SUPPLEMENTARY METHODS

Proteomics analysis

A large-scale proteomic approach was performed using a liquid chromatography-quadrupole time-of-flight mass spectrometry (LC-QTOF MS) approach as described previously (61) to analyze LNCaP-Mock and LNCaP-HSP27 proteins. Accumulated data was analyzed using Protein Lynx Global Server software (PLGS 2.3) with peptide and fragment mass accuracies of 15 ppm. This search engine was applied to the full Uniprot 15.0 database, human species. A search of SwissProt Homo Sapiens was also carried out using Mascot software search using PKL peak list files generated in PLGS. Ion score cutoff of 32 is specified as identical or highly homologous according to Mascot outputs. Peptide score ≥ 40 was considered a positive hits. Only proteins with positive peptide hits ≥ 2 were considered statistically significant.

Bioinformatics analysis of proteomics data

To evaluate HSP27 client protein’s involvement in reported biological processes, pathways enrichment analysis on the identified proteins using KEGG Pathway database was performed. We used the R Bioconductor package "Pathview" to retrieve, integrate and visualize pathway enrichments.

Antibodies used for Western blot analysis and Immunoprecipitation (IP)

For western blot, we used 1:5000 rabbit anti-HSP27 polyclonal antibody (Enzo Life Science, Villeurbanne, France), 1:500 mouse anti-Menin monoclonal antibody (SC-390345, Santa Cruz Biotechnology, CA, USA), 1:2000 rabbit anti-Menin polyclonal antibody (Bethyl, Laboratories Inc, TX, USA) and 1:500 mouse anti-ubiquitin monoclonal antibody (Santa Cruz Biotechnology, Heidelberg, Germany). Loading levels were normalized using 1:2000 mouse anti-vinculin monoclonal antibodies (Sigma Chemical, St Louis, MO) or 1:2500 rabbit anti-
GAPH polyclonal antibody (Abcam, Paris, France). For IP, the supernatant was incubated with 8 µg/ml mouse anti-Menin monoclonal antibody (Santa Cruz Biotechnology, CA, USA) or mouse anti-IgG (as an internal control) O/N at 4°C. We used rabbit or mouse True Blot anti-rabbit IgG secondary antibody (Rockland, inc, Limerick, PA, USA) to reveal western blot.

Tissue microarray experiments

Menin protein expression was assessed in Vancouver Prostate Center using two different Tissue MicroArrays (TMA) built from 291 patients (2 cores from each patient) with a total of 582 valid cores. The first "Gleason" TMA includes 198 patients from untreated radical prostatectomies specimen arrayed according to their International Society of Urological Pathology (ISUP2014) Gleason groups including benign prostate hyperplasia (BPH; n=83), G1 (n=64), G2 (n=78), G3 (n=53), G4 (n=69), and G5 (n=49). The second "NHT" TMA includes 94 patients exposed to neoadjuvant hormone therapy (NHT) before surgery includes Naïve (n=76), NHT treatment (n=42) and transurethral resection (TURP) of CRPC (n=70). Immunohistochemistry (IHC) staining was conducted by Ventana Autostainer Model Discover XT (Ventana Medical System, Tuscan, Arizona). Enzyme-labeled biotin-streptavidin system and solvent-resistant 3,3′-Diaminobenzidine (DAB) Map kit was used with 1/50 mouse anti-Menin monoclonal antibody (Santa Cruz Biotechnology, CA, USA). Digital images were obtained using the Leica SCN400 scanning system (Concord Leica Microsystems, Ontario, Canada), and scoring was performed visually by an anatomopathologist (Ladan Fazli). Nuclear immunoreactivity of Menin was evaluated using a 3-point scale scoring system by assigning zero for negative, one for weak to moderate, and two for strong staining intensity in the majority of tumor cells.

