

Functional Y₁ and I₁ Receptors Mediate Nicotinic Excitatory Regulation of Nicotinic Acetylcholine Receptor-Mediated Inward Currents

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ac... a... I₈₀c... a... a
... a... c... a... c... a...
H... c, a ac... a... I₈₀c... T.
... d c... a... ATP-d... c... -
... W... a... c... c...
... a... a... ac... I c1 (67).
I c1 b a... c... a... YY1 a... b...
... GLI-K... a... c... I c1 a...
I₈₀c... b... a... a...

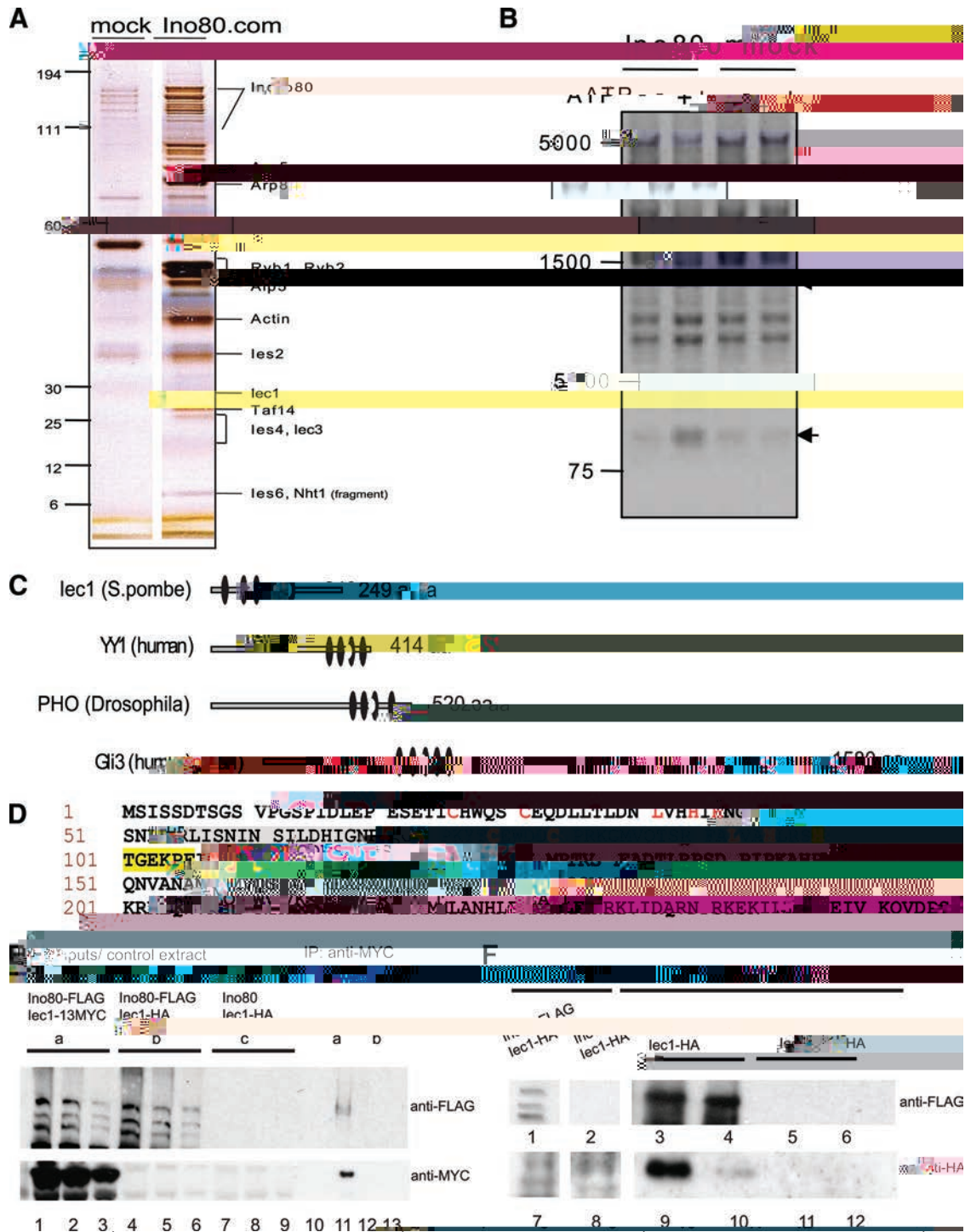


FIG. 1. Ino80 complex and its interaction with Lec1. (A) Fractionation of a pull-down from a mock Ino80.com pull-down. (B) Fractionation of a pull-down from an Ino80 pull-down. (C) Schematic representation of the domain architecture of Lec1, YY1, PHO, and Gli3. (D) Alignment of the HTGEKPF motif. (E) Co-immunoprecipitation assays for Ino80-FLAG and Lec1-HA. (a) FLAG pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (b) FLAG pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (c) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (d) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (e) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (f) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (g) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (h) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (i) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (j) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (k) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (l) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (m) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (n) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (o) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (p) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (q) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (r) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (s) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (t) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (u) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (v) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (w) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (x) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (y) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs. (z) MYC pull-downs from Ino80-FLAG, Iec1-13MYC, and Iec1-HA pull-downs.

c... ad... da (HRP) (S a) a... T... b... d
a... c... c... (ECL) c...
(M...).

