Supplemental Material

Online Methods

Cell Culture

Mouse embryonic stem cells are cultured in DMEM (high glucose) with 15% FBS and supplemented with ß-mercaptoethanol (100µM), non-essential amino acids (100 µM), leukemia inhibitory factor (LIF; 1000U/ml) and penicillin/streptomycin (50ug/ml each). Mouse embryonic fibroblasts (MEF) are cultured in DMEM with 10% FBS, Non-essential amino acids, (100 µM) and penicillin/streptomycin (50ug/ml each). H9c2 myoblasts are maintained in DMEM supplemented with 10% FBS and pen/strep. Human umbilical vein endothelial cells (HUVECs) are maintained in EGM-2 Bullet Kit (CC-3162) contains one 500 ml bottle of Endothelial Cell Basal Medium-2 and the following growth supplements: Hydrocortisone, hFGF-B, VEGF, R3-IGF-1, Ascorbic Acid, Heparin, FBS, hEGF, GA-1000 with is 2% FBS. CPCs from syngeneic male FVB mice are cultured in cardiac stem cell media [1] comprising of DMEM/F12 supplemented with 10% FBS, basic fibroblast growth factor (bFGF), leukemia inhibitory factor (LIF), epidermal growth factor (EGF) and penicillin/streptomycin/glutamine solution. CPCs were differentiated as previously described[2] with 10^{-8} mol/L dexamethasone treatment for 7days.

Exosome purification

mES and MEF cells are cultured for 40 hours, and then exosomes were collected and ultrapurified as described previously [3]. Briefly, the cells and conditioned media are separated by centrifugation (800*g* for 5 minutes); the conditioned media is clarified by centrifugation (14,000*g* for 20 minutes) and the exosomes are collected by ultracentrifugation (100,000*g* for 1 hour) on a 30% sucrose-D2O solution (density ~1.127g/cm3), washed in PBS and pelleted. The purified exosome fraction is re-suspended in PBS for use.

Electron microscopy

Cells are fixed with 4% paraformaldehyde and 1% glutaraldehyde in 0.1 M sodium cacodylate buffer (pH 7.2) (Electron Microscopy Sciences, Hatfield, PA, USA) for 3 hours at room temperature, washed with cacodylate buffer, post fixed in 1% osmium tetroxide, progressively dehydrated in a graded ethanol series (50–100%), and embedded in Epon. Thin (1-mm) and ultrathin (70- to 80-nm) sections were cut from the polymer with a Reichert (Depew, NY, USA) Ultracut S microtome, placed on copper grids, and briefly stained with uranyl acetate and lead citrate. Exosomes are fixed with 2% paraformaldehyde, loaded on 300-mesh formvar/carboncoated electron microscopy grids (Electron Microscopy Sciences, PA, USA), post-fixed in 1% glutaraldehyde, and then contrasted and embedded as described previously [3]. Transmission electron microscopy images are obtained with an FEI (Hillsboro, OR, USA) Tecnai Spirit G2 transmission electron microscope operating at 120 kV.

CyQuant and Metabolic assay

CyQuant and 3-(4,5-Dimethylthiazole-2-yl)-2,5-diphenyltetrazolium bromide assay of CPCs are performed by plating cells in quadruplicate (2000 cells/well) in 96-well plates, followed by incubation with CyQuant (Invitrogen, CA, USA) or 3-(4,5-Dimethylthiazole-2-yl)-2,5-diphenyltetrazolium bromide reagent (Sigma, Mo, USA) as previously described [4].

Matrigel tube formation assay

CPCs treated with exosomes and miRNA mimics are starved overnight prior to the Matrigel assay in CPC medium. Starved CPCs were then seeded in 48-well plate coated with growth factor-reduced Matrigel (BD Biosciences, CA, USA). Tube formation was examined by phase-contrast microscopy 6hrs and 24 hrs later.

Dynamic light scattering

Exosomes are suspended in phosphate-buffered saline (PBS) containing 2 mM ethylenediaminetetraacetic acid (EDTA); then, dynamic light-scattering measurements are performed with a Zetasizer Nano ZS (Malvern Instruments Ltd, Worcestershire, UK) as described previously[5]. Intensity, volume, and distribution data for each sample are collected on a continuous basis for 4 minutes in sets of three. At least three different measurements from three different samples are performed for each exosome population.