Menin mRNA expression correlates with prostate cancer progression
We performed expression analysis of MEN1 mRNA expression in a pooled series of 2,081 clinical tissue samples publicly available, including 272 “normal” samples (normal tissue and benign prostate hyperplasia: BPH), 1,643 PC primary tumor samples, and 90 PC metastatic samples

RNA isolation, Reverse Transcription and Quantitative Real-Time Polymerase Chain Reaction (RT-PCR)

Total RNAs were extracted from PC-3 treated with OGX-427 or control-OGX-427 using RNeasy® Mini quit (Qiagen, Courtaboeuf, France). mRNA (1 µg) was reverse transcribed to cDNA using SuperScript II Reverse Transcriptase and Random Primers (Invitrogen). To amplify MEN1 sequence, specific primers were designed as forward: 5’-CCCTCTACCACAAGGGCATT-3’, reverse: 5’-TCCCCGGCACGTAGTTGTAGTC-3’ and probe sequence Tamra 5’-FAM-ACAGTGGCCGTGTCCGCCCA-3’. Quantitative reverse transcription PCR analysis was done on the CFX96™ Real-Time PCR Detection System. Expression levels of the GAPDH (TaqMan® GAPDH Control Reagents, catalog number 402869; Applied Biosystems) gene were used as an internal control.

Sequencing and data processing

ChIP-seq libraries were generated using the MicroPlex Library Preparation Kit (Diagenode, Seraing-Belgium) following the manufacturer's instructions. Quantification and qualification of the library were performed using a Qubit® 2.0 Fluorometer (Life Technologies, Carlsbad, CA, USA) and a TapeStation System (Agilent Technologies, Les Ulis, France). ChIP-sequencing was performed with a NextSeq500 sequencer (Illumina, San Diego, CA, USA) using a 75-nt single-end protocol. Computational analyses are developed in the supplemental experimental procedure. Profiles of Menin ChIP-seq were obtained by processing normalized bigwig files through the deepTools suite (v2.2.4 computeMatrix, plotProfile). All sequencing data were visualized using Integrative Genomics Viewer (IGV v2.3.92). Statistical software R
(version 3.5.1, https://www.r-project.org/) was used for integration between ChIPseq (Differentially Bound Genes, DBGs) and RNA-seq (Differential Expression Genes, DEGs) comparative analyses. The biological significance of DBGs was explored by Gene Ontology (GO) term enrichment analysis including biological process (BP) and molecular function (MF) based on Bioconductor packages enrichR (https://cran.r-project.org/package=enrichR). KEGG pathway enrichment analysis of DBGs or Differentially Expressed and Bound Genes (DEBGs) was performed with enrichR. A value of p<0.05 was considered statistically significant.

**Computational analyses**

Reads with a Phred quality score less than 30 were filtered out. For Menin ChIP-seq, reads were mapped to Homo sapiens genome assembly (GRCh37/hg19) using default parameters of Bowtie2 (v2.3.4.1) (1). Duplicate tags were removed and mapped tags were processed for further analysis. High confidence binding sites were determined using MACS2 peak caller (2): broad mode (broad-cutoff=0.01 -p 0.01). Inputs were used as controls. To get rid of noisy signal issues, peaks located less than 2500 bp from each other were stitched together and peaks smaller than 500 bp wide were removed. To link ChIP-seq data with gene expression, only peaks located at genes TSS (TSS defined from -2kb to +5kb) were filtered in using BEDTools intersect (v2.17.0) with a minimum overlap of 1-base (3). For ChIP-seq quantitative analysis, mapped tags were counted within TSS-intersected peak coordinates (defined previously by MACS2) using featureCounts (4) and then normalized as "reads per million mapped reads" using their respective library size.

For IGV visualization, mapped tags were converted into BigWig using the deepTools suite (v2.2.4) (bamCoverage) (5) and normalized by their library size using (scaleFactor argument). For RNA-seq, PC3 and LNCaP expression tables were obtained from the publicly available Gene Expression Omnibus (GEO) repository (accession number: GSE59009) and merged. The
supervised analysis was carried out using Welch’s t-test and FDR adjustment using `p.adjust` function.

