



FIG S1 Presence of *E. typhina* × *festucae* isolate Lp1 *cloA* constructs in genomic DNA preparations of *N. fumigata* transformants as detected by PCR with primer combination 1 (Table 2). Sizes in kb of relevant fragments of *BstEII*-digested bacteriophage lambda DNA are indicated to the left of the photo.

atgatattaccatgggttatcccagcttcaatcggtctcactagggacgattttcctcacg
M I L P W L S Q L Q S V S L G T I F L T
ctattcctcgttatattgactcctttggttttcacaagcgtttaccgctgtgattttcat
L F L V I L T P L V F T S V Y R L Y F H
cctcttcgcaaaattcctggaccagcaaccgggggtttgacaagtttctatgggttctat
P L R K I P G P R T G G L T S F Y G F Y
tggaactggatacagatgaaggatactctaagctcttcaatcccctgcataaacaatat
W N W I R D E G Y S K L F N P L H K Q Y
aattcccatatcatatcgtatcggcccaaacatggtcacatcaaccaaccgcaagctttt
N S H I I R I G P N H V H I N Q P Q A F
gatgagatattcaaaagttggaacaacatggcgcaaaagacagctcattttacaagtatttt
D E I F K V G T T W R K D S S F Y K Y F
aacggcttggacgcatgattgagcgcgcaaatatcgcacctaccgaactcacttggcc
N G L D A M I E P T Q Y R T Y R T H L A
cctttatatacgcacaacgctcattgatggcttaacaccaaagctccatgacgacctcgtg
P L Y A Q R S I D G L T P K L H D D L V
gtaactgcccgaaggatggccaagagcatcgaaaatggtgaacctgtgaacatgggtgaag
V T A E R M A K S I E N G E P V N M V K
atattgcggaattgagtacctcaatgatgctttatactttgtattcgcaggacatcccg
I L R T L S T S M M L Y T L Y S Q D I P
ctctctcaatatgatgggtatcacccgtttctagaagcttttgagctgctcatgacccaa
L S Q Y D G Y H P F L E A F E L L M T Q
agttggctaattgatcaattatcccattgatgggtatgatccttggcctaattcccggcacg
S W L M I N Y P M M G M I L G L I P G T
agctttgcgaaattcaatgcccgttttcggaaccttcttgaagtactgtaaagagtggaaac
S F A K F N A A F G T F L K Y C K E W N
gacgaggatgaacgcattcaaaagcttgaactgctgaatcactgcccggactcccacatg
D E D E R I Q K L E T A E S L R D S H M
aaacgataccttgccattgacccaataacgagatcaaaaagaaggtcgtgccgatccc
K R Y L A I D P N N E I K K K V V P H P
ctggaggatataatttaactttatcgcaggcggtagtgacactacttcatatacagctgca
L E D I F N F I A G G S D T T S Y T A A
tgtgcattcttccatgttctctcgtcgtctgaggtgcactctaagctcgtggcgagctc
C A F F H V L S S S E V H S K L V A E L
gatcaagcttcttcagtgatcagggatacctttgattacaataagattcaaaacttgcca
D Q A S S V I R D T F D Y N K I Q N L P
tatctgaatgccgtgatcaaggagacgcttcgtatctcttgtccgggtaccaggggtgctt
Y L N A V I K E T L R I S C P V P G C L
ccccgagtcgcccctgaggggggaatgaatctgggttcagtaaatcttccagccgggtaca
P R V V P E G G M N L G S V N L P A G T
gtgggtgcaatctcccagctagccatccactttaatgagacgattttctcgtcacctgac
V V S I S Q L A I H F N E T I F S S P D
aagttcatccccgaaagatggcttggggacgatagaaaatcgattgagaagtggaaatc
K F I P E R W L G D D R K S I E K W N I
gcttttagcagaggacctcgacagtgcatgggacaactctcgttatatggaactacgc
A F S R G P R Q C I G T T L A Y M E L R
tgcgtcctcgttattttcttcccgtttgaatttaagtttaacgggtagctgtggagat
C V L A Y F F S R F E F K L T G S C G D
aagttgcgctgggttgatcgatttgtctcagtcacttggacgatgtcagggtcactatc
K L R W V D R F V S V N L D D V E V T I
gtgaaggaccgatgggcgtaa
V K D R W A *

FIG S2 Sequence of cDNA derived from *E. typhina* × *festucae* isolate Lp1 allele of *cloA* expressed in *N. fumigata*. Deduced amino acid sequence is indicated below.

