

Supplementary Figure 1. UV triggers synchronization of the circadian clock.

a, **b** NIH3T3 cells harboring Per2-Luc were irradiated with different doses of UV light (254 nm), and the temporal luminescence profile after the irradiation was monitored. Representative Per2-Luc profiles of the initial responses of NIH3T3 cells after different doses of UV irradiation are shown (**a**). Relative Per2-Luc intensity change before and after UV irradiation (**b**). n = 3. *: p<0.05, two-tailed t-test for each group; not significant (p>0.05) unless mentioned. **c** Cell viability of NIH3T3 cells after 36 hours from UV exposure of different strengths calculated from trypan blue staining. n = 3, *: p<0.05, two-tailed t-test for each group; not significant (p>0.05) unless mentioned. **d** Scatter plot of period and amplitude of each profiles in Fig. 1a calculated using Cosinor program.



Supplementary Figure 2. Circadian profiles of core clock gene expression after UV irradiation.

mRNA levels of core clock genes after UV irradiation. NIH3T3 cells were irradiated with UV (10 J m⁻²), and mRNA was collected at 0, 2, 6, 12, 18, 24, 30, 36, 42, 48 and 54 hours after the irradiation. mRNA levels of *BMAL1*, *CLOCK*, *Per1-3* and *Cry1-2* were quantified by qPCR. n = 3, error bar: SD, *: p<0.05, two-tailed t-test against abundance at time 0 for each group; not significant (p>0.05) unless mentioned.



Supplementary Figure 3. HSF1 activation after UV irradiation.

a mRNA levels of HSR-related genes after UV irradiation. NIH3T3 cells were irradiated with UV (10 J m⁻²), and mRNA was collected at 0, 0.5, 1, 2, and 4 hours after the irradiation. mRNA levels of HSF1 and Hsp70 were quantified by qPCR. n = 3, error bar: SD, *: p<0.05, two-tailed t-test for each group; not significant (p>0.05) unless mentioned. **b** NIH3T3 cells harboring HSE-SLR were irradiated with UV (10 J m⁻²), and single-cell HSE-SLR profiles were monitored under a luminescence microscope. Images of representative time points are shown. Each row in the heat map shows the HSE-SLR profile of an individual cell that was tracked over time.



Supplementary Figure 4. HSE2 but not HSE1 of the *Per2* promoter is transcriptionally responsive after UV irradiation.

NIH3T3 cells were transfected with reporter vectors containing luciferase driven by HSE sequences of the mouse *Per2* promoter. Each HSE site was mutated to a non-HSE consensus sequence to make mutated HSE reporters. The HSE1 mutation probe corresponds to a reporter with mutated HSE1, and the HSE2 mutation probe corresponds to a reporter with mutated HSE2. Peak intensity after UV irradiation was quantified. n = 3 *: p<0.05, ns: not significant (p>0.05), two-tailed t-test.



Supplementary Figure 5. p53 target gene expression is repressed in HSF1-/- cells after UV irradiation. The expression levels of p53 target genes, *Mdm2*, *Bax*, *Bbc3* and *Cdkn1a*, in wild-type and HSF1-/- cells were measured by qPCR. Cells were irradiated with UV (10 J m⁻²) and subjected to qPCR at 0 and 8 h post stimulation. n = 3, *: p<0.05, ns: not significant (p>0.05), two-tailed t-test.



Supplementary Figure 6. p53 deficiency does not affect UV-triggered activation of HSF1.

a-d Dose-dependent induction of HSF1 activity after UV irradiation was monitored by MEF cells harboring the HSE-SLR reporter. Schematic diagram of HSE-SLR reporter (**a**). Dose-dependent induction of HSF1 activity after UV irradiation monitored by wild-type (**b**) and p53-/- MEFs (**c**) harboring the HSE-SLR reporter. Comparison of the dose-dependent p53RE activation between wild-type and p53-/- MEFs (**d**). Luminescence intensity at 0 h and 8 h (the first peak) post UV irradiation was used for the quantification. n = 3, ns: not significant (p>0.05), two-tailed t-test. **e** The negative control for immunoprecipitation assays using mouse-IgG, corresponding to Fig. 4d and 5a. NIH3T3 cells were stimulated with UV irradiation (254 nm, 10 J m⁻²) and immunoprecipitated with mouse-IgG at 2 or 4 hours after stimulation. Representative blots for immunoblotting assay against BMAL1, HSF1 and p53 are shown. See Supplementary Figure 16 for full-size blot images.