For ChIP-seq and RNA-seq crossing, only Menin target genes (i.e. genes bound by Menin on their TSS) were analyzed. The biological significance of Differentially Expressed and Menin-Bound Genes (DEBGs) was explored by GO term enrichment analysis including biological process (BP) and molecular function (MF), based on Bioconductor packages clusterProfiler (6) (https://bioconductor.org/packages/release/bioc/html/clusterProfiler.html). KEGG pathway enrichment analysis of DEBGs was performed with clusterProfiler as well. A value of $P<0.05$ was considered statistically significant.

**Antibodies used for western blot to validate the pathways activated by Menin in CR models.**

Mouse monoclonal Anti-Akt3 (Clinisciences, Clone EE-M14, 1/500), mouse monoclonal Anti-Ets-1 (Clinisciences, Clone C-4, 1/500), Anti-PKC mouse monoclonal IgG2a Antibody (Clinisciences, clone MC5, 1/500) and mouse monoclonal Anti-MET (Clinisciences, Clone D-4, 1/500).

**Mass spectrometry analysis for Menin interactome**

The immunoprecipitated samples were stacked on NuPAGE™ 4–12% Bis–tris acrylamide gels according to the manufacturer’s instructions (Invitrogen, Life Technologies). Protein containing bands were stained with Thermo Scientific Imperial Blue, cut from the gel, and following reduction and iodoacetamide alkylation, digested with high sequencing grade trypsin (Promega, Madison, WI, USA). Peptides extracts were concentrated and analyzed by Liquid Chromatography coupled to tandem Mass Spectrometry (LC-MS/MS) using an LTQ-Velos-Orbitrap (Thermo Electron, Bremen, Germany) connected to a nanoLC Ultimate 3000 Rapid Separation Liquid chromatography system (Dionex, Sunnyvale, CA). 10% of whole sample (5
µl) were injected in triplicate on the system. After pre-concentration and washing of the sample on a Dionex Acclaim PepMap 100 column (C18, 2 cm × 100 µm i.d. 100 A pore size, 5 µm particle size), peptides were separated on a Dionex Acclaim PepMap RSLC column (C18, 15 cm × 75 µm i.d., 100 A, 2 µm particle size) at 300 nL/min flow rate and eluted by a two steps linear gradient (4-20% acetonitrile/H20; 0.1 % formic acid for 90 min and 20-45% acetonitrile/H20; 0.1 % formic acid for 30 min. The separation of the peptides was monitored by a UV detector (absorption at 214 nm). For peptides ionization in the nanospray source, spray voltage was set at 1.4 kV at 275°C. All samples were measured in a data-dependent acquisition mode. Each run was preceded by a blank MS run to monitor system background. The peptide masses were measured in a survey full scan (scan range 300-1700 m/z at 30 K FWHM resolution at m/z=400, target AGC value of 1.00×10^6 and maximum injection time of 200 ms).

In parallel to the high-resolution full scan in the Orbitrap, the data-dependent CID scans of the 10 most intense precursor ions were fragmented and measured in the linear ion trap (normalized collision energy of 35 %, activation time of 10 ms, target AGC value of 3.00×10^4, maximum injection time 100 ms, isolation window 2 Da). Parent masses obtained in Orbitrap analyzer were automatically calibrated on the 445.120025 ion used as lock mass. The fragment ion masses were measured in the linear ion trap to have maximum sensitivity and the maximum amount of MS/MS data. Dynamic exclusion was enabled with a repeat count of 1 and an exclusion duration of 30 s. Raw files generated from mass spectrometry analysis were processed with Proteome Discoverer 1.4.1.14 (Thermo Fisher Scientific). This software was used to search data via in-house Mascot server (version 2.4.1; Matrix Science Inc., London, UK) against the Human subset (20.202 sequences) of the SwissProt database (2019_05). Database search was done using the following settings: a maximum of two trypsin miscleavage allowed, methionine oxidation, N-terminal protein acetylation as variable modifications, cysteine carbamido-methylation as a fixed modification. A peptide mass tolerance of 6 ppm and a
fragment mass tolerance of 0.8 Da were used for search analysis. Only peptides with high stringency Mascot score threshold (identity, FDR<0.1%) were selected and used for protein identification. The “precursor ions area detector” option of Proteome Discoverer was activated to obtain MS area information of the 3 more intense peptides of the identified protein.

