GAGGCTCACAAGACCTATGTCTACCAGTATGAGACGACGCTCATGGGCGGAATGCAGGAGGAGAAATTGGCAAAATCCGGACTCAACTTC 180 AGTTCTAAAGTCCTAATTAGTGCTGAGTCTGGCAACATATACCTGCTTAAGCTAGAGGAACCTGAGATCTTCGAGTTAAATGGGGTCTTG 270 TCAAAAGATCCATTGGTCCCAGCCTCCAGACTGACCGCAGCCCTGAAACATCAGTTAACGGATCCCATCAAGTTTGAGTACACCAACGGC 360 GTTGTTGGGAAGATGATGGCTCCCGCTAGTGTGTCCACACTTGTGTTGAACATTTACAGGGGTATCCTGAATATCCTTCAACTCAACATC 450 AAGAAGACGCACAACGTCTATGAGTTGCAGGAGGCTGGAGCTCAGGGAGTGTGCAAGACCACATATGCCATCTTTGAGGAGGAGAAGGCT 540 GAACAAATTCATGTGACCAAGACCAGGGATTTGAACCAGTGTCAGGAGGGATTCATCAAGGATATGGGGTTGGCATACACTGAGAGATGT 630 GTCAAGTGCCAGCAGGACACGAAGAGCCTGAGAGGATCAACAGCATACGATTACATCTTTAAGTCACACGCTGAAAGTGTCCAGATCCTG 720 GTTGAGATCCAGAAGACTCCAATCGTTGTCGGCAGCTCTCCCTACGTCGATAGTGGGTCTCTTAGGTACGAGTTTCCCAGGGAGCTTTTC 900 CAGACACCTCTGCAGCTTGTCAAGATCGTCGAATTACAGAAGCAGATTGGGGAACTTCTTGATCACCTAATCGCCAACAATGTGGAGAGA 990 AGCAGCAAGAACACCTACAGATGGTGGGTTTGGGATTCCGTAGCAGCTACAGGAACTCCTGCTGCTGCTGAGATTCATTAAAGACAAGTTC 1170 CTGAAAAAGGAAGTGTCAGTCGCTGAAATGGCCCAAGCTATGGTGACATCTGTTCACATGGTGACAGCAACCACTGAGACCATTGAGATC 1260 TTTAAGGACCTGGCAAAACGCAAGGAAATAGTGGACAGCCCATTTCTGCGTGAAATAGTTCTGCTCGGCTATGGGACCATGATTGGAAAA 1350 GAAATCATCCTGCACCTGAAGGTTCTGGGAAACGCTGGCCTTCCTGACAGCATCAAGCCGATCACAAGATCCTGCCCATAGCCCACTCG 1530 CCCACGGATACATCTCTGCCCATTACAGTCTATGCTGAAGCCATCATGGCCCTGAGGAACATTGCAAAGAAGGAGCCCAAACGGATCCAG 1620 GAACTGGCTCTTCAGCTGTACATGGACAAGAACCTTGACCCCGAGCTTCGTATGCTTTCCTGCATCGTCCTGTTTGAAACCAAACCTTCT 1710 CTTGGTCTGGTGACAGCACTTGCCAACATTGTGAGGACAGAGGAGAGGAGGAGGAGTCTGCAGTTAGTGAGCTTTACCTACTCTCACATGAAGTCCCTG 1800 AGCAGGAGCACCTCCATCGTCCACTCATCAGTTGCTGCCAGCTTGCAACATTGCTCTCAGAATGTTGAGCCCCAAAGTTGAACAGACTGAGC 1890 CTCCGTTTCAGCAGAGCCATCCATCTGGATGTCTATAACAGTCATTACATGCTCGGCGCCGCTGCGGCCAATGCTTTCTACATCAACAATGCT 1980 GCCAGCATTTTGCCCAGAAATTTCATGGCGAAGGCCAGTGCATATGTTTCTGGAGCTGCTGTTGATGTTCTGGAGGTTGGAATGAGAACT 2070 GAAGGACTGCAGGAGTTACTTCTGAAAAAACCCCCACAATTCTCAACAACGAGGACAGGATCACCAAGATGAGGAAAGCCATCCAAGCTCTT 2160 CAAGCACTTCAGCTTCCTAGTGTTGAATCTATGAGGACCATTGGTAATGACATCATCAGAGAAATGGCATCTGGCATCTCGGTAAACATC 2340 GTCAAGCCTCTGCTGGTCAGTGAGGTGAGGCGTATCATGCCCACTGCTGCTGGTTTTCCATTGGAGTTCAGTCTGTTAACAGCTGCCGTG 2430 ACAGCTACAGGTGTCAGAGTCAAGGCCACCATGACACCAGCCCTGCCGGAAAACTTCAAACTTCATTGAACTCCTGCAGACCGACATGCAA 2520 ATCGAGAGTGAGATCAAACCAAGCATTGCTGTGAACACCCTTGCAACCATGGGAATAAACACTGCTATGTTCCAGTCGGGAATTGTCTCC 2610 AGAGCTAAAACTCACCACCACCACCACCACCAACATTGTTGCAAGGCTAAACCTCCACAAGGGCTACTTTAAGATTGAAGCTCTGCCCGTT 2700 TCCTTGCCTGAGAACATTGCAGCCATGCACATTGAAACCCTAGCTGTGACCAGAAACATCGAGGACTTAAGTGCTGCAAGACTAACCCCG 2790 ATCATCCCCGGAACAAGTCCAGAATCTTGCTTCAATGGAGGATCTTACATCCCCGGAATGAAAAAAGCCAGTCACATTCGTCAGAGGTTTAT 2880 TACCAAGACCATCCACGGCCATTCACAAAGTCTAAAGTTGCCCGGGTTAAGAATTACAGCGCTGAAATCCTGGGACTGAAGGGCAGTGTG 2970 ACTATGGTCACTCACAATGCTGCTTTCATCATGGACGTTCCCCCTGTACAGACTAGCTGGAACACACTATGCTGCTGTTTCTATCACACCA 3060 AAAAGCAAGGGCAGCAGCAGGGGCAGTGACTCCAGATCTGTTATAAACTTATCCAGTGGCAGCACCTCTGCATCGAACACCACGTCACGT 3330 TCATCACGACAGAGGAAACATGTCACGAAGCGAATTAAAAACCCATAAGTTCAACAAGAACCACAGGAAGCAGTATAAAAGTCAAATGGCA 3420 GATTCCTCTGGCAGTGCTGCAAGCTTTGAGGCAGTCTACGAGCAGAACAAATTCCTTGGCAGCTCTGGAGTTCCTAGCTTTGCTGTAGTC 3510 GTGCGTGCTGTCAAAGGTAAGATGGTAGCAGGATACCAGTTGGCAGTCTACATGGACAAACCCACGAATAGAATTCAGATGATTCTGGCT 3600 GATCTGGACTCTGAAAACAACTGGAAACTCTGTGCTGATGGAATTGTGCTTAGCGAGCACAGAGTCGTAGCTAGAATTGGCTACGGAGCT 3690 GCATGCAACAAGTACAGGGCCACCATCACTGCAGAGACTGGTCTCGTTGGTCCCGAGCCCTGCAGGTCGCCTCAGAGTGGACTGGAACGAA 3780 ATACCCGATGCCCTTAAGCGCACGTTGGAAAAGGCAATGAAGAGCATTCCTTTATCCATCGAGTGTACTTTTGACAAGCAAAGGAGCAAA 3870 AACCTGGATGTCCTCTTCCGATTACCCTTCCAATCAATGAAATACATGGCCTCGCCCCCTTTGGTGACATCGTTGATGAACTCCATGTT 4050 **TfVWDF** primer GTGGCAGCCAAGGCCATAGCAGCTCAGTGTAACTACGCCCAGAACTTGCTGACCACCTTCAACGACATCAAATACGAGCCCCAGATGCCG 4140 T T F N D I K Y E P Q M P CCGTCTTGCTACCAAATTCTGGTTCAGGACTGCACACCCGAGCTGAAATTCATTGTTATGCTGAAGAATGATAACTTTGAACAGAAACAC 4230