Supplementary Figure 7. Hierarchal regulation of BMAL1-HSF1-p53.

a Synchronization of Per2-Luc after UV irradiation (10 J m⁻²) in U2OS cells. Raw values were detrended to emphasize circadian oscillation. **b-d** Split-luciferase complementation assay to monitor BMAL1–HSF1 and HSF1–p53 interactions. Averaged luminescence profiles of reconstituted luciferase for BMAL1–HSF1 (**b**) or HSF1–p53 (**c**) interactions after UV irradiation. Quantification of first peak time after the irradiation (**d**). n = 3, *: p<0.05, two-tailed t-test.



Time post Dex treatment [h]

Supplementary Figure 8. (Corresponds to Fig. 6) Reporter profiles with UV irradiation at various CTs. a All measured Per2-Luc profiles upon UV irradiation at various CTs used for plotting the PRC and the PTC (Corresponds to Fig. 6a-c). Dex-synchronized NIH3T3 cells were irradiated with UV (10 J m⁻²) at 24-48 hours (CT 0-24 h) post Dex treatment. The color of each vertical line corresponds to the time at which cells were irradiated; thereafter, each Per2-Luc profile, each a corresponding color, is shown. **b** All measured HSE-SLR profiles upon UV irradiation at various CTs used for the calculation of the first peak time and their peak intensity in Fig. 6d. Dex-synchronized NIH3T3 cells were irradiated with UV (10 J m⁻²) at 24-48 h (CT 0-24 h) post Dex treatment. The color of each vertical line corresponds to the time at which cells were irradiated; thereafter, each a correspond to the time at which cells were irradiated with UV (10 J m⁻²) at 24-48 h (CT 0-24 h) post Dex treatment. The color of each vertical line corresponds to the time at which cells were irradiated; thereafter, each NIH3T3 cells were irradiated with UV (10 J m⁻²) at 24-48 h (CT 0-24 h) post Dex treatment. The color of each vertical line corresponds to the time at which cells were irradiated; thereafter, each HSE-SLR profile, each a corresponding color, is shown. **c** All measured p53RE/E-box-Luc profiles upon UV irradiation used for the calculation of the first peak time and their peak intensity in Fig. 6e. Dex-synchronized NIH3T3 cells were irradiated with UV (10 J m⁻²) at 24-48 h (CT 0-24 h) post Dex treatment. The color of each vertical line corresponds to the time at which cells were irradiated; thereafter, each p53RE/E-box-Luc profile, each a corresponding color, is shown.



Supplementary Figure 9. Reversible inhibition of HSF1 at the onset of UV irradiation impairs the Per2-Luc surge in a time-dependent manner.

Per2-Luc profiles upon UV irradiation at various CTs with or without transient HSF1 inhibition by 100 μ M KNK437. Dex-synchronized NIH3T3 cells were irradiated with UV (10 J m⁻²) at 24-48 h (CT 0-24 h) post Dex treatment. KNK437 was added to the medium 15 min before the stimulation, and the medium was replaced with fresh medium 10 min after the irradiation. The first peak time after the irradiation was calculated, and the effect of KNK437 was compared to that of the untreated sample. Error bar: SD, n = 3, two-tailed t-test, *: p<0.05; not significant (p>0.05) unless mentioned.

Heat-shock response



DNA damage response



Circadian related



Oxidative stress response





Pmaip1 Pmaip2 Casp3 Casp7 Casp7 Casp7 Casp7 Casp7 Casp6 Casp7 Casp





Supplementary Figure 10. Expression profiles of genes related to the clock and stress response pathways affected by UV irradiation.

Gene expression profiles of UV (20 J m⁻²)-treated MEF cells reanalyzed from datasets from the microarray database. Values corresponding to the log2 fold change compared to those of the non-stimulated sample are color-coded from red, indicating increased expression, to blue, indicating decreased expression after UV stimulation. Gene expression profiles of apoptosis-related, cell-cycle-related, circadian-related, HSR, oxidative stress response and DNA damage response pathways extracted from the dataset are shown.