SUPPLEMENTARY FIGURES LEGENDS

Figure S1: (A) Quantification of Menin expression levels in LNCaP-HSP27 compared to LNCaP-Mock. Bands were quantified using ImageJ software normalized to vinculin protein levels. (B) Quantification of Menin expression levels in PC models LNCaP, DU-145 and PC-3. Bands were quantified using ImageJ software normalized to GAPDH protein levels (C) Quantification of Menin and HSP27 interaction in LNCaP-HSP27 compared to LNCaP-Mock. Bands were quantified using ImageJ software normalized to HSP27 protein levels. (D) Quantification of Menin expression levels after OGX-427 treatment. Bands were quantified using ImageJ software normalized to vinculin protein levels. Data were normalized to Control-OGX (100%) and are shown as mean ±SEM, n=3. Two-tailed, unpaired Student's t-test, *P≤0.05, **P≤0.01, ***P≤0.001. (E) Estimation of Menin half-life by a cycloheximide treatment in PC-3 cells. Cells were treated with cycloheximide (CHX) (10 μg/ml) or CHX followed by MG-132 (10 μmol/l) for indicated periods. Cells were harvested for protein extraction and evaluated by western blot analysis using anti-Menin and anti-vinculin antibodies.

Figure S2: (A) Inhibition of Menin enhances cell apoptosis specifically in AIPC (PC-3) model. PNT1A and PC-3 cells were transfected with 100 nMol Menin- or control-ASO for 2 days after the second transfection. Data were normalized to untreated cells, and are shown as mean ±SEM, n=3. Two-tailed, unpaired Student's t-test *P≤0.05, **P≤0.01, ***P≤0.001. (B) Quantification of Menin protein levels after ASO-Menin treatment in vivo. Bands were quantified using ImageJ software normalized to vinculin protein levels. Data were normalized
to Control-OGX (100%) and are shown as mean ±SEM, n=3. Two-tailed, unpaired Student's t-test, \( *P \leq 0.05, **P \leq 0.01, ***P \leq 0.001 \).

**Figure S3:** (A) Average profile of ChIP peaks is a graph showing the read count frequency in the range from \(-1000\)bp to \(+5000\)bp. (B) Volcano plot representing differential genes expression RNA-Seq in LNCaP compared to PC-3 (GSE59009). The y-axis corresponds to the mean expression value of negative log10 (adjusted p-value), and the x-axis displays the log2 fold change value. The yellow dots represent the up-regulated expressed genes (p-adj <0.01, log2 fold change >2). (C) The median effective concentration (EC50) of cisplatin is at 27.9 μM. PC-3 cells were transfected with 100 nM Menin- or Control-ASO for 2 days and treated at 0, 1, 2.5, 5, 10, 25, 30 or 40 μM of cisplatin for 48 hours after the second transfection. Cell viability was determined using MTT test.
SUPPLEMENTARY TABLES

Table S1. Protein list detected in LNCaP-HSP27 and LNCaP-Mock cells (MS Excel file).