Vab → a... C... YES... EMM... a 30-G. T.
c... H₂

TABLE 3. Oligonucleotide PCR primers

Locus	Sequence		
	Forward	Reverse	
1	5'-CGTTCATGCCTCATCTTAATACAGG-3'	5'-ACTACCGCGTAGCGATGAGAA-3'	
	5'-TTGGAACGGCAATGTGCTAGT-3'	5'-GCAGAGTGCGAACATGAGACC-3'	
	5'-TTAAGGTAGCGTTCAATGTTCTTGC-3'	5'-ATTCAGCGAACTGAGCGTTG-3'	
	5'-GACATGGTTCGTAATCCTACCG-3'	5'-CAATAGGAACTGATCCATTAAGCAAT-3'	
	5'-CAACTGGACTCCTGTTATTGAAGC-3'	5'-AGAGCTCATGGTAAACGCTGTA-3'	
	5'-TTACGAGCGTTACCATGAGCTC-3'	5'-GAATCTACAACGCGCTGCTG-3'	
	5'-TCAGAGAATGCTACTGCTGGATC-3'	5'-TTCTCCAAGCTTCGAGAGCA-3'	
	5'-CCTGCTCTCGAAGCTTGGAG-3'	5'-CGTCGTTGGTAAGATTATAGTTGGAA-3'	
	5'-TCCAATAAATCTTACCAACGACGA-3'	5'-CTTACAGAATTCGCTATAGTCTTGAA-3'	
	5'-GAGGTGCCTATGCTAATAGCCTTG-3'	5'-CAACAGGAATAATATTGGCATATG-3'	
	5'-CTTCTACCAAGTCCACTCTCACTC-3'	5'-GCGGACATAGTACTTAGAATCTTCAC-3'	
	5'-GCCATCTTGTTAACGAGGAGGT-3'	5'-AGGCGTACAACCTCACACATACCA-3'	
	5'-ATGGTATGTGTGAGTTGTACGCCTA-3'	5'-AGCAGCCACAGCACTAGCATT-3'	
	5'-AACCTATACGTGCACAACGTTGT-3'	5'-GGTATCGCGTAATTAGTCATCTCTTC-3'	
	2	5'-AGATAGCAGACTAATACTCTTCAATGCC-3'	5'-TTGTACGAGCTGCTACATAATATTGC-3'
5'-TCCAATGTAGTCGTGCCGAA-3'		5'-AAGCGCAAGAAGCAGCAGAT-3'	
5'-AAGTTCGCTGATTGAATAGACTACCG-3'		5'-TGTATGAATAACCAACGACCAG-3'	
5'-CATCCATCGTTCATGCTGGTA-3'		5'-CATGATATACACTAATCGTTCCAAGATG-3'	
5'-AGGACCATCTTGGAACGATTAGTG-3'		5'-GATCCATGACGCTGAAGCAA-3'	
5'-AATGCCGACCAAGTTGTCAG-3'		5'-GAGCAGCAGTATTGATATCGTAGACATC-3'	
5'-GATGTCTACGATATCAATACTGCTGCTC-3'		5'-CGGAAGAAGTATGAAAGTTGGTT-3'	
5'-AGCCTATCGCCAATCGCTTA-3'		5'-TCGCTGTTATCTCTGAGAGCGA-3'	
5'-GTGTGAATAATGCTTCCGATCGT-3'		5'-AAGAGCAGCCTCGACTGGAAT-3'	
5'-CCATTAGAGGTAGAATTAACACGAACC-3'		5'-TTACTGATTGCGGTTATGGTCTC-3'	