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Supplementary Figure 11. Similarly, or differentially regulated clock and stress-responsive genes after UV and oxidative stress exposure-induced clock synchronization.

Comparison of the gene expression profiles of UV (20 J m⁻²)-treated MEFs as in Supplementary Figure 10 and hydrogen peroxide-treated (ROS; 5 mM, 10 min) NIH3T3 cells. Genes with a fold change of at least 1.5 were defined as "responsive genes" for each condition. **a** Circadian and several stress response-related transcripts were extracted and depicted as a heat map of the log2 fold change compared to that of the non-stimulated sample. Expression profiles at 3 h or 6 h post UV stimulation and 4 h post ROS stimulation are shown. Among these responsive genes, genes whose expression increased in a similar pattern post UV and ROS stimulations, those whose expression increased only in response to UV stimulation, and those whose expression increased only in response to ROS stimulations were clustered respectively (up arrow indicates upregulated genes). **b** Venn diagram showing the number of responsive genes that were increased similarly or differentially in each stimulation condition. The number in each area shows the number of genes associated with each category. Gene names for transcripts that were upregulated by both stimulations (at least 1.5-fold change at either 3 h or 6 h post UV stimulation) are shown below the Venn diagram. Genes that are considered to be important for this study are indicated in bold characters. Red and green colored gene names indicate HSF1 regulatory or p53 regulatory genes, respectively.



Supplementary Figure 12. UV-induced signaling pathways and their respective expression profiles.

Pathway maps representing UV stress-responsive expression genes for circadian, HSR, DNA damage-related, apoptosis-related, cell-cycle-related and anti-oxidative pathways. Pathway maps were generated using PathVisio software with maps adopted from WikiPathways. Arrow indicates signaling pathways hypothesized to be activated upon UV irradiation. Gene names are color-coded with the time course fold change after UV irradiation. Arrows indicate mutual control among these pathways.



Supplementary Figure 13.

Full sized immunoblots corresponding to Fig. 1d.



Supplementary Figure 14.

Full sized immunoblots corresponding to Fig. 4d. 'load' denotes that the bands are used for the loading control. The sign "X" on the top of the bands denotes that these bands were not used in this study.



Supplementary Figure 15.

Full sized immunoblots corresponding to Fig. 5a. 'load' denotes that the bands are used for the loading control. The sign "X" on the top of the bands denotes that these bands were not used in this study.





Supplementary Figure 16.

Full sized immunoblots corresponding to Supplementary Figure 6e.

Supplementary Table 1. Primers used for the qPCR assay.

Primers used in Supplementary Figure 2.

mPeriod1 Forward	TGA AGC AAG ACC GGG AGA G
mPeriod1 Reverse	CAC ACA CGC CGT CAC ATC A
mPeriod2 Forward	GAA AGC TGT CAC CAC CAT AGA A
mPeriod2 Reverse	AAC TCG CAC TTC CTT TTC AGG
mPeriod3 Forward	AAA AGC ACC ACG GAT ACT GGC
mPeriod3 Reverse	GGG AGG CTG TAG CTT GTC A
mCry1 Forward	CAC TGG TTC CGA AAG GGA CTC
mCry1 Reverse	CTG AAG CAA AAA TCG CCA CCT
mCry2 Forward	CAC TGG TTC CGC AAA GGA CTA
mCry2 Reverse	CCA CGG GTC GAG GAT GTA GA
mBmall Forward	ACA GTC AGA TTG AAA AGA GGC G
mBmall Reverse	GCC ATC CTT AGC ACG GTG AG
mClock Forward	CTT CCT GGT AAC GCG AGA AAG
mClock Reverse	GTC GAA TCT CAC TAG CAT CTG AC

Primers used in Supplementary Figure 3.

mHSF1 Forward	ACT CCA ACC TGG ACA ACC TG
mHSF1 Reverse	GGA GGC TCT TGT GGA GAC AG
mHsp70 Forward	CCC TCA GGA ATC CGT ACT C
mHsp70 Reverse	AAT CAG CGA GCC GGA GGA G

Primers used in Supplementary Figure 5.