Western blotting

Western blot analysis is carried out as previously described [6]. Briefly, Cells or purified exosomes are lysed with 0.1M Tris, 0.3 M NaCl, 0.1% SDS, 0.5% sodium deoxycholate, and 1% Triton X-100 in a cocktail of antiproteases (Sigma-Aldrich Corporation, St. Louis, MO, USA); then, the nuclei and membranes are cleared by centrifugation (15,000*g* for 10 minutes). Protein extracts are separated on an 8% SDS-PAGE gel, blotted on Immobilon (Millipore, Billerica, MA, USA) with TSG101 (4A10; Abcam Inc.), and visualized with enhanced chemoluminescence substrate (Thermo Fisher Scientific, Rockford, IL, USA). Images are acquired with a Chemidoc XRS (Kodak, Rochester, NY, USA).

Real-Time Quantitative reverse transcription Polymerase chain reaction

Total RNA is isolated from frozen heart or cultured cells using RNA-stat and reverse-transcribed into cDNA using iScript cDNA Synthesis kit (Bio-Rad, CA, USA). Quantitative reverse transcriptase polymerase chain reaction is performed on all samples in triplicate using iQ SYBR Green (Bio-Rad, CA, USA) according to the manufacturer's instructions. Primer sequences are provided in Online Table I.

Flow Cytometry

Cell death is measured by Annexin V staining (BD Biosciences, CA, USA) according to the manufacturer's instructions. Cell cycle analysis with propidium iodide (PI, BD Biosciences, CA, USA) is performed by fixing the cells in cold ethanol for 15min followed by labeling with PI for 40min at 37°C. Cytometry is performed using a BD LSRII Flow Cytometer (BD Biosciences, CA, USA).

TaqMan® MicroRNA Array

Single-stranded cDNA is synthesized from all samples using the TaqMan® MicroRNA Reverse Transcription Kit (Part Number 4366593) and the Megaplex™ RT Primers, Rodent Pool Set v3.0 (Part Number 4444746) as described in the Applied Biosystems protocol "Megaplex™ Pools for microRNA Expression Analysis (Part Number 4399721 Rev. C). The reverse transcription product is pre-amplified using Megaplex™ PreAmp Primers, Rodent Pool B v3.0 (4444308). The pre-amplified product is used to run real time PCR reactions using TaqMan® Universal PCR Master Mix, No AmpErase® UNG (Part Number 4324018) on a TaqMan® Array Rodent MicroRNA A+B Cards set v3.0 (Part Number 4444909). The array cards are run on a 7900HT system.

MicroRNA treatment and quantification

Cells are transfected with mouse miR-291a-5p, miR-294-3p, miR-295-3p (mimics) or negative control mimics. CPCs are grown in DMEM/F12 media without antibiotics and transfected with either miRNA mimics or controls (25nM, Invitrogen, CA, USA) using Lipofectamine RNAiMAX (Invitrogen, CA, USA) for 24 hrs as per manufacturer instructions.

Total RNA from the CPCs and heart tissue is extracted using the miRNeasy Mini Kit (Qiagen) according to the manufacturer's protocol including a DNase step. RNA concentrations are verified on the NanoDrop Spectrophotometer (NanoDrop, Thermo Scientific, DE, USA). Equal amount of RNA (5ng) is reverse transcribed using the Taqman MicroRNA Reverse Transcription Kit (Applied Biosystems, CA, USA) using a specific miRNA primer to generate cDNA for use with individual Taqman MicroRNA Assays (Applied Biosystems, CA, USA). Real time Reactions are performed in triplicate on a 7500FAST Real-Time PCR system (Applied Biosystems, CA, USA). Ct values are averaged and normalized to snoRNA234. Relative expression is determined by ΔΔCt comparative threshold method.

Neonatal Rat Cardiomyocyte Isolation

Neonatal rat cardiomyocytes (NRCM) are prepared by enzymatic digestion of hearts obtained from newborn (0–2 day old) Sprague–Dawley rat pups using percoll gradient centrifugation and plated on six-well cell culture grade plates (coated with collagen IV) at a density of 0.85 × 106 cells/well in DMEM/M199 medium and maintained at 37°C in humid air with 5% $CO2$[7].

Animal Studies

All mice (C57BL/6; 8-12 weeks old) used in this study were obtained from The Jackson Laboratories (Bar Harbor, ME). All surgical procedures and animal care protocols were approved by the Temple University Animal Care and Use Committee.

