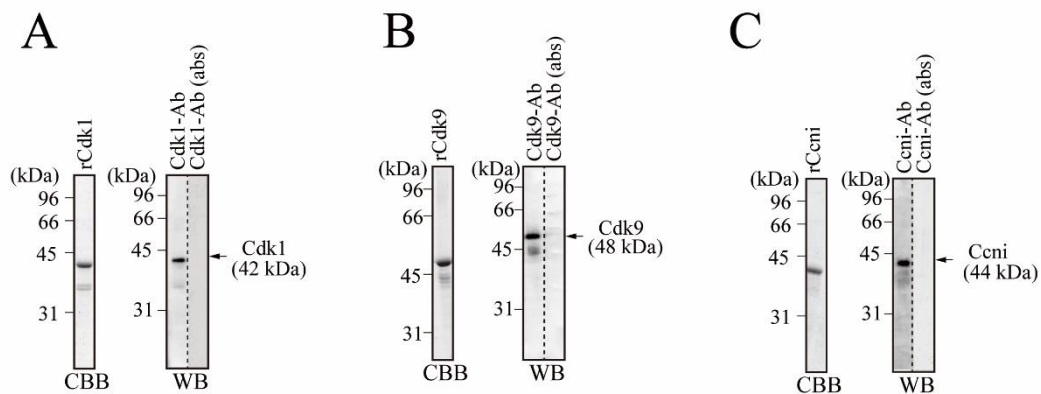


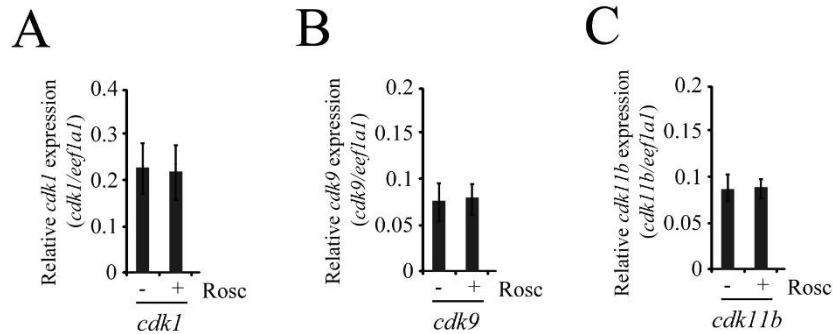
**Figure S1.** Expression of *cdk1*, *cdk9*, and *cdk11b* mRNA in medaka preovulatory follicles during 24-h spawning cycle.

Total RNAs were prepared from the follicles at various time points in the 24-h spawning cycle and were used for real-time RT-PCR of *cdk1*, *cdk9*, and *cdk11b* mRNA. Relative follicular expression is shown (N=5-7).



**Figure S2.** Characterization of antibodies prepared in this study.

Antibodies raised in this study were characterized. Recombinant Cdk1 (A), Cdk9 (B), and cyclin I (C), which were separately used as antigens for immunizing mice, were analyzed for purity by SDS-PAGE/CBB staining (CBB). SDS-PAGE/Western blot analysis was conducted for Cdk antibodies using extracts of ovaries isolated from spawning female fish at 5 h before ovulation (left lane in each panel of WB). As negative control, the antibodies were preincubated with the respective antigens, and the absorbed antibodies (abs) were used (right lane in each panel of WB). Positions of various molecular masses are indicated.



**Figure S3.** Expression of *cdk1*, *cdk9*, and *cdk11b* mRNA in preovulatory follicles cultured with or without roscovitine.

The -14 h-follicles were incubated with or without Rosc (50  $\mu$ M) for 18 h, and the expression levels of *cdk1*, *cdk9*, and *cdk11b* were determined by real time RT-PCR (N=5).