ATGTCGCAACTATGGCTATAACAAGGCTCCAAGTCCCGGGCTTCTGATCACGGCATGTTTCTTCATTGTCTTGCCATGGCTCGTCA
 AAGGCATCTACAATCTGTACTTCCACCCTCTCGAAACATTCCTGGGCCCAAACCTGGCGCCTTGACCAGCTTTTACGCCTTCTA
 CTGGAATTGGATTTCGGGAGGGAGGATACTGTAAGAAATTCGCGCAATATACAAGGACTACAGTACGAATCTCCAGTCAAAGACT
 TTCTTTTTCTCACGGGACCTGCGCTAACCTGCAGACAAGACTCTCCCGTCGTTTCGCATCGGCCCAAACGCTGTTCAATACCAACCA
 GGTGGAACCTACGACGTGTAAGCACGGGGTCCCTTGATCATAATGTGAGAAATGCTTTCTAACTCGAGACGCAAAAAAAAAAAAA
 AAAAAAAAAAAAAAGAATATCAAAGGTGGCTCGGCATGGCTGAAGGACTCGCAATTCTACCAACACTTCAATGGCGTGGATGCCAT
 GATTGGACCGAAGCATTTCCTGACCTATCGCAACCACCTGGCCCCTTTGTATGCGCAGCGGGCGGTGGACGGCCTCACGCCCAAG
 ATTCATAACGACCTCGAACGCTGTGCTGCCAAAATCCACAAGACGGCCGGAACCGGCAACCGGTGAACATGGCAAAGATGTTAC
 GACTGTTGAGTGTGAGTATCCATGCATCGATGCTGCTCCGATACAAGGAGGCTGGGGGCTGAAAGAAAGAGGAGCTTAGAGCTCC
 ATGATTCTCTACAACATTTTTTCGCTCGAAATCTCGCTCTTTGAGCACGACGGGTACCATCCGTTCCCTCGCAGCGTTTGAACAG
 TCATGACACAAAAGCTGGCTGTGTGAGTTCCCTCGGTCCCTGGCCGCTCGCATCCTCGCGGAGGGATTAGAAATAGGGGGAAC
 CTGCTGATCAAGGTTCTCGGTAACCTGGTTGCGCAAAAAAAAAATGCCTAGTTGTGACGTATCCGCTGGTACCCGCGTGGCTGGGC
 CTCATTCCGGGAACGATCTTCTCGCAGTTCAACTCATCTTGGAAACACATTGATGAAGTACTGCACGGCTTGAACGAGGAAGACA
 TGCGCAGGCAGCACGCGAGCGACGAGCAGTCGATTGCGGATTCCCATTCCAAGCGGTACCTTGCCATGAAGAACGAGGGCGACGA
 GGAGAAGAAGAGCATCATCCCCGACCCGATAGATGATGTTTTCAATTTCAATTGCGGGGGTAGCGACACCACGGCCTACACCACT
 TCGGCCGCCCTTCTTCTACATCCCTCTCGTCCGATCCGTTGTACAAAGCTGGTGGAGGAACTCGACGAGAATCGCTCCGTCATCC
 GCGATGCAATGGACTATCACAAGATTACAAGTCTACCTTATCTCGTACGTGCTAGAAACGACCGGCATCCTTCGCGCTCTGTTAT
CCGGAGAAAAAGGAAAAAGAAAAAAAAGGAAAGAAAAAAGGAAAGAAAAAGAAAAATGCCCTGTCACTACTGACTCCTGACTC
CTGCCTCCCCACCTCTAGAACGCCGTGATCAAGGAGACGCTACGAATCTCCGTTCCACTTCTTGCTGCTGCCACGTGCTGCTG
 CCTGAAGGTGGCATACCCGTGGGGTTCGTTCCATCTCCAGCTGGGGTGGAGTTCATGTCGTCGCCCCATGACCCATTTTCGAGAGC
 CTCCTGACCGTGGGGTGACTGATGAACTACAAATCGATCCACACAGACGCTGTGTCTCTTACGCACCAAGTGATTAGCTTCAA
 TGAAGAAATTTTCCCCTCGTCAAAGACGTTTCTTCCGAAAGATGGCTGGGTCCGGAAGCGGTTCGGGCTGATAAGTGGAAATGTG
 GCGTTTAGCCGAGGCCCTCGCCAATGCCTTGGGACAACGTAAGTCGCAAAAAAGAAAAAGAAAAAGAAAAAAGAAAAAAGAAAAA
 AAGAAAAAAAAGGAAAAAAAAGAAAAAAGAAAAAGCAGAGTTTAAATCTACACATTATCTTACGCCGTGAGCCCCCAAGAGCTGC
AGTTAACCCTTTTGACACAGTCTGGCGTACTTGGAGCTTCGCTGCTCCTCGCTTACTTCTTCTCTCGGTTCCAGATGGTCCTGA
 CGGGAAACTCGGGGATCGACTCCGCTGGGTTGACCGATTTGTTGCTGCAACGTTGACGACGTGGAGGTGCGAGTAGTAGCTGA
 TCGATGGACAGGATACGTTCTGA

FIG S3 Sequence of *C. africana cloA* allele to demonstrate issues with intron processing in *N. fumigata*. Initiation and termination codons are highlighted with green and red, respectively. Intronic sequences processed from the mRNA are highlighted in blue. Retained introns are highlighted in gray. Yellow highlight indicates exonic sequences that were erroneously processed from the mRNA. Putative branch site sequences (consensus CTRAC) are underlined.

