

# Bisphenol A (BPA) directly activates the G protein-coupled estrogen receptor 1 and triggers the metabolic disruption in the gonadal tissue of *Apostichopus japonicus*

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**Table S1.** GenBank accession numbers corresponding to the sequence used for phylogenetic tree analysis and multiple sequence alignment.

Gene type	abbreviation	GenBank accession numbers
Estrogen receptor $\alpha$	HsER $\alpha$	NP_000116.2
	MmER $\alpha$	NP_001289460
	CfER $\alpha$	XP_533454
	SsER $\alpha$	NP_999385.1
	GgER $\alpha$	NP_990514.1
	DrER $\alpha$	NP_694491.1
	CaER $\alpha$	XP_026089164.1
	XlER $\alpha$	NP_001083085
	BmER $\alpha$	XP_014332422
	LcoER $\alpha$	NP_001290305
	LcfER $\alpha$	XP_018528117
	HcER $\alpha$	XP_019750588
	CsER $\alpha$	XP_008310919
	OsER $\alpha$	XP_029644783
	LaER $\alpha$	XP_013404576
	OgER $\alpha$	XP_046148097
	MaER $\alpha$	XP_020443618
	CmER $\alpha$	XP_007892594
Estrogen receptor $\beta$	HsER $\beta$	NP_001428
	MmER $\beta$	NP_034287
	CfER $\beta$	XP_038445068
	SsER $\beta$	NP_001001533
	GgER $\beta$	NP_990125
	DrER $\beta$	NP_777287
	CaER $\beta$	XP_026079489
	XlER $\beta$	NP_001124426
	BmER $\beta$	XP_005898314
	LcmER $\beta$	XP_005986456
	LcoER $\beta$	XP_010737659
	LcfER $\beta$	XP_018538032
	HcER $\beta$	XP_019726315
	CsER $\beta$	NP_001281164
	OgER $\beta$	XP_046147746
	MaER $\beta$	XP_020459576
	RtER $\beta$	XP_020380159
G protein-coupled estrogen receptor 1	HsGPER1	NP_001035055
	MmGPER1	NP_084047

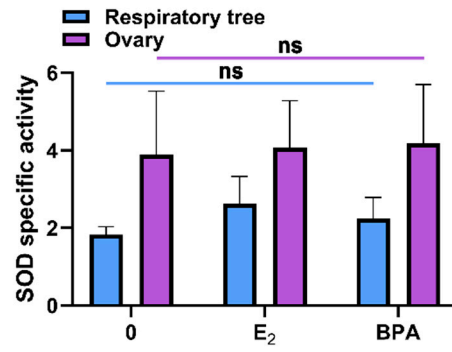
	CfGPER1	XP_005621261
	SsGPER1	XP_003124292
	GgGPER1	NP_001155877
	DrGPER1	NP_001122195
	CaGPER1	XP_026056231
	XIGPER1	XP_018091862
	BmGPER1	XP_014338621
	LcmGPER1	XP_006010235
	LcoGPER1/LcGPER1	XP_010728847
	LcfGPER1	XP_018537294
	HcGPER1	XP_019731372
	CsGPER1	XP_008312438
	OgGPER1	XP_046163069
	MaGPER1	XP_020466193
	CmGPER1	XP_007901691
Kisspeptin1 receptor	HsKISS1R	NP_115940
	MmKISS1R	NP_444474
	SsKISS1R	NP_001038089
	DrKISS1R	NP_001099149
	CaKISS1R	XP_026051331
	XIKISS1R	XP_018080755
	LcoKISS1R	XP_010727548
	LcfKISS1R	XP_018528723
	HcKISS1R	XP_019739218
	CsKISS1R	XP_008332638
	OgKISS1R	XP_046178411
	CmKISS1R	XP_007898035

Table S2. The primers used in the experiments

Primer	Sequence
<i>AjGPER1-F</i>	CTTCAACACAGTTCTTCTGTCAAGA
<i>AjGPER1-R</i>	TGTCGACCTGTATAATCTCGTCAT
<i>q-AjGPER1-F</i>	GTCGTAGCGACTGTCCATATAGTT
<i>q-AjGPER1-R</i>	CATCTTGCCTGTTCTGATACTGACT
<i>β-tublin-F</i>	CACCACGTGGACTCAAAATG
<i>β-tublin-R</i>	GAAAGCCTTACGACGGAACA

Table S3. Multiple sequence alignment

abbreviation	% identity	alignment length	E value
AjGPER1	100.000	398	0.0
HsGPER1	27.891	294	1.59e <sup>-26</sup>
GgGPER1	26.074	326	5.48e <sup>-24</sup>
MmGPER1	25.965	285	1.59e <sup>-23</sup>
DrGPER1	28.045	353	2.20e <sup>-23</sup>
XIGPER1	28.472	288	1.31e <sup>-22</sup>
HcGPER1	27.445	317	1.94e <sup>-20</sup>
MaGPER1	28.615	325	3.99e <sup>-20</sup>
LcGPER1	27.066	351	6.07e <sup>-20</sup>
CmGPER1	25.402	311	6.26e <sup>-19</sup>



**Figure S1.** Effects of 4 h exposure to E<sub>2</sub> or BPA on SOD enzyme activity in the respiratory tree and ovary tissue of *A. japonicus*. Values shown are multiples of the lowest activity of the control group expressed as means  $\pm$  SEM (n = 3). The bars of different colors represent different tissues. Data were analyzed by using one-way ANOVA followed by Tukey's multiple comparison tests (ns  $P > 0.05$ ).