

Supporting Information

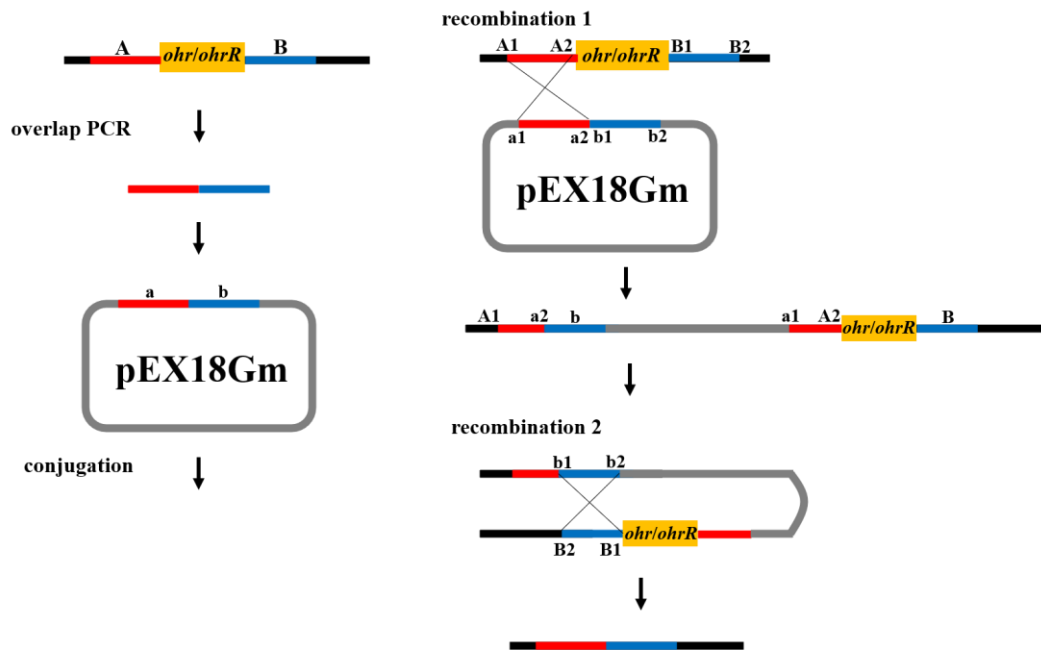


Figure S1. Schematic representation of the strategy used to construct the *ohr* and *ohrR* mutant strains by homologous recombination.

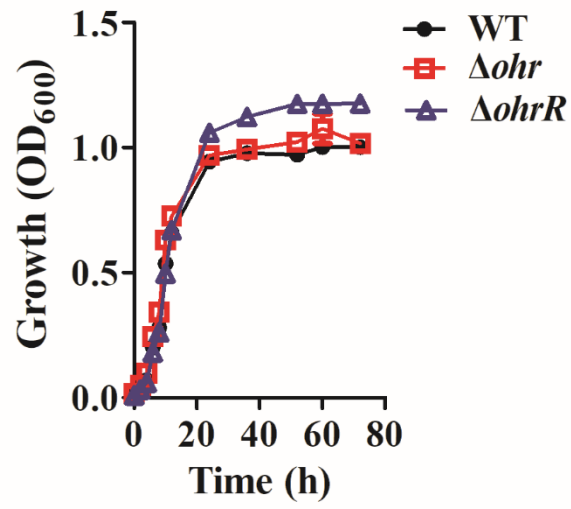


Figure S2. Bacterial growth of *A. caulinodans* WT, Δohr and $\Delta ohrR$ mutant strains.

The cultures were grown in TY medium at 28 °C and OD₆₀₀ was measured at different time points. Experiments were repeated 3 times.

CAAAGGGTTAGTTTTCCGTTGCGGAAGGTTGGGTGATG**TTGACGGTGG**
 TCTGCAAAATAAGTT**TGACACAAT**CAAATTGTGTGCTACTTAAATGGCC
 ATGGAAGCGGCTTGGCCGTTTCTCATCGATTTCTCAAGGCACGAGGATC
 AAGCGATGTCCGTCGATGTGAAGTACACCACCAAG

Figure S3. The promoter sequence of *ohr* gene. The putative -10 and -35 promoter elements are shown in bold, and the inverted repeat motifs are indicated by arrows

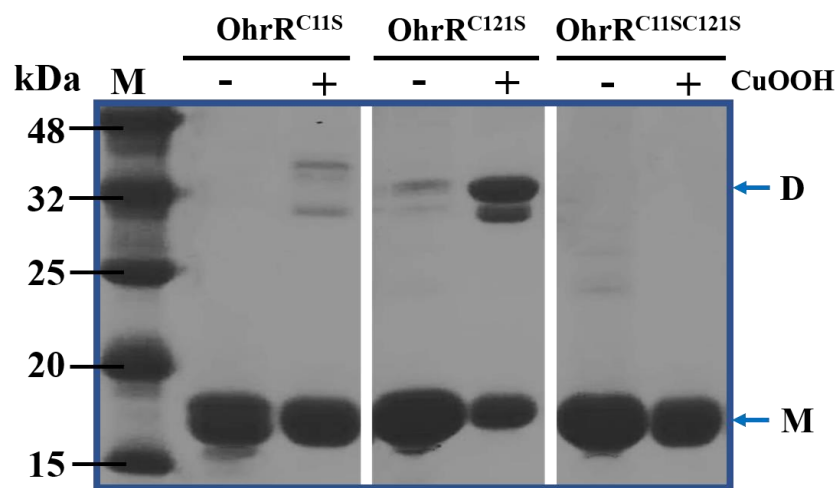


Figure S4. Investigation of intermolecular disulfide bond formation on OhrR variants by nonreducing SDS-PAGE. Samples of reduced proteins were either untreated (-) or treated (+) with CuOOH. The monomeric (M) and dimeric (D) forms of OhrR are indicated by arrows.