Table S2. List of KEGG pathways where HSP27 is implicated.

| KEGG Identifier | Name                                |
|-----------------|-------------------------------------|
| hsa04210        | Apoptosis                           |
| hsa05215        | Prostate cancer                     |
| hsa04110        | Cell cycle                          |
| hsa00620        | Pyruvate metabolism                 |
| hsa04010        | MAPK                                |
| hsa04151        | PI3K/AKT                            |
| hsa04014        | Ras                                 |
| hsa05202        | Transcriptional misregulation in cancer |
| hsa04370        | VEGF                                |
| Hsa04115        | P53 signaling pathway               |

Table S3. List of Menin isoforms detected in LNCaP-HSP27 compare to LNCaP-Mock.

| Menin isoforms | Number of peptides detected | LNCaP-HSP27 | LNCaP-Mock |
|----------------|----------------------------|-------------|------------|
| O00255         | 6                          | 0           | 0          |
| O00255-2       | 5                          | 0           | 0          |
| O00255-3       | 5                          | 0           | 0          |
| E7EN32         | 5                          | 0           | 0          |
| E7EPR4         | 4                          | 0           | 0          |
| E7ET29         | 4                          | 0           | 0          |
| E7ENS2         | 3                          | 0           | 0          |
| Q9GZQ5         | 2                          | 0           | 0          |
| References | Source of data | Technological platform | Nº of probe sets/genes | Nº of samples | Normal prostate samples (N) | Primary PC samples (N) | Metastatic PC samples (N) |
|------------|----------------|-----------------------|-----------------------|---------------|---------------------------|---------------------|--------------------------|
| (7) | GEO database, GSE55945 | Affymetrix, array U133 Plus 2.0 | 54K | 21 | 0 | 13 | 0 |
| (8) | GEO database, GSE35988 | Agilent, array 4x44K G4112F(014850) | 44K | 122 | 0 | 59 | 35 |
| (9) | Array-Express database, E-MTAB-6128 | Affymetrix, Human Gene 2.0 ST Array | 54K | 141 | 40 | 101 | 0 |
| (10) | Array-Express database, E-TABM-26 | Affymetrix, array U133 A+B | 2x22K | 57 | 13 | 44 | 0 |
| (11) | GEO database, GSE10645 | Illumina, DASL Human Cancer + Custom Prostate Panel | 500+500 (1K) | 596 | 0 | 596 | 0 |
| (12) | BROAD Institute, Cancer Program portal [https://portals.broadinstitute.org/cgi-bin/cancer/datasets.cgi](https://portals.broadinstitute.org/cgi-bin/cancer/datasets.cgi) | Affymetrix custom array, Hu6800 + Hu35KsubA | 7K+9K | 23 | 9 | 10 | 4 |
| (13) | GEO database, GSE6811 | Non-commercial spotted DNA/cDNA, YN Human 36K array | 37K | 35 | 0 | 24 | 11 |
| (14) | GEO database, GSE21034 | Affymetrix, Human Exon 1.0 ST Array | 43K | 185 | 29 | 131 | 19 |
| TCGA, PRAD, Cell 2015 | TCGA portal, [https://tcga-data.nci.nih.gov](https://tcga-data.nci.nih.gov) | Illumina, RNA sequencing V2 | 25K | 551 | 52 | 498 | 1 |
| (15) | GEO database, GSE6099 | Non-commercial spotted DNA/cDNA, Chinnaiyan | 20K | 104 | 18 | 32 | 20 |
Table S5. Correlations of MEN1 mRNA expression with clinicopathological variables.