mMdm2 Forward	GGA TCT TGA CGA TGG CGT AAG
mMdm2 Reverse	AGG CTG TAA TCT TCC GAG TCC
mBax Forward	TGA AGA CAG GGG CCT TTT TG
mBax Reverse	AAT TCG CCG GAG ACA CTC G
mBbc3 Forward	TTC TCC GGA GTG TTC ATG C
mBbc3 Reverse	TAC AGC GGA GGG CAT CAG
mCdkn1a Forward	CCT GGT GAT GTC CGA CCT G
mCdkn1a Reverse	CCA TGA GCG CAT CGC AAT C

Supplementary Table 2. Primers used for the ChIP-qPCR assay.

p53RE/E-box2 Forward	CAG GTT CCG CCC CGC CAG TAT
p53RE/E-box2 Reverse	GTC GCC CTC CGC TGT CAC ATA G
HSE1 Forward	GCC TCC TTT CCA TTC CTG
HSE1 Reverse	GGA GAA GGC AAG CTT GTC
HSE2 Forward	GAA GAC GTG ACA AGC TTG C
HSE2 Reverse	CTG TCC AAA GGG TCA AAG G
E-Box5 Forward	CTC TGT AGG GTG GAG CGG CGA
E-Box5 Reverse	ATC CCC ACT GCT CCT TCG CAC
Mdm2 Forward	GTT GAC TCA GCT CTT CCT GTG G
Mdm2 Reverse	GGC TGC GGA AAC GGG GCA GCG

Supplementary Table 3.

List of p values

Fig. 1d Protein abundance change of HSF1 and Hsp70 after UV irradiation, compared to time 0 h.

HSF1		Hsp70	
Hour	p value	Hour	p value
2	0.030682 *	2	0.026439 *
8	0.039357 *	8	0.144687
14	0.130678	14	0.101302
20	0.091954	20	0.375521
26	0.007278 *	26	0.13332
32	0.137196	32	0.06001
38	0.350130	38	0.135767
44	0.159351	44	0.304443
50	0.045099 *	50	0.352204

Fig. 2c Fold induction of mutated Per2-Luc reporter after UV irradiation, compared to unstimulated sample.

	p value
Native	0.021948 *
HSE1	0.014618 *
HSE2	0.055106

Fig. 2d ChIP assay of HSE1 and HSE2 sequence on Per2 promoter after UV irradiation, compared to unstimulated sample.

		p value	
HSE1	HS	0.44467	
	UV	0.72989	
LIGES	HS	0.03536 *	
IISE2	UV	0.03690 *	

Fig. 3b First peak time of Per2-Luc after UV irradiation compared to wild-type.

	p value
p53-/-	0.003275 *

Fig. 3d ChIP assay of p53RE on Mdm2 or Per2 promoter after UV irradiation, compared to unstimulated

sample.

	p value
Mdm2	0.018418 *
E-Box2	0.040913 *
E-Box5	0.378366

Fig. 4c Analysis of p53RE/Luc peak intensity after UV irradiation between wild-type and HSF1-/- MEFs.

Strength [J m ⁻²]	p value
2	0.296354
5	5.08×10 ⁻⁵ *
10	0.002541 *
15	0.007382 *

Fig. 4e Coimmunoprecipitation assay of HSF1-p53 interaction, compared to the band intensity at time 0 h.

p53–HSF1		HSF1-	-p53
Hour	p value	Hour	p value
2	0.0102680 *	2	0.014769 *
4	0.0188642 *	4	0.010246 *

Fig. 4h Split-luciferase complementation assay of HSF1-p53 interaction, compared to unstimulated sample.

Strength [J m ⁻²]	p value
2	0.004568 *
5	0.002999 *
10	0.000896 *

Fig. 5b Coimmunoprecipitation assay of BMAL1-HSF1 interaction, compared to band intensity at time 0 h.

HSF1–BMAL1		BMAL1-HSF1	
Hour	p value	Hour	p value
2	0.007393 *	2	0.025951 *
4	0.032426 *	4	0.038298 *

Fig. 5e Analysis of HSE-SLR peak intensity after UV irradiation between wild-type and BMAL1-/- MEFs.

Strength [J m ⁻²]	p value
2	0.031158 *
5	0.026923 *
10	0.011466 *

Fig. 5h Analysis of p53RE/Luc peak intensity after UV irradiation between wild-type and BMAL1-/- MEFs.