CLUSTAL O(1.2.4) multiple sequence alignment

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Epichloe_typhina_x_festucaae      MILPWLSQLQSVSLGTIFLTLFLVILTPLVFTSVYRLYFHPLRKPFGPRTGGLTSFYGFY
Claviceps_africana                MSQLWLWLYKAPS--P-GLLITACFFIVLPWLVKGIYNLYFHPLRNIPGPKLGALTSFYAFY
C_africana_synthetic              MSQLWLWLYKAPS--P-GLLITACFFIVLPWLVKGIYNLYFHPLRNIPGPKLGALTSFYAFY
*   ** : *   :::* :.*: * :...:*.*****:****: *.*****:**

Epichloe_typhina_x_festucaae      WNWIRDEGYSKLFNPLHKQYNSHIIRIGPNHVHINQPQAFDEIFKVGTTWRKDS SFYKYF
Claviceps_africana                WNWIREGGYCKKFAQYHKDYNSPVVRI GPNVHTNQVELYDIFKGGSAWLKDSQFYQHF
C_africana_synthetic              WNWIREGGYCKKFAQYHKDYNSPVVRI GPNVHTNQVELYDVIKGGSAWLKDSQFYQHF
*****: **.* *   **:* ** :***** * * * : : * ** *:* * **.***:*

Epichloe_typhina_x_festucaae      NGLDAMIEPTQYRTRYRTHLAPLYAQRSIDGLTPKLHDDLVTVAERMAKSIENGE PVMNVK
Claviceps_africana                NGVDAMIGPKHFRTYRNHLAPLYAQRVDGLTPKIHNDLERCAAKIHKTAGTGKPVNMAK
C_africana_synthetic              NGVDAMIGPKHFRTYRNHLAPLYAQRVDGLTPKIHNDLERCAAKIHKTAGTGKPVNMAK
**:* ** * :.*:***.*****:*****:*** * : : * : .*:*****.*

Epichloe_typhina_x_festucaae      ILRTLSTSMMLYTLYSQDIPLSQYDGYHPFLEAFELLMTQSWLMINYPMMGMILGLIPGT
Claviceps_africana                MLRLLSSSMILYNI FSL EISLFEHDGYHPFLAAFEHVMTQSWLFVTYPLVPAWLG LIPGT
C_africana_synthetic              MLRLLSSSMILYNI FSL EISLFEHDGYHPFLAAFEHVMTQSWLFVTYPLVPAWLG LIPGT
.* **.***.***.*:.* : * * :***** ** * .*****:.***:* *****

Epichloe_typhina_x_festucaae      SFAKFNAAGFTFLKYCKEWNDEDERIQKLETAESLRD SHMKRYLAIDPN-NEIKKKVVPH
Claviceps_africana                IFSQFNSSWNTFMKYCTAWNEEDMRRQHASDEQSIRD SHSKRYLAMKNEGDEEKSIIPD
C_africana_synthetic              IFSQFNSSWNTFMKYCTAWNEEDMRRQHASDEQSIRD SHSKRYLAMKNEGDEEKSIIPD
*:* **:*:.***.***.* **:* * * : . :*:* ** * **.* : : * **.***.*

Epichloe_typhina_x_festucaae      PLEDIFNFIAGGSDTTSYTAACAFFHVLSSSEVH SKLVAELDQASSVIRDTFDYNKIQNL
Claviceps_africana                PIDDFVNF IAGGSDTTAYTTSAAFFYILSSPSVCTKLVEELDENRSVIRDAMDYHKITSL
C_africana_synthetic              PIDDFVNF IAGGSDTTAYTTSAAFFYILSSPSVCTKLVEELDENRSVIRDAMDYHKITSL
*:*:***** **.***.***.***.***.***.***.***.***.***.***.***.***.***.*

Epichloe_typhina_x_festucaae      PYLNAVIKETLRISCPVPGCLPRVVPEGGMNLGSVNL PAVTVVVISQLAIHFNETIFSSP
Claviceps_africana                PYLNAVIKETLRISVPLPGCLPRVVPEGGITVGSFHL PAGTSVSLTHQVISFN EEIFSS
C_africana_synthetic              PYLNAVIKETLRISVPLPGCLPRVVPEGGITVGSFHL PAGTSVSLTHQVISFN EEIFSS
***** **.***.***.***.***.***.***.***.***.***.***.***.***.***.*