5'-TTGTGTCGTTGCTGTGTGTTAA-3'		5'-TCTCAAGATATAACCGCTACATTAACG-3'	
5'-AACCAGCTATCCGGAATTACCA-3'		5'-ACACATGCGGTAATCAGATGAGTT-3'	
3		5'-GAACGGAGAGAAGGAGCGAT-3'	5'-AATCTTACGCTCTACATTACTCGTT-3'
		5'-TAATGATGAACAACGGATATGACTGAA-3'	5'-AACTGTCAGCCACAACCTCCATGT-3'
		5'-TTCGCTGCATAGACGGTAACG-3'	5'-AGGTAATTGATTCTATCGTCGGACA-3'
	5'-GAGGTTGGATAGGAACGGTGA-3'	5'-TTAAGCAATTGCAAGGCGTAA-3'	
	5'-TTGGTCGTCGTAAGCTACAATAAGG-3'	5'-CCACCAGTAGCAAGAATGTCGTC-3'	
	5'-GTTGACGACATCTTGTACTGGT-3'	5'-GGAAGAGATGTCCAACCTAATTCACC-3'	
	5'-CGTAAGCGTCTTATGGCTCCTACT-3'	5'-ATTCACTACCATCAGGTGCTTCATC-3'	
	5'-ATATGACCGCAAGTCTATACACATGC-3'	5'-TCGTACTTCCAAGGAACTCCAGTT-3'	
	5'-CATCCGTGATGAGAAATTAACAATCC-3'	5'-GCTCCACTTGGCAACTGCTC-3'	
	5'-GGGAATCTTGTGTTGAAGATATAATGCC	5'-CCAACAATCACATCAATGTTATTGA-3'	
	4	5'-AGATTCACCAGACATGATTACTGAGTG-3'	5'-TTCATCACGACATGGCCAAT-3'
		5'-GAAGTTGTCAAGAGTGTGCTGCT-3'	5'-ATCGCTCCATCCGTTACACC-3'
		5'-AATGCGAGCATCATGTGCAT-3'	5'-CGCAGCATCACTAGGAAGA-3'
		5'-TGCTGTGCTGTGTTGATTGAGG-3'	5'-TGAACCTCTGTGAATGAGCGAT-3'
		5'-TCTGCGTCACTTGCCCTTCTG-3'	5'-CCTGCAACAATTCCATTCTCAA-3'
5'-GCAGGTGAAGAAGGTGATCCA-3'		5'-TCTGCAACTCGCTTCACTAATCT-3'	
5'-GAGTTAATGAAGCGAGTTGCAGAAG-3'		5'-GGCAATAGAATTGACACAACGAAGT-3'	
5'-GGCGGTTACACCTTGGAGAA-3'		5'-GCCGTTGATAGCAGCATTAGC-3'	
5'-AATGAGCAGGCCATGTTAACATC-3'		5'-CCTGAAGACCTAGCCAGCCAT-3'	
5		5'-TTGTGCTCGCATCTGGAATG-3'	5'-GTAATGTACAATATCGCTGCATCGTT-3'
		5'-CGGTCGTGAACATACCATCG-3'	5'-AGATCCGCTCCATTGCTTG-3'
		5'-GTTCAATCTCATAGAGCAGTTGGTAG-3'	5'-TACGCGTCTCAATTGCAACG-3'
		5'-CCTTCCGTCCTCTATGCCATC-3'	5'-TCAATAATCATCTTACAAGACCGGAA-3'
		5'-CCCCAAATCCAACCGTGAGA-3'	5'-GGCATACAAAGACAAAACAGCTTG-3'
		5'-TGCTCAGTGCCAGAGTGTGATAG-3'	5'-GGTCCGAAGCCGTAGTGTA-3'

U1 RNA, a1, ca1.