**Supplemental Table S1.** Primers used in this study.

| Primer name          | gene          | Sequence                     | Accession No. |
|----------------------|---------------|------------------------------|---------------|
| <b>Real-time PCR</b> |               |                              |               |
| mmp2 ss              | <i>mmp2</i>   | 5'- GCGACGACGGCTTTTTGTGG-3'  | AB033754      |
| mmp2 as              | <i>mmp2</i>   | 5'- CATCTCCATTCCTCCAG-3'     |               |
| mmp14 ss             | <i>mmp14</i>  | 5'-CGGAGGGTTTCCACGGCGAC-3'   | AB185847      |
| mmp14 as             | <i>mmp14</i>  | 5'-CCAATCGTCCACGGCTCAGC-3'   |               |
| mmp15 ss             | <i>mmp15</i>  | 5'-ACGGCTCCCTCCAGTTTTA-3'    | AB072928      |
| mmp15 as             | <i>mmp15</i>  | 5'-GGTGTTCCTGCGTCTTC-3'      |               |
| timp2b ss            | <i>timp2b</i> | 5'-TGGAGACTGATGGGACGATG-3'   | AB193468      |
| timp2b as            | <i>timp2b</i> | 5'-GCACGCAGGAATGGAAGTG-3'    |               |
| cdk1 ss              | <i>cdk1</i>   | 5'-ATGGAGGACTACGTGAAAATAG-3' | AB040436      |
| cdk1 as              | <i>cdk1</i>   | 5'-GGAAGTCCCTCCTCCTCACTCT-3' |               |
| cdk2 ss              | <i>cdk2</i>   | 5'-ATGGATACGTTTCAGAAAGTGG-3' | XM_020703479  |
| cdk2 as              | <i>cdk2</i>   | 5'-CGAATCGCTGTGCTTGGTACGC-3' |               |
| cdk4 ss              | <i>cdk4</i>   | 5'-ATGGCGCAGTGCACCGGTCTGC-3' | XM_011475449  |
| cdk4 as              | <i>cdk4</i>   | 5'-CGCGCACGTTCTTCAGGGCGAC-3' |               |
| cdk5 ss              | <i>cdk5</i>   | 5'-ATGCAGAAATATGAAAAGCTTG-3' | XM_004079396  |
| cdk5 as              | <i>cdk5</i>   | 5'-TCGTCATCGTCTAGTCTGACTC-3' |               |

|              |               |                               |              |
|--------------|---------------|-------------------------------|--------------|
| cdk7 ss      | <i>cdk7</i>   | 5'-ATGTCTGTGGATGTGAAAACAA-3'  | XM_004072423 |
| cdk7 as      | <i>cdk7</i>   | 5'-AATGGCAACTATTGTGTCAGTC-3'  |              |
| cdk8 ss      | <i>cdk8</i>   | 5'-ATGGACTATGACTTTTAAACTGA-3' | XM_004081058 |
| cdk8 as      | <i>cdk8</i>   | 5'-GTACACATGACCGTAGGTGCCT-3'  |              |
| cdk9 ss      | <i>cdk9</i>   | 5'-ATGCAACGAGACAAAACAAGCA-3'  | XM_004086584 |
| cdk9 as      | <i>cdk9</i>   | 5'-TCGTACTTGGAGAACTCGTCGC-3'  |              |
| cdk10 ss     | <i>cdk10</i>  | 5'-ATGGACGCCACTGGAGAGGAAG-3'  | XM_011476444 |
| cdk10 as     | <i>cdk10</i>  | 5'-ACCTATCCGATTGAGTTTCTCA-3'  |              |
| cdk11b ss    | <i>cdk11b</i> | 5'-ATGGAAATAACAATTCGTAATT-3'  | XM_023956292 |
| cdk11b as    | <i>cdk11b</i> | 5'-GAGACTTTTCCTTTCTTGCCAC-3'  |              |
| cyclin I ss  | <i>ccni</i>   | 5'-ATGAAGAGCCCAGGAGCCGCAG-3'  | LC435346     |
| cyclin I as  | <i>ccni</i>   | 5'-GATGTTGGGAAAAGGAGATGTC-3'  |              |
| cyclin G2 ss | <i>ccng2</i>  | 5'-ATGGACGCCGTCAGCTGATGA-3'   | XM_004075148 |
| cyclin G2 as | <i>ccng2</i>  | 5'-CACTTGGCGGAGATCCTGCTGT-3'  |              |
| cyclin E2 ss | <i>ccne2</i>  | 5'-ATGTCAAGACGCAGTGGTCGCA-3'  | XM_023964550 |
| cyclin E2 as | <i>ccne2</i>  | 5'-CTTGGAGGCAGGCTGGAGCTTC-3'  |              |
| ef1a ss      | <i>ee1a1</i>  | 5'-CACCGGTCACCTGATCTACA-3'    | AB013606     |
| ef1a as      | <i>ee1a1</i>  | 5'-GCTCAGCCTTGAGTTTGTCC-3'    |              |

