: the $[M-H]^-$ was used as the precursor ion.
[2)]Two pairs of collision energy and product ions were used for the MRM analysis to guarantee the precision of analytes.
[3)]The precursor ion of astragaloside IV was $[M + HCOOH - H]^-$ under the negative mode.

Table S2

Calibration curves, LOD, LOQ, precision, repeatability and recovery of 4 marker chemicals in ginseng extracts

Ginsenoside	Calibration curve[1)]	Correlation coefficient (r^2)	Range (ng)	LOD (ng)[2)]	LOQ (ng)[3)]	Precision (n = 6)[4)]		Repeatability (n = 5)		Recovery[7)] (n = 3)	
						Intra-day[5)]	Inter-day[6)]	Mean (%)	RSD (%)	Mean (%)	RSD (%)
Rb1	y =76.2x - 293.2	0.9995	2-200	0.329	1.146	1.99	1.72	98.66	1.25	98.66	1.25
Rd	y = 192.2x - 1102.8	0.9994	1-100	0.156	0.632	2.81	2.12	99.87	2.81	99.87	2.81
Re	y = 169.2x - 22.1	0.9992	1-50	0.199	0.651	2.88	2.18	98.19	2.98	98.19	2.98
Rg1	y = 401.1x - 1109.1	0.9991	1-50	0.167	0.631	1.89	1.32	99.19	2.11	99.19	2.11

[1)]The calibration curve was derived from six data points, n = 3.
[2)]LOD refers to the limits of detection.
[3)]LOQ refers to the limits of quantification.
[4)]The value of RSD (%) were presented.
[5)]The intra-day analysis refers to the sample examined for six replicates within one day.
[6)]The inter-day analysis refers to the sample examined in duplicates over three consecutive days.
[7)]Recovery (%) = 100 × (amount found - original amount)/amount spiked. The data were presented as average of three independent determinations, and the SD was < 5% of the mean, which was not shown for clarity.

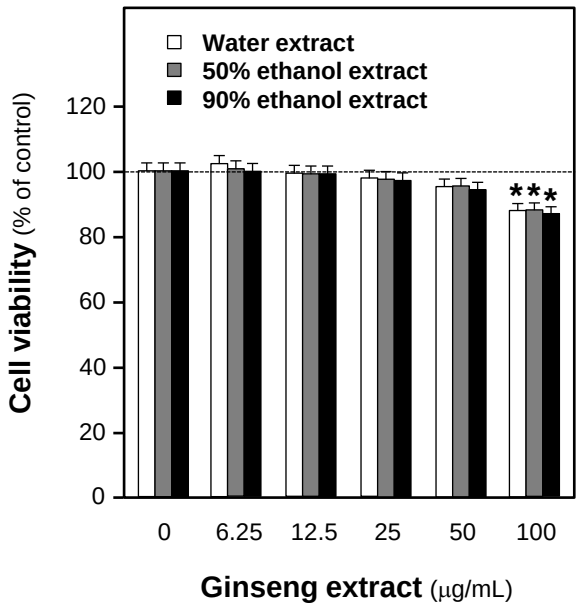
Water extract
50% ethanol extract
90% ethanol extract
Cell viability (% of control)
120
100
80
60
40
20
0
0
6.25
12.5
25
50
100
***
Ginseng extract (μg/mL)

Fig. S1

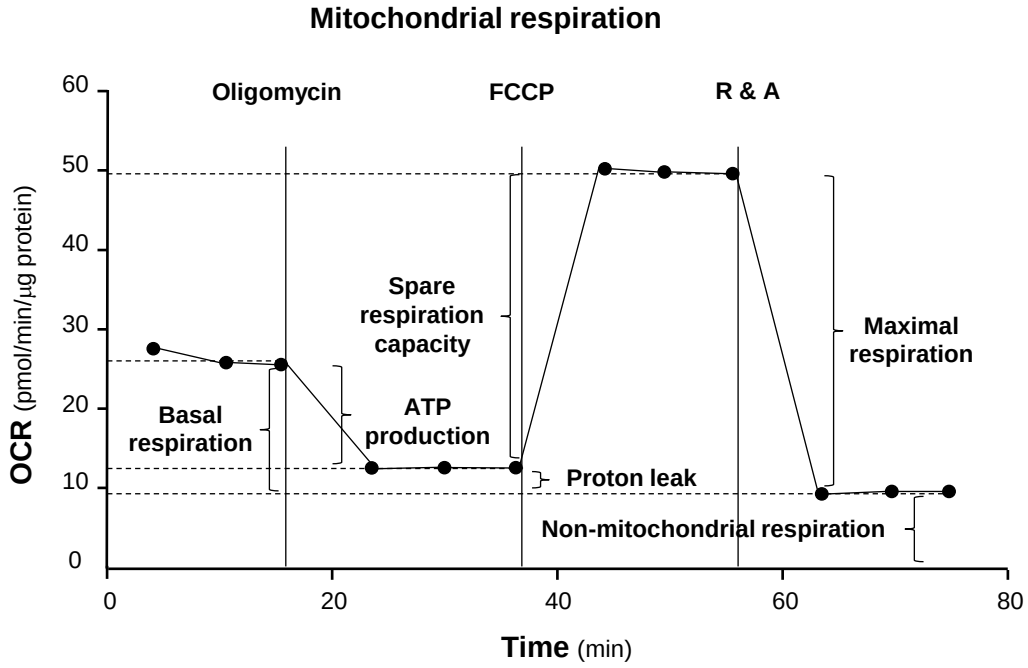
Mitochondrial respiration
Oligomycin
FCCP
R & A
OCR (pmol/min/μg protein)
60
50
40
30
20
10
0
Spare respiration capacity
Maximal respiration
Basal respiration
ATP production
Proton leak
Non-mitochondrial respiration
0
20
40
60
80
Time (min)