| Characteristics                              | N     | "MEN1-low" class | "MEN1-high" class | p-value |
|----------------------------------------------|-------|------------------|-------------------|---------|
| Patients' age, median (range)                | 1373  | 62.67 (37.3-83)  | 62.87 (45.98-78) | 0.654   |
| ISUP2014 grade                              |       |                  |                   |         |
| 1                                            | 294   | 233 (29%)        | 61 (22%)          | 1.03E-02|
| 2                                            | 149   | 119 (15%)        | 30 (11%)          |         |
| 3                                            | 133   | 91 (12%)         | 42 (15%)          |         |
| 4                                            | 175   | 127 (16%)        | 48 (17%)          |         |
| 5                                            | 320   | 221 (28%)        | 99 (35%)          |         |
| Serum PSA level (ng/ml), median (range)      | 544   | 37.59 (0-3247)   | 16.02 (0-623)    | 0.105   |
| AJCC pathological tumor size (pT)            |       |                  |                   | 1.60E-04|
| pT2                                          | 686   | 551 (51%)        | 135 (39%)         |         |
| pT3                                          | 712   | 512 (48%)        | 200 (58%)         |         |
| pT4                                          | 21    | 12 (1%)          | 9 (3%)            |         |
| AJCC pathological lymph node                 |       |                  |                   | 0.0625  |
| pN0                                          | 926   | 704 (86%)        | 222 (82%)         |         |
| pN1                                          | 160   | 110 (14%)        | 50 (18%)          |         |
| Surgical resection margins                   |       |                  |                   | 1       |
| R0                                           | 490   | 366 (67%)        | 124 (67%)         |         |
| R1-2                                         | 239   | 178 (33%)        | 61 (33%)          |         |
| Follow-up, months (range)                    | 1298  | 34 (1-219)       | 27 (1-194)        | 3.28E-02|
| BRFS event                                   | 1298  | 383 (39%)        | 140 (44%)         | 0.148   |
| 10-year BRFS                                 | 1298  | 42% [38-46]      | 34% [27-42]       | 9.68E-03|
| OS event                                     | 1222  | 135 (15%)        | 59 (20%)          | 3.53E-02|
| 10-year OS                                   | 1222  | 83% [80-87]      | 74% [67-81]       | 9.75E-04|
Table S6. Univariate and multivariate prognostic analyses for Biochemical Recurrence-Free Survival (BRFS) and Overall survival (OS) (MS Excel file)
Table S7. List of selected ASOs covering Menin mRNA.

| ASO Number | Position | Antisense 5′-3′                  | % GC |
|------------|----------|---------------------------------|------|
| 6          | 211-230  | CACCAAGGAAAGGAGGCACCA          | 55   |
| 7          | 231-250  | AAATCGTTCACGAAGCCCAAG          | 55   |
| 8          | 251-270  | TGACCGGTTTACAGCCACAGCA         | 60   |
| 9          | 271-290  | CTCGGGAACGTTGAGGAGGCA          | 60   |
| 12         | 331-350  | CACGGGAAAGTACAGGCTAGGC         | 60   |
| 13         | 351-370  | GGATAGGAGGCACGTTGACCA          | 60   |
| 16         | 411-430  | GGATAGAGGAGGCACGTTGACCA        | 60   |
| 17         | 431-450  | TGGGAGACACCCCTTCTCAGA          | 60   |
| 18         | 451-470  | CTTCTCACCAGGCTCAGGC           | 60   |
| 19         | 471-490  | TCCCATATGACATCGGAGAC          | 45   |
| 21         | 511-530  | GATGTTGGGCCGCGACCTTGGA        | 60   |
| 22         | 531-550  | ATGAAACTGGAAGGGAGCTGCA        | 60   |
| 23         | 551-570  | TGTCAAATCTGCTGCTTGGA          | 50   |
| 27         | 631-650  | ATCCCTACAGGAGGCGGGGTA         | 60   |
| 28         | 651-670  | CCAAACACTACCCAGGCATG          | 55   |
| 34         | 771-790  | TATGACTTCTTTTACGATAGC         | 40   |
| 35         | 791-810  | TCTTGCGGTACAGGCGCATG          | 60   |
| 36         | 811-830  | CACCATGTTCGCCACCTCAG          | 50   |
| 37         | 831-850  | TGATGGCAACAGTGGAGGCTG         | 50   |
| 39         | 871-890  | CTGCAGCTCAGAGACCTGCA          | 60   |
| 40         | 891-910  | AGCAGACAGACAGCTCTCTG          | 60   |
| 41         | 911-930  | CCAGATGTCGCCAGGTCTAGA         | 55   |
| 43         | 951-970  | TCTAGATCTGCCAGGTCCCTC       | 55   |
| 46         | 1011-1030| GCAATGCCCCTTGTGAGTAGAG        | 55   |
| 48         | 1051-1070| GGGGTAGATGTGTTGTCATCC         | 55   |
| 50         | 1071-1090| CATTGGCTGTCGACGAGTTG          | 60   |
| 63         | 1351-1370| GATGCCGCCAGATGAATCGCA         | 55   |
| 64         | 1371-1390| CTCGCCCTCTCCTCCATTTGCC        | 60   |
| 66         | 1411-1430| AAGAAGATGGCCGCCAGGCA          | 60   |
| 67         | 1431-1450| AACCGGCTAAGGGACTGCA          | 60   |
| 69         | 1471-1490| TCGGGCTCAGATAGCGCAGCT         | 60   |
| 86         | 1811-1830| CGCTTGAGTTGATCTTGTGTG         | 55   |
| 87         | 1831-1850| GTGAGTTGACAGCTTGTG           | 55   |
| 88         | 1851-1870| ATCTGCACCTTGCAGCTGTG         | 55   |