#### ChIP

|                            |              |                               |  |
|----------------------------|--------------|-------------------------------|--|
| Primer pair-1 SS for mmp15 | <i>mmp15</i> | 5'-GAAAGTCATGACGTCCTGG-3'     |  |
| Primer pair-1 AS for mmp15 | <i>mmp15</i> | 5'-GGCTCCGCTCCTCCAGAGCC-3'    |  |
| Primer pair-2 SS for mmp15 | <i>mmp15</i> | 5'-GTTAGGTTAGGCACACTTAA-3'    |  |
| Primer pair-2 AS for mmp15 | <i>mmp15</i> | 5'-TGGAATGTAAACCATTACAA-3'    |  |
| Primer pair-3 SS for mmp15 | <i>mmp15</i> | 5'-CATGTTATAACACATGGATG-3'    |  |
| Primer pair-3 AS for mmp15 | <i>mmp15</i> | 5'-TTAAGTGTGCCTAACCTAAC-3'    |  |
| Primer pair-4 SS for mmp15 | <i>mmp15</i> | 5'-CCAATGCTAATGGATACGCT-3'    |  |
| Primer pair-4 AS for mmp15 | <i>mmp15</i> | 5'-ATGTGAACCACTTTGATGAT-3'    |  |
| Primer pair-5 SS for mmp15 | <i>mmp15</i> | 5'-CTAAAGTTTCCCATTAACT-3'     |  |
| Primer pair-5 AS for mmp15 | <i>mmp15</i> | 5'-GTCATATTTCACTCCCACAA-3'    |  |
| Primer pair-6 AS for mmp15 | <i>mmp15</i> | 5'-CTTTTGTACCGTCTCCAT-3'      |  |
| Primer pair-6 AS for mmp15 | <i>mmp15</i> | 5'-GACAAGATGGCCACCAGCTT-3'    |  |
| Primer pair-7 AS for mmp15 | <i>mmp15</i> | 5'-TTTCGTGCGGATGATATATC-3'    |  |
| Primer pair-7 AS for mmp15 | <i>mmp15</i> | 5'-TTGCAGCACATTAAGATTGG-3'    |  |
| Primer pair-8 SS for mmp15 | <i>mmp15</i> | 5'- AGATATTGCAGTTGCTATACGG-3' |  |
| Primer pair-8 AS for mmp15 | <i>mmp15</i> | 5'- TCATCTGTGTTTCCTCTTTCAC-3' |  |

| <b><u>Recombinant proteins</u></b> |             |                              |                    |
|------------------------------------|-------------|------------------------------|--------------------|
| Cdk1 pET SS                        | <i>cdk1</i> | 5'-ATGGAGGACTACGTGAAAATAG-3' | AB040436           |
| Cdk1 pET AS                        | <i>cdk1</i> | 5'-TCAGACGCCGTTGATGCAGGCG-3' |                    |
| Cdk9 pET SS                        | <i>cdk9</i> | 5'-ATGCAACGAGACAAAACAAGCA-3' | XM_004086584       |
| Cdk9 pET AS                        | <i>cdk9</i> | 5'-CTAAAAGACTCGGTGGAAGTCC-3' |                    |
| Cyclin I pET SS                    | <i>ccni</i> | 5'-ATGAAGAGCCCAGGAGCCGCAG-3' | LC435346           |
| Cyclin I pET AS                    | <i>ccni</i> | 5'-CTAGTTGACAGCAGGGTGCAAT-3' |                    |
| Rpl7 pET SS                        | <i>rpl7</i> | 5'-ATGGCGGACGCAGAAAAAAG-3'   | DQ118296           |
| Rpl7 pET AS                        | <i>rpl7</i> | 5'-TTAGTTCATCCTTCGGATCATT-3' |                    |
| <b><u>Cloning</u></b>              |             |                              |                    |
| cyclin I 5'-SS                     | <i>ccni</i> | 5'-ATCATCTCAATGCTCCGTCCAT-3' | ENSORLT00000001568 |
| cyclin I 3'-AS                     | <i>ccni</i> | 5'-TCATTTGACCCTGGAGAGCTTG-3' |                    |
| <b><u>Crispr/Cas9</u></b>          |             |                              |                    |
| cdk9-SS for Crispr                 | <i>cdk9</i> | 5'-TAGGCTCTGAAGAAAGTGCTGA-3' | XM_004086584       |
| cdk9-AS for Crispr                 | <i>cdk9</i> | 5'-AAACTCAGCACTTTCTTCAGAG-3' |                    |
| Hyg SS                             |             | 5'-GCGCAGCACCATGGCCTGAA-3'   |                    |
| Hyg AS                             |             | 5'-ACACAAAAAACCAACACACA-3'   |                    |