Fig. S2

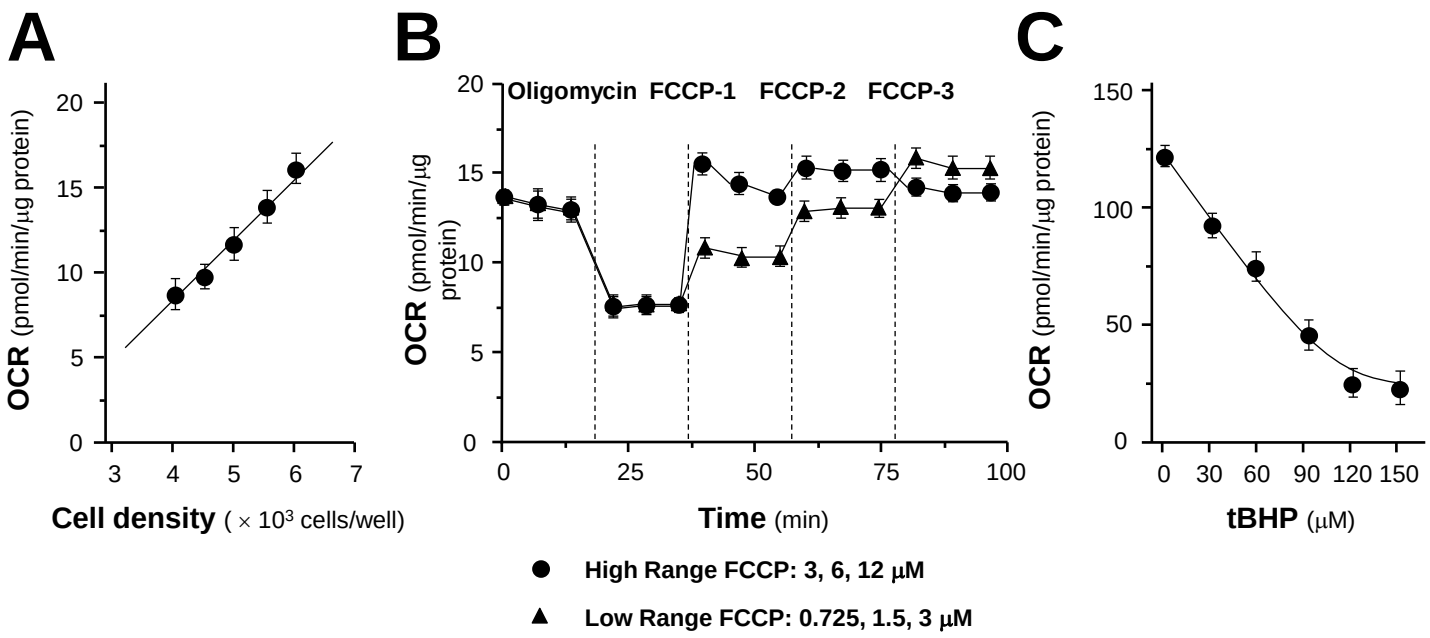
A
B
C
OCR (pmol/min/μg protein)
Cell density (× 10³ cells/well)
Oligomycin FCCP-1 FCCP-2 FCCP-3
OCR (pmol/min/μg protein)
Time (min)
OCR (pmol/min/μg protein)
tBHP (μM)
High Range FCCP: 3, 6, 12 μM
Low Range FCCP: 0.725, 1.5, 3 μM

Fig. S3