UM/UWC Linkage Program

/LQNDJH 5HSRUWV

6XEPLWWHG E\ 'U 0DUVKDOO .H\VWHU

'HSDUWPHQW RI %LRWHFKQRORJ\

8QLYHUVLW\ RI WKH :HVWHUQ &DSH

+RVWHG E\ 3URI $QWMH +HHVH 'HSDUWPHQW RI %LRF
0HQGR]D &R]DWO 'LYLVLRQ RI 3ODQW 6FLHQF

9LVLW SHULRG 6HSWHPEHU WR 1RYHPEH

5HFHQW VWXGLHV VKRZ WKDW D IXQFWLRQDO YHVLFXODU
WUDIILFNLQJ RI SURWHLQV WR DQG IURP WKH 30 SOD\V D
DEXQGDQFH DW WKHLU VLWH RI IXQFWLRQ QDPHO\ WKH
SDWKZD\V WKDW FRQWULEXWH WR PRGXODWLQJ WKH FRP
HQGRF\WRVLV ,Q VHFUHWLRQ QHZO\ V\QWKHVL]HG SU
(QGRSODVPLF 5HWLFXOXP (5 WR WKH 30 (QGRF\WRVLV
SODQW FHOOV UHPRYH DQG LQWHUQDFPLUÀ WHUQDFPE p
SOLF0P O

,RQRPLF SURILOLQJ 3URI 0HQGR]D &R]DWO RI WUD SODQWV H[SRVHG KHDY\ PHWDO LGHQWLILHG LQ

5HVXOWV IURP DQG ZRXOG JLYH XV DQ LGHD RI Z LRQV PD\ EH DIIHFWHG LQ HDFK RI WKH PXWDQWV 7KL WDUJHWHG DSSURDFK WR VWDUW WUDQVIRUPLQJ WKH PX ZLWK IOXRUHVFHQW SURWHLQV WR ORRN DW SRWHQWLDO OLYH FHOO LPDJLQJ 3URI +HHVH¶V ODE DV ZHOO DV ZL 0HQGR]D &R]DWO V RSWLPL]HG ODE DSSURDFKHV

$V D ORQJ WHUP JRDO ZH DOVR H[SHFW WR SXUVXH :DOWHU *DVVPDQQ 08 ,3* WR WHVW WKH WUDQVSRUWH RRF\WHV 7KHVH H[SHULPHQWV ZLOO JLYH XV LQVLJKW LQ RI VHOHFWHG KHDY\ PHWDO WUDQVSRUWHUV

$IWHU JURZLQJ YHVLFOH PXWDQWV ZLWK RU ZLWKRX LVRODWH HQULFKHG SODVPD PHPEUDQHV ZLWK RU ZLWK W\SH DQG WKHQ LQ FROODERUDWLRQ ZLWK 3URIH6FRWW TXDQWLWDWLYH PDVV VSHF DQDO\VHV WR FRPSDUH WK GLIIHUHQW JURZWK FRQGLWLRQV

WpH 0 U R ZWK H Original project timeline D_7°H àHG SHUIRUPUP @Y 97°H ºêHƒ FRPSDUD

