BRUSH LjCNGC.IVA3 LjCNGC.IVA4 LjCNGC.IVA5	MPQFDKDGVPVLLET • HAQQSDEFMDSNCRRLSYRTRSAS • ISIPMVPIEPYEGGTHLVGHTGPLRSVRKPPSGQMSGPLYATTAGA    .AEM.SRF.LE.F.VF.SMSMDTKPNLK.T.FVP.T    .ASSENNEH.TA.EE.SYG.PLHTRFQ.ASN.QCSS.KES.KQRS    .AN.KN.EM.A.KAR.RPG.GPF.KFQ.VT.QT.VV.LSSM.Q.D.SVIRQ.K.NA.LMK.P.T
LjCNGC.IVA3 LjCNGC.IVA4	GNHFQHSIAVPGKKAVEGKTQQLSTFDGTDENLWNNNYDRKNEHLLRSGQLGMCNDPYCTTCPTYIKASQKGNPKVSTVFDSKFHN LLA.E.TA.SN.DDFSHN
LjCNGC.IVA3 LjCNGC.IVA4	SLYGEAKGFGRKLFSFCSSCVPGVMNPHAKVVQQWNKFLAMFCLMAIFVDPLFFFLFYVRKDSNCIVIDLTMTKILLLRSINDVVYL FDL.L
BRUSH LjCNGC.IVA3 LjCNGC.IVA4 LjCNGC.IVA5	LNILLQFRLAYVSPESTVVGAGDLVDNPKKIALNYVKGYFFFDLFVVSPPLPQIMILFVLPTSLGSPGPNSTKNVLHVAVLLQYVPRL    F
BRUSH LjCNGC.IVA3 LjCNGC.IVA4 LjCNGC.IVA5	FRFLPLLIGQSPTGFIFESAWVNFVINLLIFMLSGHVVGSCWYLFGLQRVNQCLQNVCHSSIKHGCTELIDCDSRMGQM
BRUSH LjCNGC.IVA3 LjCNGC.IVA4 LjCNGC.IVA5	SAMWRNNTNATACLNSTS-GSFPYGIYDRAVALTTETKVVKKYVFALFWGFQQISTLAGNQNPSYFEWEVLFTMAIIGLGLLLFALLI .RL.SI.ED.ANAKR.N.YEVEVS. .EQ.ND.IWG.P.TDGN.P.K.DM.TSLT.N.VT .NQ.KD.INPP.D.GEL.P.I.N.N.YSI.V.VV.VS.MI.
BRUSH LjCNGC.IVA3 LjCNGC.IVA4 LjCNGC.IVA5	GNIQNFLQALGRRRLEMQLRGRDVEQWMSHRRLPEGLRRKVRQAERYSWAATRGVNEEMLLENLPEDLQTDIRRHLFKFAKKVRIFAL
BRUSH LjCNGC.IVA3 LjCNGC.IVA4 LjCNGC.IVA5	MDEPILDAIRERLKQKTYIKGSRILSRGSLVEKMVFVVRGKLESIGEDGIGVPLSEGDACGEELLTWYLENSSVSKDGKKVRLPGQRL   C
LjCNGC.IVA3 LjCNGC.IVA4	LSNRTVRCLTNVEAFSLHAADLEEVTFLFTRFLRSPQVQGALRYESPYWRSLAANRIQVAWRYRKKRLGRARANISQSDQTPKS QKGRS.NTSQGL S.IKRR.LH.RTQN.HSSL.G VKS.VNSSVTTKH

**Figure 1-figure supplement 3. Protein sequence comparison of Group IVA CNGCs from** *Lotus japonicus.* Protein sequence alignment of BRUSH (CNGC.IVA1), CNGC.IVA3, CNGC.IVA4, and CNGC.IVA5. Residues identical to BRUSH are shown as dots and gaps are shown as dashes. Shown is the Group IVA CNGC conserved domain (CD, green), *brush* mutation (red asterisk), predicted transmembrane domains (TM, light blue), putative selectivity filter (F, orange) in the pore region, and the cyclic nucleotide-binding domain (CNBD, purple). CNGC.IVA3 (81%), CNGC.IVA4 (65%), and CNGC.IVA5 (72%) show high sequence identity to BRUSH.