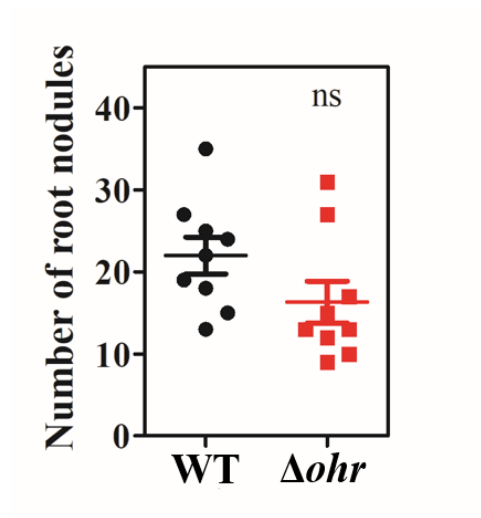


Figure S5. The number of root nodules on *S. rostrata* formed by *A. caulinodans* wild type and Δohr . Horizontal lines are the average number of nodules formed.

Table S1. Strains and plasmids used in this study.

Strains or Plasmids	Relevant characteristics	Source
<i>A. caulinodans</i> strains		
WT	<i>Azorhizobium caulinodans</i> ORS571, AZC0 wild-type, Amp ^R	[1]
Δohr	Derivative of WT carrying an <i>ohr</i> in-frame deletion	This study
$\Delta ohrR$	Derivative of WT carrying an <i>ohrR</i> in-frame deletion	This study
Δohr^c	Mutant Δohr harboring the expression construct pYC12- <i>ohr</i>	This study
$\Delta ohrR^c$	Mutant $\Delta ohrR$ harboring the expression construct pYC12- <i>ohrR</i>	This study
<i>E. coli</i> strains		
DH5 α	Host for cloning	[2]
SM10 λ pir	Host for conjugation	[3]
BL21(DE3)	Host for protein expression	[2]
Plasmids		
pEX18Gm	Suicide cloning vector, Gm ^R	[4]
pYC12	<i>In trans</i> expression vector, Gm ^R	[5]
pRA302	Translational fusion vector, Spe ^R	[6]
pET-28a	Protein expression vector, Kan ^R	[7]

Table S2. PCR primers used in this study.

Primer	Sequence (5'-3') ^a	Restriction sites
For deletion		
AZC2977-1	CGGAATTCTGCCGACTGCCAGCTGACA	<i>EcoRI</i>
AZC2977-2	GCCGTCAGACGACGACGGACATCGCTTGATCCT	
AZC2977-3	AGCGATGTCCGTCGTCGTCTGACGGCGAGGCTG	
AZC2977-4	CCAAGCTTAGCCCGCCAATGTTGAAGAG	<i>HindIII</i>
AZC3555-1	CGGAATTCTCCACGCCCTTCATCTTCTG	<i>EcoRI</i>
AZC3555-2	CCTAGTCGACGGCGAAGGTGGACATGGCAAGGG	
AZC3555-3	CATGTCCACCTTCGCCGTCGACTAGGCGGCAAC	
AZC3555-4	CCAAGCTTTGCGATTGTCCACCTGATGG	<i>HindIII</i>
For <i>in trans</i> expression		
AZC2977-pYC12-F	GGGAATTCATGTCCGTCGATGTGAAGTACAC	<i>EcoRI</i>
AZC2977-pYC12-R	CCAAGCTTGCAACGGCGGACTTACTTC	<i>HindIII</i>
AZC_3555-pYC12-F	GGGAATTCATGTCCACCTTCTGCCTCGACGA	<i>EcoRI</i>
AZC_3555-pYC12-R	CCAAGCTTCTAGTCGACGGCGGCGTTCA	<i>HindIII</i>
For qRT-PCR		
16s rRNA-F	ACGGATTCTTCCAGCAATG	
16s rRNA-R	ACCGGCAGTCCCTTTAGAGT	
AZC2977-F	ATGTGAAGTACACCACCAAGG	
AZC2977-R	CGAACAGTTGCTCGGGATT	
For transcriptional fusion		
<i>ohr</i> -pRA302-F	CGGAATTCCTAAGGGCCGCATGAGAAAGC	<i>EcoRI</i>
<i>ohr</i> -pRA302-R	CCAAGCTTCCCAGTTCCTTCGGCGTGTC	<i>HindIII</i>
For site-directed mutagenesis		
OhrR-11S-1-F	GCGCTTTCGACTTCAGGTTCTGGG	
OhrR-11S-1-R	GGAATAGACGGCGAAGCTCAGCAGG	
OhrR-11S-2-F	GACGACCTGCTGAGCTTCGCCGTCT	
OhrR-11S-2-R	CTAGTCGACGGCGGCGTTTCAGG	
OhrR-121S-1-F	ATGTCCACCTTCTGCCTCG	
OhrR-121S-1-R	GTCCTCGGCGCTCAGGCCGATGGC	
OhrR-121S-2-F	CATCGGCCTGAGCGCCGAGGAC	
OhrR-121S-2-R	CTAGTCGACGGCGGCGTTTC	
For protein expression		
OhrR-pET28a-F	CGGAATTCATGTCCACCTTCTGCCTCG	<i>EcoRI</i>
OhrR-pET28a-R	CCAAGCTTCTAGTCGACGGCGGCGTTTC	<i>HindIII</i>

^aThe underline sequence is the restriction site of indicated enzymes.

References

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