Strength [J m ⁻²]	p value
2	0.194965
5	0.000178 *
10	0.000561 *

Fig. 5i Cell viability after different dose of UV irradiation in wild-type and null-mutant MEFs.

p value compared to "wild-type".

	Cell type		
Strength [J m ⁻²]	HSF1-/-	BMAL1-/-	p53-/-
No stimulation	0.057191	0.235447	0.092735
2	0.928389	0.156726	0.183503
5	0.911955	0.825922	0.67925
10	0.069051	0.098097	0.094539
20	0.046346	0.147987	0.02286
30	0.015598	0.029113	0.009501
50	0.019804	0.014548	0.016356
100	0.016932	0.299699	0.378003

p value compared to "No stimulation".

	Cell type			
Strength [J m ⁻²]	Wild-type	HSF1-/-	BMAL1-/-	p53-/-
2	0.020204	0.723974	0.434083	0.008163
5	0.026148	0.337734	0.422649	0.275931
10	0.041577	0.109129	0.154999	0.062333
20	0.029286	0.036817	0.132528	0.009852
30	0.018829	0.011389	0.023958	0.004827
50	0.014059	0.010788	0.007878	0.002748
100	0.011315	0.00249	0.022261	0.009398

Fig. 6d Analysis of circadian time-dependency of HSE-SLR reporter.

p value

	0-3 h	4-7 h	8-11 h	12-15 h	16-19 h	20-23 h
0-3 h	\mathbf{i}	0.026964	0.435501	0.838656	0.601761	0.820042
4-7 h		\searrow	0.012786	0.402869	0.089758	0.938245
8-11 h			\mathbf{i}	0.628982	0.276719	0.761276
12-15 h				\mathbf{i}	0.994837	0.863645
16-19 h					\searrow	0.860903
20-23 h						\mathbf{i}

Fig. 6e Analysis of circadian time-dependency of p53RE/Luc reporter.

p value

	0-3 h	4-7 h	8-11 h	12-15 h	16-19 h	20-23 h
0-3 h	$\overline{}$	0.006281	0.079596	0.127815	0.909733	0.821556
4-7 h		\mathbf{i}	0.372077	0.099518	0.015526	0.025017
8-11 h			\mathbf{i}	0.618666	0.075248	0.084441
12-15 h				/	0.119359	0.133658
16-19 h						0.902921
20-23 h						$\overline{\}$

Fig. 6f Circadian time-dependent ChIP assay of HSE on Per2 promoter.

ANOVA for % input of HSF1

	F value	p value
UV irradiated	16.6	1.2×10 ⁻⁶
No stimulation	4.2	0.014

HSF1

p value for circadian time comparison.

	0 h	6 h	12 h	18 h
0 h		0.131069	0.0001598	0.0001778
6 h		\backslash	0.0044577	0.007475
12 h			\backslash	0.743352
18 h				\backslash

p value for effect of UV irradiation.

0 h	0.0014378 *
6 h	0.0098022 *
12 h	0.3078513
18 h	0.3168514

IgG p value for circadian time comparison

-				
	0 h	6 h	12 h	18 h
0 h	\mathbf{i}	0.066335	0.597258	0.139604
6 h		\mathbf{i}	0.310401	0.689109
12 h			$\overline{\}$	0.469887
18 h				$\overline{\}$

p value for effect of UV irradiation.

0 h	0.4140059
6 h	0.4570973
12 h	0.2886741
18 h	0.8128733

Fig. 6g Circadian time-dependent ChIP assay of p53RE on Per2 promoter.

ANOVA for % input of p53

	F value	p value
UV irradiated	0.30	0.83
No stimulation	1.67	0.17

<u>p53</u>

p value for circadian time comparison.

	0 h	6 h	12 h	18 h
0 h	$\overline{\}$	0.691956	0.079723	0.629479
6 h		\backslash	0.085974	0.418027
12 h			$\overline{\}$	0.077741
18 h				\mathbf{i}

p value for an effect of UV irradiation.

0 h	0.046336 *
6 h	0.049214 *
12 h	0.073879
18 h	0.419613

IgG

p value for circadian time comparison.

	0 h	6 h	12 h	18 h
0 h	\mathbf{X}	0.478605	0.282845	0.300022
6 h		\backslash	0.571548	0.621415
12 h			\mathbf{i}	0.990628
18 h				\mathbf{X}

p value for an effect of UV irradiation.