Epichloe_typhina_x_festucaae      DKFIPERWLGDDRKSIEKWNIAFSRGPRQCLGTTL AYLELRCVLAFFSRFEFKLTGSCG
Claviceps_africana                KTF LPERWLGPEAVGLDKWNVAFSRGPRQCLGTTL AYLELRCVLAFFSRFQMVLTGNCG
C_africana_synthetic              KTF LPERWLGPEAVGLDKWNVAFSRGPRQCLGTTL AYLELRCVLAFFSRFQMVLTGNCG
..*:* **.***.***.***.***.***.***.***.***.***.***.***.***.***.*

Epichloe_typhina_x_festucaae      DKLRWVDRFVSVNLDDVEVTIVKDRW----
Claviceps_africana                DRLRWVDRFVAANVDDVEVRV VADRWTGDTF
C_africana_synthetic              DRLRWVDRFVAANVDDVEVRV VADRWTGDTF
*:* **.***.***.***.***.***.***.***.***.***.***.***.***.***.*
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FIG S4 Alignment of CloA amino acid sequences deduced from the indicated alleles. The red-highlighted valine residue represents the last amino acid in the version of CloA that would be translated from the misprocessed *C. africana* genomic *cloA* allele expressed in *N. fumigata*. Alignment was produced with CLUSTAL Omega.

C.afr	961	TTCTTCTACATCCTCTCGTCGCCATCCGTGTGTACAAAGCTGGTGGAGGAACTCGACGAG	1020
Synth	961	TTCTTCTACATCCTCTCTAGCCCTTCCGTTTGCACATAAGCTGGTTGAGGAGCTGGACGAG	1020
C.afr	1021	AATCGCTCCGTCATCCGCGATGCAATGGACTATCACAAGATTACAAGTCTACCTTATCTC	1080
Synth	1021	AACCGCTCCGTCATCCGCGACGCTATGGACTACCACAAGATCACTAGTCTGCCTTACCTC	1080
C.afr	1081	AACGCCGTGATCAAGGAGACGCTACGAATCTCCGTTCCACTTCCTGGCTGCCTGCCACGT	1140
Synth	1081	AACGCCGTTATCAAGGAGACTCTGCGCATCTCCGTTCCCTCTGCCTGGTTGCCTGCCTCGT	1140
C.afr	1141	GTCGTGCCTGAAGGTGGCATCACCGTGGGGTTCGTTCCATCTCCCAGCTGGGACGTCTGTG	1200
Synth	1141	GTCGTTCCCTGAGGGTGGTATCACCGTGGCTCTTTCCACCTCCCTGCTGGCACTTCTGTT	1200
C.afr	1201	TCTCTTACGCACCAAGTGATTAGCTTCAATGAAGAAATTTTCCCCTCGTCAAAGACGTTT	1260
Synth	1201	TCTCTGACTCACCAAGTTATTAGCTTCAACGAGGAGATCTTCCCCTCTTCTAAGACTTTC	1260
C.afr	1261	CTTCCCGAAAGATGGCTGGGTCCGGAAGCGGTCCGGCTTGATAAGTGAATGTGGCGTTT	1320
Synth	1261	CTGCCGAGCGCTGGCTGGGTCTGAGGCTGTCCGTTCTCGACAAGTGAACGTTGCCTTC	1320
C.afr	1321	AGCCGAGGCCCTCGCCAATGCCTTGGGACAACCTCTGGCGTACTTGGAGCTTCGCTGCGTC	1380
Synth	1321	AGCCGAGGCCCTCGCCAATGCCTCGGTACTACTCTGGCTTACCTGGAGCTCCGCTGCGTC	1380
C.afr	1381	CTCGCTTACTTCTTCTCTCGGTTCCAGATGGTCCTGACGGGAAACTGCGGGGATCGACTC	1440
Synth	1381	CTGGCTTACTTCTTCTCTCGTTTCCAGATGGTCCTGACTGGCAACTGCGGTGACCGACTG	1440
C.afr	1441	CGCTGGGTTGACCGATTTGTTGCTGCAAACGTTGACGACGTGGAGGTGCGAGTAGTAGCT	1500
Synth	1441	CGCTGGGTTGACCGTTTCGTTGCTGCTAACGTTGACGACGTTGAGGTTGAGGTTGTTGCT	1500
C.afr	1501	GATCGATGGACAGGAGATACGTTCTGA	1527
Synth	1501	GACCGCTGGACTGGTGACACTTTCTGA	1527

FIG S5 Alignment showing changes in coding sequences to optimize codon usage in the synthetic allele derived from *C. africana cloA* (lower sequence) as compared to the coding sequences from the genomic sequence of *C. africana cloA* (upper sequence). Changes in nucleotide sequence did not change amino acid sequence (Fig S3).