7a, ac, HTGEKP(F), a, a, YY1, ac, c, a, GLI-K, a, b, a, SPAC144.02 (F, 1D), (, GLI3, HTGE (KPH).

T, c, I c1, I, 80 c, a, c, b, c, a, -c, ac, MYC, a, HA, a, I c1- a, FLAG, a, I, 80- a, b, W.

b... a... (F... 1E a... F). HA... a... d I c l... a... d...
c... a... c... a... a... c... a... I c l... c...
ac... b... a... c... ca... a... I c l... c...
I... 80... AAA⁺ ATPa... R b2, a... ac... a... a...
a... b... a... I c l-I... 80 c... c... (Tab 4). T... c...
a... a... a... YY1/PHO-I... 80 c...
a... a... a... c... a... c... a... YY1/PHO,
I... 80, R b1/2, ac... (), a... A... 5/A... 8 (Ta-
b 4) (10, 41, 82). W... c... c... d... a... a... I... 80
ac... I c l, a... C₂H₂... c... a...
GLI-K... a... b... c... a... YY1, a... a...
I... 80 c... c... I... 80... b... YY1-
I... 80 c... c...
I c l a d... b... f... e I 80 c... e... a... e... ed
e... ca... a... d... e DNA da... a... e... e... T... d...
b... d... a... I... 80 c... a... DNA... a...
ca... a... d... a... c... (c... c... 4 a... d 77).
W... a... d... ab... a... a... c... a... a...
a... c... I... a... a... a... a... a...
I... 80... a... a... a... a...
T... d... ca... a... I... 80... a... c... ab... (F...
2A). A... ab... a... a... d... d... b... ca...
0... a... d... d... a... a... a... c... d... c...
0^{-/+}... d... c... (da... a...). T... 10,
a... d... b... I... 80 c... d... d...
ab... T... Δ... Δ...
Δ... a... Δ... 10... a... c... DNA... ca...
a... DNA da... a... b... a... a... d...
ac... a... a... c... a... HU... b... c...
a... a... UV... ad... a... a... a... ab...

a... HU... ca... a... a...
ca... a... a... d... d... b... c...
d... ca... a... a... ca... (8, 57). B...
c... ca... a... d... b... a... DNA b... a... (53, 65).
UV... ad... a... a... a... d...
a... DNA da... a... (70). Δ... a... Δ... c... b...
Δ... 10 c... HU... b... c... a... UV... a-
da... c... a... d... c... (F... 2B). A... Δ...
a... c... d... a... a... a... a...
c... d... (YES),... a... a... HU,
b... c... a... UV... ad... (F... 2C). T... a...
I... 80 c... DNA da... a... DNA
ca... W... Δ... I...
a... HU... a... b... c...
I... UV... ad... U... a... c...
I... d... c... a... c... ab... (F... 2D). E...
I c l... Δ... I bac... a... a...
(F... 2D). T... a... a... a... ac... b...

Δ I c ... (F. 3B). W
ac ... a ad ... a ... a ad ...
a ... a ac ... a d, D ... a ... a ...
a ... a ... a ... 5' ... b ... -5-
a ... da ... (AIR), a ... c ... acc ... a d
(F. 3C). U ... c ... Δ I c ...
a ... c ... (YES) acc ... a d ...
(F. 3B). T ... ca ... a ... a ...
AMP, b ... (58). S ... c ...
a ... a ... a ... a ... a ...
b ... a ... a ... b ... a ... a ...
c ... a ... b ... a ... a ... a ...
a ... c ... I ... Δ I c ...
-ad ... c ... (YES), b ... c ...
d ... d ... (F. 3B). T ... c ...
ad ... a ... a ... a ... a ...