Induction of Acute Myocardial Infarction and Injections

Myocardial infarction. Mice underwent surgery to ligate the left anterior descending coronary artery as reported previously [8] followed by administration of exosomes from mES cells (n=6) and MEF (n=6) cells suspended in saline intramyocardially into the left ventricular wall (border zone) at two different locations immediately after left anterior descending ligation. The saline group underwent the same surgery but received saline without exosomes (n=6). Tissue was harvested at 5 days, 2, 4 and 8weeks after AMI for histological analysis.

Acute myocardial infarction (AMI) was induced as described previously [7, 8]. Briefly, mice are anesthetized, orally intubated and placed in a supine position. Respiration is controlled by mechanical ventilation using a rodent ventilator (Nemi Scientific, Inc., Framingham, MA) with tidal volume set to 0.4 ml at a rate of 110 strokes/min. The chest is then shaved, cleaned free of hair and sterilized. Under a dissecting microscope, a left thoracotomy is performed in the fourth intercostal space. After displacing the pericardium, an 8-0 monofilament nylon suture on a curved tapered needle is passed under the left anterior descending coronary artery (LAD) 4 mm below the left atrium and permanently tied to eliminate blood flow distal to the suture. Following verification of induced ischemia via epicardial blanching, exosome re-suspended in sterile PBS are injected into the infarct border zone as two separate 10 µl injections (one on either side of the ligation). Pericardium is re-draped over heart, and the chest was then closed following the injection. A 22 gauge syringe was used to re-establish negative pressure within the chest cavity prior to extubation. Animals received post-surgical pain management with buprenorphine and surgical inflammation control with meloxicam. Animals were recovered until freely mobile on a

heating pad at which point they were then placed into a clean cage and housed for the duration of the experiment.

Echocardiography

Transthoracic two-dimensional M-mode echocardiography was performed using the Vevo770 (VisualSonics, Toronto, ON, Canada) equipped with a 30-MHz transducer. Mice were anesthetized for analysis with a mixture of 1.5% isoflurane and oxygen (1 L/min) before AMI (baseline) at 1, 2, 3, 4, 6 and 8 weeks after AMI. M-mode tracings were used to measure left ventricular wall thickness and left ventricular inner diameter in systole and diastole. The mean value of three measurements was determined for each sample. Percentage fractional shortening and percentage ejection fraction were calculated as described previously [7, 8].

Histology and Staining

Immunostaining of CPCs is performed on cells grown on permanox or glass chamber slides[10]. Cells are fixed by 4% paraformaldehyde (PFA), permeabilized in PBS supplemented by 0.2% Triton-X for 10 min, and blocked in PBS supplemented with 10% horse serum for 1 hr. Primary antibodies diluted are applied overnight at 4°C after blocking in PBS with 10% horse serum. The next day, cells are washed with PBS and incubated for 1 h at room temperature with secondary antibodies (Jackson Laboratories, USA) diluted in blocking solution. Sytox Blue or To-Pro (Molecular Probes, USA) is diluted in Vectashield (Vector Labs, CA, USA) mounting media at 1:500 vol/vol and used as nuclear staining.

Paraffin heart sections are deparaffinized in xylene and rehydrated through graded alcohols to distilled water. Antigen retrieval is achieved by boiling the slides in 10 mmol/L citrate pH 6.0 for 12–15 min. Slides are washed several times with distilled water and once with TN buffer (100 mmol/L Tris, 150 mmol/L NaCl). Endogenous tissue peroxidase activity is quenched with TN buffer supplemented with 3% H_2O_2 for 20 min whenever necessary. Slides are then washed in TN buffer and blocked in TNB buffer (TSATM kit from Perkin-Elmer) at room temperature for at least 30 min. Primary antibodies are applied overnight at 4°C in TNB buffer. The next day, samples are washed in TN buffer and incubated with secondary antibodies at room temperature in the dark for 1 hr. When amplification of signal is needed, slides are washed in TN buffer and incubated with streptavidin horseradish peroxidase conjugated diluted 1:100 vol/vol in TNB buffer for 30 min at room temperature, and signals are developed using Tyramide substrate diluted 1:50 vol/vol in Amplification Diluent (Perkin-Elmer) for 10 min. Slides are washed in TN buffer and coverslipped using Vectashield in the presence of DNA staining. List of primary and secondary antibodies is reported in OnlineTable II.