P S C Y Q I L V Q D C T P E L K F I V M L K N D N F E Q K H

ATAAATATAAAGATCGCTGACATCGACATCGACCTCTTCCCCCAAGAGTGGCAACATAGGTGTGAAGGTCAATGGTGTGGAAATACCAATG 4320 Κ D I D L F P K S G Ν Ι G V Κ G Е Т Ν Ι Ι А D Ι V Ν V T Р M GAAAACCTGCCATACCATCATCCCACAGTTAAAATCCAGATCAGGCAAAAGGGTGAAGGCATCTCTGTTGTTGCTCCCAGCCTCGGGCTT 4410 Е Ν L Р Ү Н Н Κ RQKGE GΙ S V V А S L G L Р ΤV Ι Q Ι Р AGTGAAGTCTATATGGACAGTAAATCATGGAAGGTTGATGTTGTGGACTGGATGAAGGGACAGACTTGTGGACTCTGTGGGAAGGCTGAT 4500 V D V М Κ S Е Y М D S Κ S Κ V D W G Q Т С G L G Κ D V W С А GGGGAGATCAAACAGGAGTTCCGTATGCCCAACGGGCACCTGACCAAGAATGCGGTCACTTACGCTCATTCCTGGATCCTGGCGGCCGAG 4590 E Τ Κ Q Е F R M Р Ν G AGCTGCAGAGACAACAGCGAGTGCCGAATCAAGCTCGACTCTGTAGAGCTGGAAAAGCAGGCGGTCATCTATGGTCAGGAATCTAGATGT 4680 TfVWDR primer TTCTCCGTTGAGCCGGTGCTCCGCTGCCTGCCCGGCTGCTTCCCTGTGAAGACCACTGCCGTCACCGTTGGCTTCCACTGCGTGGCTGCT 4770 GATTCCAATGTGAACAAATCGGAGGTTCTCAGAGGAATCCAAGGGAAGAGGGTTGACCTGAGGGAAAAAGCTGATGCTCACCTGGCCTGC 4860

Figure S1. The cDNA fragment of the vitellogenin gene. The VWD domain sequence is underlined, the primers are

shown in bold, and the conserved Cys residues are shaded in gray.

AGCTGCACGGCTCAGTGTGTTTGA 4884



**Figure S2.** Construction of the recombinant expression vector. A. Structural map of the pET28a/TfVWD recombinant plasmid vector. B. Electropherogram of the pET28a plasmid following double enzymatic digestion. Lane M: DL2000 Plus DNA Marker; lane 1: Following enzymatic cleavage of the vector. **C** Electropherogram of the PCR product of the TfVWD expression fragment. Lane M: DL2000 Plus DNA Marker; lane 2: PCR product of TfVWD.