Table S8. List of targets Menin detected using ChIP-seq (MS Excel file)
Table S9. List of microRNAs regulated by Menin in normal prostate model.

| MicroRNAs   | Targets                                                                 | Roles                              | References |
|-------------|-------------------------------------------------------------------------|------------------------------------|------------|
| MIR-1-2     | FN1 (fibronectin), LASP1 (LIM and SH3 protein 1) and PTMA (prothymosin alpha) | Inhibit cell adhesion and migration | (18)       |
| MIR-133a/b  | EGFR (epidermal growth factor receptor)                                  | Inhibit proliferation, migration and invasion | (19)       |
| MIR-155     | CTHRC1 (collagen triple helix repeat containing 1)                       | Inhibit cell proliferation, cell cycle and promoted cell apoptosis | (20)       |
| MIR-15a, MIR16-1 | CCND1 (cyclin D1)                                           | Inhibit cell cycle and proliferation | (21)       |
| MIR-29-a    | KDM5B (Lysine Demethylase 5B)                                            | Inhibit cell cycle and promoted apoptosis | (22)       |
| MIR-29-b    | CDH2 (cadherin 2, type 1, N-cadherin (neuronal))                        | Inhibit cell adhesion              | (23)       |
| MIR-30-a    | SIX1 (Sine oculis homeobox homolog 1)                                    | Inhibit cell proliferation, and invasion | (24)       |
| MIR-30-d    | Src (tyrosine kinase) NT5E (Ecto-5′-nucleotidase)                        | Inhibit cycle migration and invasion | (25), (26) |
| Let-7-a-1   | E2F2 (E2F transcription factor 2) CCND2 (Cyclin D2)                     | Inhibit cell cycle progression     | (27)       |
| Let-7-d     | AEG1 (Astrocyte elevated gene-1)                                         | Inhibit proliferation and invasion and promotes apoptosis | (28)       |
| Let-7-f     | AKT2 (protein kinase B)                                                  | Inhibit proliferation and promotes apoptosis | (29)       |
**Table S10.** List of the Differentially Expressed and Menin-Bound Genes (DEBGs) in PC-3 cell line. The genes in red were selected for biological validation.