Iec1 e ed f e e e f e e ed
ade e a d a e e ab W a d ab
I a a a d c a d a
D I 80 b a ca PHO5, PHO12, a
PHO89 a a ad ab AAH1
a d ADK1 (64, 71, 76). W I cl
I a d ac
a a a T c d
b d a PHO5 a PHO3 (49,
60). P 4 a c ac a a c
ca b a (49, 60). P 1 a d b
a a d a a (49, 60, 61). W a d
I, c a d b a
a 1, a a a a ca a
c a d AMP (F 3C). N b a a
d a RNA a
a c d Δ I c c a a a
a (F 4A a d a a) b
ca d a d a Δ I c c
a c c (F 4A a d B). T d ca a
a a a I cl c a c c
ab W I cl a d a
a a d a a a a c
a a a a a a a
(F 4C).
R b c d d ca (RNR) a
b c d ab a c b c d
b c d (55). T a c a a
RNR cc DNA da a (12, 20). I
a, ac RNR a a a
(Cdc22) a d a (S, c22) b (21). RNA
d c a d I a a EMM a d a
EMM/ P (F 4D). W
c a d HU, a c d a b d a
ad a RNR c a d
b d 0 (52, 84). D I d a c
a a c a d HU (da a
) W a b c a a
d a I a c EMM a EMM/ P
a d a d CTP a d GTP d d c a
a d d TTP c a d a d ATP
I a a c c a d c (F 4E).
T a c a a a b RNR, a
d c a d ATP a d c a d TTP a b
3T6 c a d b c d d ca
b HU (8). T b a a I cl-
I 80 c a d a d c d b
a a c HU ac a
I cl-I 80.
I 80 a e d e f a e e
e e a e a a T d c
I 80 a a a
c d ab d c a
c a b b d a a a
a a 20-b I c a I 80-b

c a a a d c EMM
EMM/ P. I a a a ca d
c I 80 b b a a a a
T a a b a a c a b a
(5 c) a I 80 c
a a a (F 5A) a
I 80 a a a a a
I 80 a a a a ca b
a a d a a a d a
(F 5B). T C IP- c a a
a a acc a I 80 a I, I,
I, I, a a a (F 5C). I
c d ad d a a c d a a d
a a a I a d d d