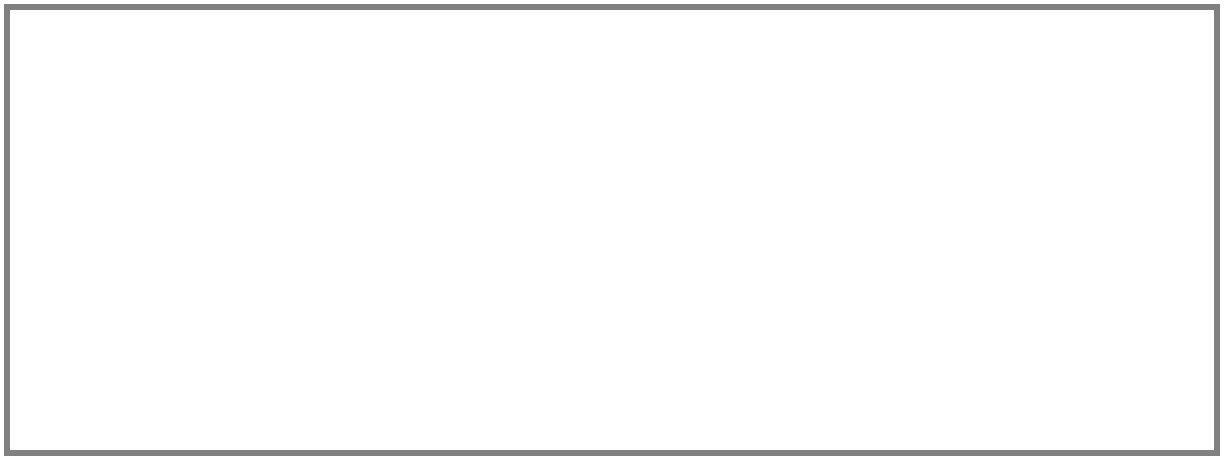
Deviations from the original timeline

'XH WR SHUVRQDO FRPPLWPHQWV LQ 'U .H\VWHU¶V VFKH
WULS WR &ROXPELD 0LVVRXUL 7KHUHIRUH 6HSWHPEHU &D
DQG DUULYHG LQ &ROXPELD 6HSWHPEHU 7KH +H WKH
0LVVRXUL RQ WKH 1RYHPEHU DQG DUULYHG EDFN LQ &D
RI 1RYHPEHU 'XH WR WKH VKRUWHU VFKHGXOH ZH G
VFUHHQLQJ RI WKH PXWDQWV RQ WKH VDPH GD\ DV WKH K

Preliminary Results obtained

Screening the vesicular trafficking (*ves*) mutants for Cd tolerance

7KH FRQWURO &RO *ves5* DQG *ves6* *ves4* PXWDQWV *ves6* *ves4* ZHUH VSRQVRUH
E\ 3URI +HHVHH *opt3-2* 7KH FRQWURO IRU LURQ GHILFLHQF\ DQG
ZDV VSRQVRUHG E\ 3URI 0HQGR]D &R]DWO 6HHGV ZHUH
RQO\ SODWHV LURQ GHILFLHQW SODWHV FRQWDLQLQJ
SODWHV FRQWDLQLQJ —0 &G 6HHGV ZHUH DOORZHG
FRQWUROOHG HQYLURQPHQWDO FKDPEHUV $IWHU GD
GLJLWDO SKRWRJUDSK\ )LJ :H REVHUYHG JUHHQ OHD
DV ZHOO DV SURSHU URRW IRUPDWLRQ)XUWKHUPRUH D
LURQ GHILFLHQW SODWHV *ves6* *ves4*)LJ SHUIRUPHG EHWWHU WKDQ
GHILFLHQW FRQGLWLRQV :KHQ WKH SODWHV *ves5* *ves4* ZHUH H[SR
ZHUH REVHUYDEO\ PRUH WROHUDQW WR FDGPLXP WKH
H[SHFWHG *vti3-2* ZDV VHQVLWLYH WR ERWK LURQ GHILFLHQF\ DQ