Infarct Size Determination

All fixed hearts were sectioned starting from the height of the ligating suture and then sequentially at 250-μm distances below the suture as far as effective sectioning would permit. Infarct size was evaluated on Masson's trichrome-stained heart sections cut 500 μm below the ligation point with ImageJ (National Institutes of Health), and the transmural, fibrotic infarct perimeter was then assessed as a percentage of the entire LV chamber perimeter.

Capillary Density Analysis

Capillaries were identified by injecting mice with BS-1 lectin (Vector Laboratories, Burlingame, CA) 10 min prior to sacrifice. Subsequent staining of sections included a goat anti-lectin primary antibody (Vector Laboratories) and FITC-conjugated donkey anti-goat IgG secondary antibody. Slides were imaged using fluorescent microscopy (Zeiss), and capillary density was evaluated by counting positively stained tubular structures within the infarct border zone in sections 500 μm below the ligation point in all hearts. Three high-power visual fields (×20) were analyzed from three independent mouse sections.

Oxygen consumption rate (OCR) measurment

A Seahorse Bioscience XF96 Extracellular Flux Analyzer was utilized to measure oxygen consumption rates (OCR) in cardiac progenitor cells (CPCs) using a protocol similar to that previously reported[9]. CPCs were plated at 3,000 cells per well in XF media supplemented with 1 mM pyruvate, 2 mM glutamine, 10 mM glucose. Three independent OCR measurements were acquired for each condition: baseline, following the addition of 3 μM oligomycin, 2 μM FCCP, and 0.5 μM Antimycin A and 0.5 0.5 μM Rotenone. At the conclusion of each experiment, cells were lysed in RIPA buffer and protein concentration was determined for each well using a standard Bradford assay. All calculations for assessment of OCR were evaluated from the third reading in each condition and all values reported as mean +/- SEM, (pmoles O2/min/mg).

Online Figure legends

Online Figure I: Characterization exosomes derived from embryonic stem cells (ESC). A-C) Exosomes secreted by MEF cells with higher magnification of the exosome in adjacent panels compared to mES exosomes (D-F) as confirmed by electron microscopy Panel A, D scale bar=5μm, Panel B, E scale bar=1μm, Panel C, F scale bar=100nm. G) Exosomes from MEF and mES cells express exosome marker Flotillin-1 and are negative for nuclear marker Lamin B as confirmed by immunoblot analysis. Protein from MEF and mES cells was used as controls. H) Increased mRNA levels of pluripotency markers OCT-4, SOX2, Nanog in mES exosomes compared to MEF cells and media alone confirming the ESC origin of exosomes. mES cells were used as positive control and showed significantly high expression of the pluripotency markers.

Online Figure II: Reduced fibrosis in the heart after mES Ex treatment. A-C) Decreased fibrosis in mice receiving mES Ex compared to MEF Ex and saline treated animals at 4 weeks after infarction as evidenced by Masson's trichrome staining with corresponding quantification of infarct size (D) (n=6). Scale bar =20μm. Saline vs. mES Ex $^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$ and MEF Ex vs mES Ex $^{\#}p < 0.05$, $^{\#\#}p < 0.01$, $^{\#\#\#}p < 0.001$.

Online Figure III: Assessment of tumor formation in mice receiving mES Ex. Hematoxylin and Eosin staining of the heart samples showed no tumor in the heart of animals receiving saline A), MEF Ex B) and mES Ex C).

Online Figure IV: mES Ex modulate expression of markers of proliferation. A-C) Increased BrdU+ cardiomyocytes in mES exosomes treated hearts compared to MEF exosomes and saline treated hearts along with corresponding quantification. D) BrdU (green), nuclei (blue) and sarcomeric actin (red). E) Increased mRNA expression of cyclinA2, D1, D2 and E1 after treatment with mES Ex compared to saline and MEF Ex in hearts after myocardial infarction by qRT-PCR (n=5). F) Reduced expression of cell cycle inhibitors p16, p19, p21 and p53 after mES exosome treatment 2 days after infarction (n=4). Saline vs. mES Ex $^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$ and MEF Ex vs mES Ex $^{\#}p < 0.05$, $^{\#\#}p < 0.01$, $^{\#\#\#}p < 0.001$.