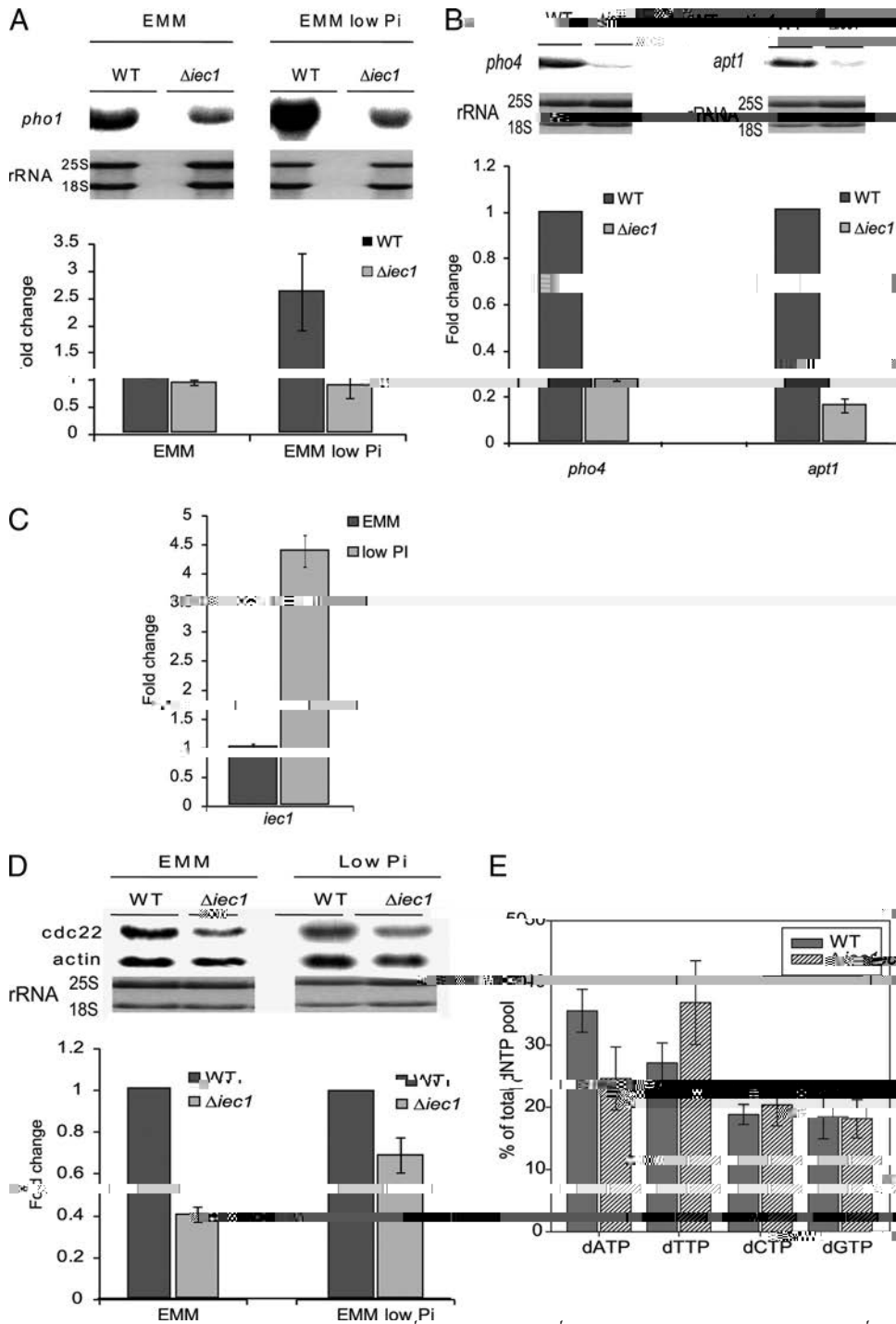


FIG. 4. Iec1 function in yeast. (A) Western blot and bar graph showing *pho1* and rRNA (25S/18S) levels in WT and $\Delta iec1$ strains under EMM and EMM low Pi conditions. (B) Western blot and bar graph showing *pho4* and *apt1* levels. (C) Bar graph showing *iec1* fold change. (D) Western blot and bar graph showing *cdc22* and actin levels. (E) Bar graph showing dNTP pools.

... a a d ad ... ab ... a
... a a ... ad ... a c ... a d ... a l c l ... d ...
... c b d ... l 80 c ... a ... a ...
Iecl ed a e ... ce ... e de e ... e a e e e . W

H. ab. I. 80. d. c. a. O. a. a. I. cl. a. I. 80. c. a. a. b. da. c. c. c. a. W. a. I. 80. a. b. d. (F. 8). c. a. a. I. 80. da. c. ac. a. a. c. a. b. RNA. ca. a. c. a. A. e. f. I. e. c. a. b. e. e. p. a. e. a. d. ce. de. e. ab. W. da. a. Δ. I. a. a. d. a. c. I. 80. c. a. a. Δ. I. a. a. a. ac. a. a. a. a. ad. ab. (F. 3B). T. a. c. d. b. a. a. Δ. I. a. I. 80. c. a. a. d. a. d. (F. 3B). T. a. a. a. d. c. d. (F. 3D). Ta. c. a. b. a. a. ad. ab. c. a. a. T. c. a. c. ca. b. a. d. b. ac. a. c. da. ab. da. b. c. a. a. ca. d. ca. a. c. I. a. a. c. a. a. a. a. a. a. a. c. a. c. a. a. F. a. b. ed. a. a. P. 2. a. c. ac. a. a. a. (14, 58). Ad. 1. c. c. a. ad. a. a. a. PHO5. a. a. ad. a. a. (Ad. 1). PHO. a. ADE. AMP. (27, 33). O. a. a. a. a. I. cl. I. 80. c. a. W. a. I. cl. a. a. c. a. a. ad. ab. a. c. I. 80. c. a. (F. 4C, 5C, a. d. 6C). T. ca. a. I. cl. I. 80. c. a. a. a. I. 80. a. a. b. b. a. ab. a. a. a. c. a. b. ed. a. (52, 56, 74-76). T. b. a. a. d. DNA. da. a. c. a. a. I. 80. c. a. S. c. a. a. d. c. d. a. a. c. a. a. a. b. c. a. a. a. c. c. a. c. a. ca. a. a. W. a. b. a. a. a. c. d. ab. I. 80. c. a. T. a. a. a. d. c. a.

d, b - d ; C, G d, b, c

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