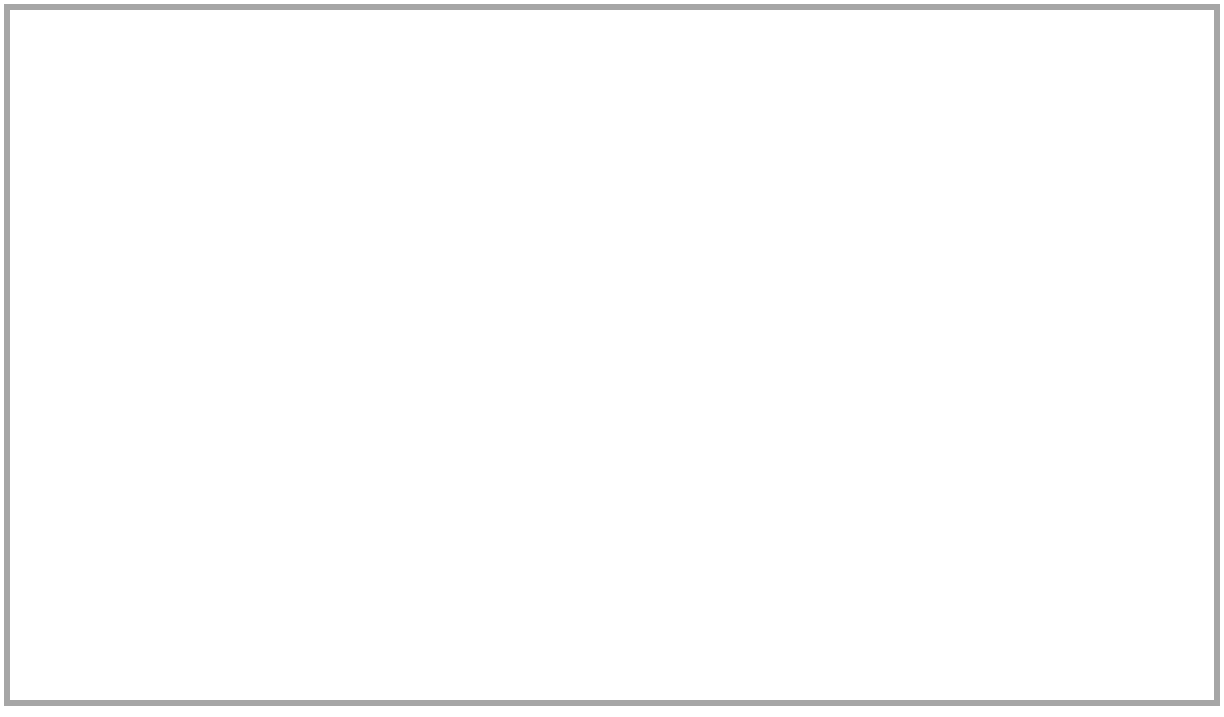


)LJ 5HSUHVHQWDWLYH 0XUDVKLJH DQG 6NRRJ 0HGLXP ZLWK DJDU RQO\ 7KH ,QI
SODWH FRQWDLQV 0XUDVKLJH DQG 6NRRJ PHGLXP ZLWK DJDU RQO\ 7KH ,QI
DQG 6NRRJ PHGLXP ZLWK DJDU DQG VXSSOHPHQWHG ZLWK)HUURXV 7KH
PHGLXP ZLWK DJDU DQG VXSSOHPHQWHG ZLWK &X6 7KH
VHHGV ZHUH SODWHG DQG JURZQ IRU GD\V DW f& LQ D K OLJKW K
FKDPEHUV

Microscopic observation of screened vesicular trafficking (***ves***) mutants

$IWHU GLJLWDO SKRWRJUDSK\ SODQWV ZHUH VXEMHFWH
WKH SKRWRJUDSK\ REVHUYDWLRQV SODQWV ZHUH JURHG
V\QWKHVLV+R)ZHYHU ZKHQ SODQWV ZHUH H[SRVHG WR LU
OHDI \HOORZLQJ ZDV REVHUYHG REVHUYHG \HOORZLQJ ZKH
H[SRVHG WR FDGPLXP)LJ EXW QRW DV VHYHUH DV LQ