Online Figure V: Enhanced CPC proliferation and survival in heart after mES Ex administration. A-C) Increase in number of c-kit+/pH3+ CPCs in the heart 5 days after mES Ex administration compared to MEF Ex and saline treated animals along with corresponding quantification in (D) (n=4)**.** c-kit (red), pH3 (green), sarcomeric actin (blue) and nuclei (white). E-G) Reduction in c-kit+/TUNEL+ CPCs in mES Ex administered hearts after 5 days compared to MEF Ex and saline treated animals. H) Quantification of c-kit+/TUNEL+ cells in all treatment groups (n=4). c-kit (red), TUNEL (green), sarcomeric actin (blue) and nuclei (white). Scale bar=40μm. Saline vs. mES Ex $^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$ and MEF Ex vs mES Ex $^{\#}p < 0.05$, $^{\#\#}p < 0.01$, $^{\#\#\#}p < 0.001$.

Online Figure VI: A) Characterization of cell survival in CPCs treated with exosome free media, MEF exosome free media and mES and MEF growth media shows no significant effect on the number of Annexin-V+ cells as measured by FACS based cell death assay. B) mES Ex pretreated CPCs are observed in the heart 14 days after infarction. C) GFP+ mES Ex-CPCs are observed in close proximity to blood vessels along with formation of a few small myocytes. D) MEF Ex-CPCs form rare GFP+ myocytes in the heart after 14 days. Scale bar=40μm.

Online Figure VII: Reduced apoptosis and increased blood vessel formation by GFP+ mES Ex-CPCs. A-B) Reduced GFP+/TUNEL+ cells in mES Ex-CPC hearts compared to MEF Ex-CPC animals 5 days after infarction along with corresponding quantification (C). GFP

(green), TUNEL (red), sarcomeric actin (blue) and nuclei (white). Scale bar=20μm. D-E) Increased GFP+/SM22+ cells in the heart transplanted with mES Ex-CPC compared to controls at 8 weeks after infarction with quantification (F). GFP (green), SM22 (red), sarcomeric actin (blue) and nuclei (white). Scale bar=20μm. mES Ex-CPC vs. MEF Ex-CPC $*p < 0.05$, $**p < 0.01$, $***p < 0.001$.

Online Figure VIII: A) Differential expression analysis of 338 miRs in mES Ex and MEF Ex shown on the right pie. The left pie indicates total number of miRs with no change in expression (430) and with significant change (338). 59 miRs had significantly high expression in mES exosomes including certain ES-specific miRs (miRs-291, 294, 295). Total identified indicate significant expression of 338 miRs in ES exosomes (left pie). B) Increased expression of miR-291 and miR-294 in hearts transplanted with mES Ex while no expression was detected in saline and MEF Ex transplanted animals (n=6). Saline vs. mES Ex $*p < 0.05$, $**p < 0.01$, $***p < 0.001$. C) Enhanced expression of cell cycle markers (Cyclin E1, A2 and Cdk2) in neonatal rat cardiomyocytes 24 hrs after treatment with mimics for miR-291 (50nM) and miR-294 (50nM) as measured by miRNA qRT-PCR (n=3). Expression of miRs was normalized to endogenous control snoRNA234 while fold expression was calculated against non-treated group that represents cells treated with miR negative control (50nM). D) Increased levels of pluripotent markers (c-myc, Klf4) in CPCs treated with miR-294 (n=3). NT vs. miR-294 $*p < 0.05$, $**p < 0.01$, $***p < 0.001$.

Online Figure IX: mES Ex increase CPC spare-respiratory capacity. CPCs treated with either MEF Ex or mES Ex were examined for oxidative phosphorylation function by measuring oxygen consumption rates (OCR) in a Seahorse XF96 Analyzer. OCR was evaluated at baseline, after the addition of Oligomycin, FCCP, and Rotenone + Antimycin A. **A**) Mean oxygen consumption rates for all experimental conditions for control NT (black line), MEF Ex (grey line), and mES Ex (red line). **B**) Basal respiration rate, calculated as (baseline OCR - Rot/AA OCR), Rot/AA OCR = non-mitochondrial respiration. **C**) ATP production, calculated as (baseline corr. OCR – Oligo corr. OCR). **D**) Maximal respiration, calculated as (FCCP OCR – Rot/AA OCR). **E)** Spare respiratory capacity, calculated as (Max Resp. - Basal Resp.). *(n=28 for all groups, data corrected to protein and shown as mean +/- SEM, *p<0.05, **p<0.01)*