| ECM-receptor interaction | Focal adhesion | ErbB signaling pathway | Ras signaling pathway | EGFR tyrosine kinase inhibitor resistance | PI3K/AKT | Pathways in cancer | Transcriptional misregulation in cancer | Proteoglycans in cancer | Axon guidance |
|--------------------------|----------------|-----------------------|-----------------------|------------------------------------------|----------|------------------|----------------------------------------|-------------------------|--------------|
| COL4A1                   | COL4A1         | AREG                  | FGF5                  | NRG2                                     | COL4A1   | COL4A1           | RUNX1                                  | ITGB1                   | ITGB1        |
| COL4A2                   | COL4A2         | NRG2                  | VEGF5                 | AXL                                      | COL4A2   | COL4A2           | JMD1C                                  | FGFR1                   | EPHA2        |
| LAMA5                    | LAMA5          | PIK3CA                | EPHA2                 | KDR                                      | LAMA5    | LAMA5            | HMG2                                   | KDR                     | EPHA5        |
| ITGA1                    | ITGA1          | SOS1                  | FGFR1                 | MET                                      | ITGA1    | ITGA1            | METJ                                   | MET                     | MET          |
| ITGA3                    | ITGA3          | AKT3                  | MET                   | PIK3CA                                   | ITGA3    | ITGA3            | PIK3CA                                  | CD44                    | CAMK2D       |
| ITGB1                    | ITGB1          | PRKCA                 | MET                   | AKT3                                     | ITGB1    | ITGB1            | AKT3                                   | PRKCA                   | PRKCA        |
| ITGB4                    | ITGB4          | CAMK2D                | PRKCA                 | RALBP1                                   | ITGB4    | ITGB4            | METJ                                   | RALBP1                  | RALBP1       |
| CD44                     | MYL12A         | VCL                   | ETS1                  | ETS2                                     | ETS1     | ETS1             | ETS1                                   | ETS1                    | ETS1         |
|                          | ACTN1          | VEGF5                 | EPHA2                 | PIK3CA                                   | PIK3CA   | PIK3CA           | BIO5                                   | PIK3CA                  | PIK3CA       |
|                          | KDR            | KDR                   | KDR                   | KDR                                      | KDR      | KDR              | KDR                                    | KDR                     | KDR          |
|                          | MET            | MET                   | MET                   | MET                                      | MET      | MET              | MET                                    | MET                     | MET          |
|                          | SOS1           | SOS1                  | SOS1                  | SOS1                                     | SOS1     | SOS1             | SOS1                                   | SOS1                    | SOS1         |
|                          | AKT3           | AKT3                  | AKT3                  | AKT3                                     | AKT3     | AKT3             | AKT3                                   | AKT3                    | AKT3         |
|                          | PRKCA          | PRKCA                 | PRKCA                 | PRKCA                                    | PRKCA    | PRKCA            | PRKCA                                  | PRKCA                   | PRKCA        |
|                          | CAV1           | CAV1                  | CAV1                  | CAV1                                     | CAV1     | CAV1             | CAV1                                   | CAV1                    | CAV1         |

**Table S11.** List of Menin interactors detected using IP-MS (MS Excel file).

**Table S12.** List of transcription factors interacting with Menin.
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Supplementary Figures

A.

Expression antibody compared to vinculin

- LNCaP-Mock
- LNCaP-HSP27

B.

Menin expression compared to GAPDH

- LNCaP
- DU-145
- PC-3

Figure S1
C. Menin expression compared to HSP27

D. Expression antibody compared to vinculin

E. Western blot analysis with anti-Menin IB (67 KDa) and anti-vinculin IB (130 KDa)

Figure S1
Figure S2

A.

Apoptotic cells (% of control)

Control-ASO  |  Menin-ASO
---|---
PNTA1  |  PC3

B.

Expression anti-Menin compared to vinculin

Control-ASO  |  Menin-ASO
---|---

*  |  **
Figure S3

A.

LNCaP (AS)
PC3 (AI)
PNT1A (N)

B.

PC3 vs LNCaP RNAseq GSE59009

- Log(pval) vs Log2(fold change)

FC > 2 & pval < 0.05
FC > 2 & adjpval < 0.05
FC > 2 & adjpval < 0.01

Menin1
C.

Figure S3

Concentration of cisplatin (µM)

Cell viability %