)LJ 5HSUHVHQWDWLYH PLFURVFRSLF LPDJHV RI 0XUDVKLJH DQG 6NRRJ (
ves PXWDQWV



)LJ 5HSUHVHQWDWLYH PLFURVFRSLF LPDJHV RI 0XUDVKLJH DQG 6NRRJ
)HUUR]LQH XVHG IRU *ves* SHFWURSKRWRPHWULF RI

)LJ 5HSUHVHQWDWLYH PLFURVFRSLF LPDJHV RI 0XUDVKLJH DQG 6NRRJ P
XVHG IRU SKHQRW\SLF DQDO\VLV

Fresh weight measurement of ***ves*** mutants

:H LQYHVWLJDWHG WKH HIIHFW RI LURQ GHILFLHQF\ DQ
ELRPDVV REWDLQHG DIWHU GLJLWDO SKRWRJUDSK\ )LJ
:H XVHG WKUHH VHHGOLQJV RI VLPLODU VL]H WR PHDVX
:H FDOFXODWHG WKH DYHUDJH IUHVK ZHLJKW RI WKH VH
VHWV RI VHHGOLQJV Q *ves5* H DQG *ves4* ZHUH WKH
&RO DQG *ves4* ZHLJKHG WKH OHDVW RI DOO WKH
)XUWKHUPRUH *ves3* H[SHULHQFHG PRVW VHQVLWLYH WR LURQ
IROORZHG E\ *ves6* *ves4* PXWDQW ZDV WKH PRVW WROHUDQW WR
WKH *ves5* PXWDQW

)LJ 6HHGOLQJ IUHVh*ves*ZPXWDQWV WUHP FRQWURO)HUU]LQH DQG &G 06
ZHLJKHG DV VHWV RI DQG DW OHDVW UHSOLFDWHV ZHUH PHDVXUHG SHU

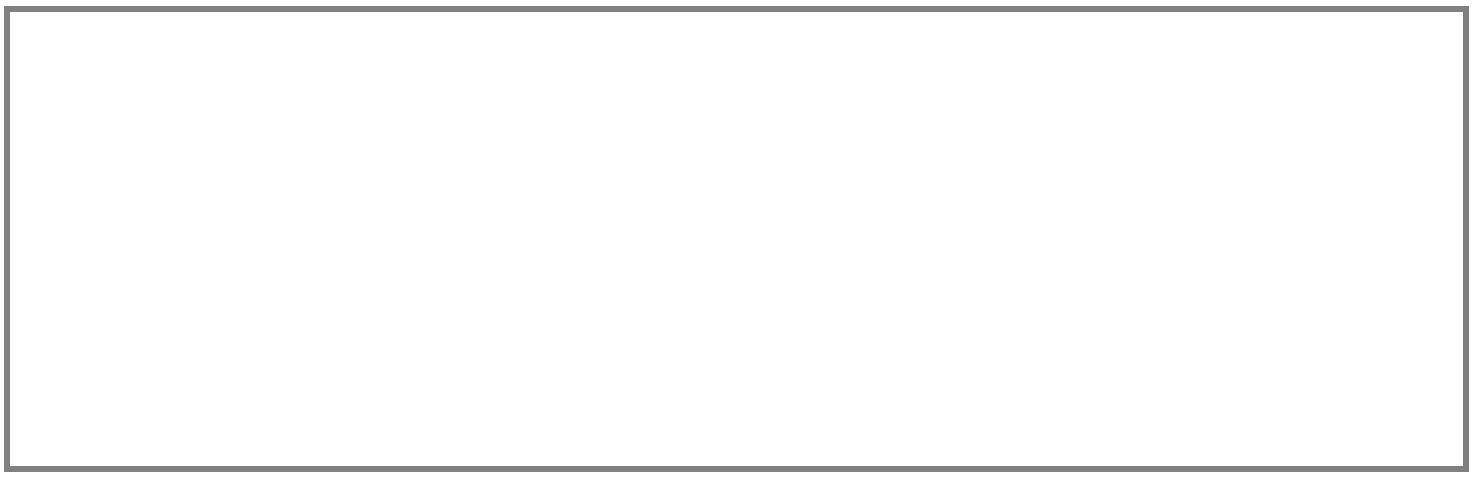
Screening of a second set of ***ves*** mutants for Cd tolerance