0 h	0.513813
6 h	0.435149
12 h	0.070892
18 h	0.042677 *

Supplementary Figure 1b Dose-dependent fold induction of Per2-Luc intensity compared to unstimulated sample.

Strength [J m ⁻²]	p value
2	0.031152 *
5	0.002807 *
10	0.002218 *
15	0.023812 *
20	0.008170 *
30	0.016121 *
50	0.072485 *
100	0.365545 *

Supplementary Figure 1c Cell viability after different dose of UV irradiation, compared to unstimulated sample.

Strength [J m ⁻²]	p value
2	0.166364
5	0.011143 *
10	0.29563
15	0.793317
20	0.049203 *
30	0.018209 *
50	0.012321 *
100	0.008517 *

Supplementary Figure 2 Circadian fluctuation of core clock genes.

ANOVA

	F value	p value
Perl	8.061	0.00115 *
Per2	3.57	0.03240 *
Per3	5.112	0.00923 *
Cryl	1.548	0.235
Cry2	4.178	0.01980 *
Clock	4.14	0.02040 *
Bmall	5.686	0.00594 *

p values for two tailed t-test.

t-test p value Hour Clock Bmal1 Perl Per2 Per3 Cry1 Cry2 0.994798 0.1690685 2 8.03658×10⁻⁵ 0.0341623 0.0005858 0.6738449 0.1510075 6 0.6811744 0.024118 0.9172012 0.8649320 0.1362905 0.0568834 0.0140846 12 0.0289605 0.7773849 0.0119116 0.121244 0.0016522 0.9134892 0.0016118 18 0.0556526 0.0857629 0.146486 0.0765497 0.0109139 0.0590109 0.0052727 24 0.0308640 0.0164572 0.311734 0.0225892 0.0775266 0.0323833 0.0267052 30 0.0001832 0.01398190.232725 0.0248494 0.00075090.0071302 0.0092821 36 0.0059255 0.361294 0.0004234 0.0077292 0.0028809 0.0430308 0.0031816 42 3.53998×10⁻⁵ 0.0460458 0.054283 0.0094175 0.0006157 0.0208516 0.003247648 0.0211079 0.0074227 0.006070 0.0021239 0.0017623 0.0011975 0.0311170 54 0.00720860.0025513 0.974213 0.003520 0.0264419 0.0025602 0.0052761

Supplementary Figure 3a Expression of Hsf1 and Hsp70 after UV irradiation, compared to unstimulated sample.

Hsfl		Hsp70	
	p value		p value
0.5 h	0.165337	0.5 h	0.021763 *
1 h	0.046716 *	1 h	0.039199 *
2 h	0.455541	2 h	0.258675
4 h	0.472773	4 h	0.947661

Supplementary Figure 4 Analysis of fold induction of mutated Per2HSE-Luc reporter after UV irradiation,

Strength [J m ⁻²]	Reporter	p value
2	HSE1 mutation	0.259346
Z	HSE2 mutation	0.251643
5	HSE1 mutation	0.820035
5	HSE2 mutation	0.257982
10	HSE1 mutation	0.943489
10	HSE2 mutation	0.022243 *

compared to the response in wild-type reporter.

Supplementary Figure 5 p53 target gene expression in wild-type and HSF1-/- MEFs after UV irradiation,

compared to unstimulated sample.

0 h		8 h	
	p value		p value
Mdm2	0.879744	Mdm2	0.169397
Bax	0.809252	Bax	0.431081
Bbc3	0.143847	Bbc3	0.036769 *
Cdkn1a	0.453971	Cdkn1a	0.045296 *

Supplementary Figure 6d Analysis of HSE-SLR peak intensity after UV irradiation between wild-type and

p53-/- MEFs.

Strength [J m ⁻²]	p value
2	0.939721
5	0.91342
10	0.35645
15	0.547087

Supplementary Figure 7 First peak time of BMAL1-HSF1 and HSF1-p53 interaction monitored by splitluciferase complementation assay after UV irradiation.

p value 0.034102 *

Supplementary Figure 9 Analysis of the effect of reversible inhibition of Per2-Luc at different circadian times.

CT	p value
0 h	0.783705
6 h	0.046774 *
12 h	0.034578 *
18 h	0.183503