Online Table I: Quantitative Real time PCR

Primer Name	Forward/Reverse	Sequence
OCT4	Forward	TCGGACCAGGCTCAGAGGTA
OCT4	Reverse	ATCCCTCCGCAGAACTCGTA
SOX2	Forward	AAACCAAGACGCTCATGAAGAAG
SOX2	Reverse	CGCTCGCCATGCTGTTC
Nanog	Forward	CAGCATCCATTGCAGCTATCC
Nanog	Reverse	CTGCCCCACATGGAAAGG
Cyclin A2	Forward	GGCTTTTAATGCAGCTGTCTCTTT
Cyclin A2	Reverse	CAAAACTGCCATCCATTGGA
Cyclin D1	Forward	AGCCAGCTGCAGTGCTGTAG
Cyclin D1	Reverse	TGGTGGTGCCCGTTTTG
Cyclin D2	Forward	GCTCTGTGCGCTACCGACTT
Cyclin D2	Reverse	CACGCTTCCAGTTGCAATCA
Cyclin E1	Forward	CACGGGTGAGGTGCTGATG
Cyclin E1	Reverse	AGGACGCACAGGTCTAGAAGCT
p16	Forward	GAACTCTTTCGGTCGTACCC
p16	Reverse	CGAATCTGCACCGTAGTTGA
p19	Forward	AGAGGATCTTGAGAAGAGGGCC
p19	Reverse	GCAGTTCGAATCTGCACCG
p21	Forward	CCTCATATTTCTGGAGTCAGTGTCA
p21	Reverse	GGAAAACCCAAGACTACTGTCACA
p53	Forward	GCTGCCCACCTGCACAA
p53	Reverse	GGTGAAATACTCTCCATCAAGTGGTT
cTnT	Forward	AGATGCTGAAGAAGGTCCAGTAGAG
cTnT	Reverse	CACCAAGTTGGGCATGAAGA
MEF2c	Forward	TTCCACTCCCCCATTGGA
MEF2c	Reverse	TGCGCTTGACTGAAGGACTTT
GATA-6	Forward	CGGTTATCCCAGAACCCATTC
GATA-6	Reverse	TCCTCTCCACGAACGCTTGT
SMA	Forward	CTGCCTCTAGCACACAACTGTGA
SMA	Reverse	ACCACGAGTAACAAATCAAAGCTTT
Ang1	Forward	GGGACAGCAGGCAAACAGA
Ang1	Reverse	TGTCGTTATCAGCATCCTTCGT
CD31	Forward	TCCCCGAAGCAGCACTCTT
CD31	Reverse	ACCGCAATGAGCCCTTTCT
VE-cadherin	Forward	GTGGATGAGCCCCCTGTCT
VE-cadherin	Reverse	CAGCGGTTTCTTCTGGTTTTCT
c-myc	Forward	AGGCCCCCAAGGTAGTGATC
c-myc	Reverse	GTGCTCGTCTGCTTGAATGG
Klf4	Forward	TGCCAGGAGAGAGAGTTCAGTATTT
Klf4	Reverse	CTGGGAACTTGACCATGATTGTAG
Cdk2	Forward	CGGACGGAGCTTGTTATCTCA
Cdk2	Reverse	AGGGCTGCTTTGGCTGAAAT

Online Table II

Antibodies

Application	Antibody	Dilution	Amplify	Cat No.	Company
Western Blot	Flotillin 1	1:500	No	ab41927	Abcam
Western Blot	Lamin B	1:500	No	SC-6216	Santa Cruz
Western Blot	Phospho AKT S473	1:1000	No	4060S	Cell Signaling
Western Blot	AKT	1:3000	No	9272	Cell Signaling
Western Blot	Nucleostemin	1:500	No		R&D Systems
Western Blot	LIN28	1:500	No	8641	Cell signaling
Western Blot	βactin	1:2000	No	3700	Cell Signaling
ICC	Cleaved Caspase 3	1:1000	No	9664S	Cell Signaling
ICC	BrdU	1:100	No	Ab6326	Abcam
ICC	Sarcomeric Actin	1:100	No	A2172	Sigma Aldrich
ICC	c-kit	1:50	Yes	AF1356	R&D Systems
ICC	Phospho Histone 3	1:100	Yes	441190G	Life technologies
ICC	GFP	1:100	Yes	A10262	Life technologies

Supplemental references

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