6HHGV ZHUH VWHULOL]HG DQG SODWHG RQ 06 RQO\
FRQWDLQLQJ)HUU]LQH DQG FDGPLXP FRQWDLQLQJ S
)XUWKHUPRUH VHHGV ZHUH DOORZHG WR JHUPLQDWH
HQYLURQPHQWDO FKDPEHUV K OLJKW K GDUN F\FOH
SURFHVVHG E\ GLJLWDO SKRWRJUDSK\ )LJ :H REVHU
SODWHG RQ WKH 06 SODWHV :H REVHUYHG SURSHU UR
H[FHSW *ves3* PXWDQWV PXWDQW VKRZHG D ³VPDOO URRW
SODWHV)XUWKHUPRUH *ves1* &GLG QRW SHUIRUP ZHOO RQ WK
SODWHV *ves3* DQG *ves2* SHUIRUPHG REVHUYDEO\ EHWWHU XQ
:KHQ WKH SODQWV ZHUH H[SRVHG WR FKORURVH PRUH
WROHUDQW WR LURQ GHILFLHQF\ ZHUH REVHUYDEO\ PRU
WKH PXWDQWV $JDLQ *ves3* DQG *ves2* H[KLELWHG VLPLODU
FDGPLXP VWUHVV

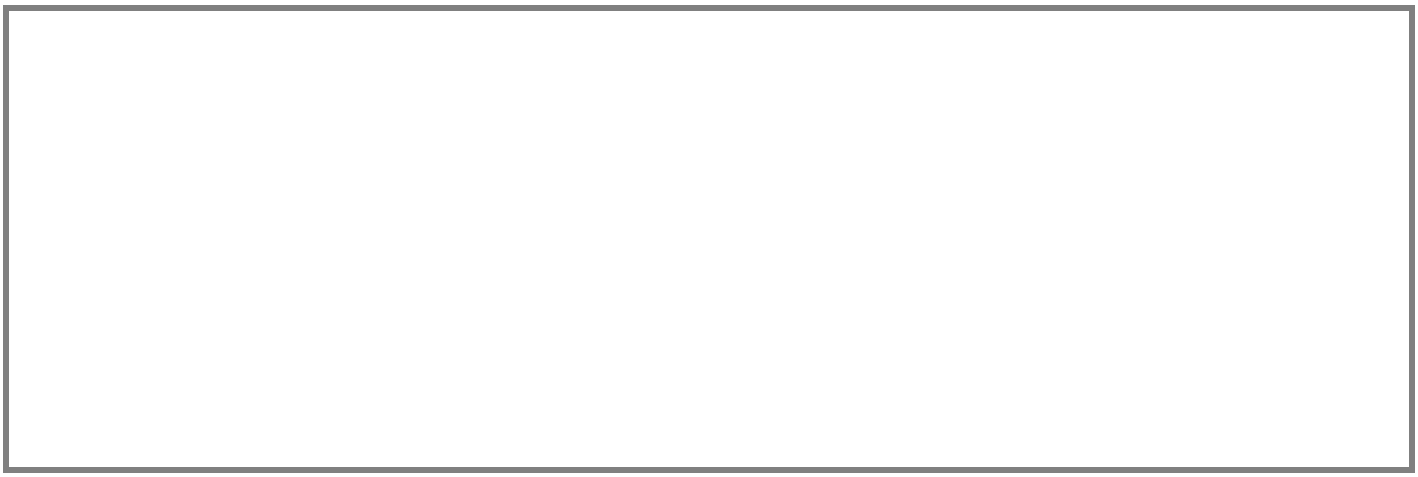
)LJ 5HSUHVHQWDWLYH PLFURVFRSLF LPDJHV RI 0XUDVKLJH DQG 6NRRJ 06
PXWDQWV ve01XDQW2V

)LJ 5HSUHVHQWDWLYH PLFURVFRSLF LPDJHV RI 0XUDVKLJH DQG 6NRRJ
)HUURQH XVHG IRU ve1,ve2 DQG ve3 0XWDQWV

)LJ 6HHGOLQJ IUHVK ZHLJKW RI WKH VHW PXWDQWV IURP FRQWURO)HU
ZHLJKHG DV VHWV RI DQG DW OHDVW UHSOLFDWHV ZHUH PHDVXUHG SHU

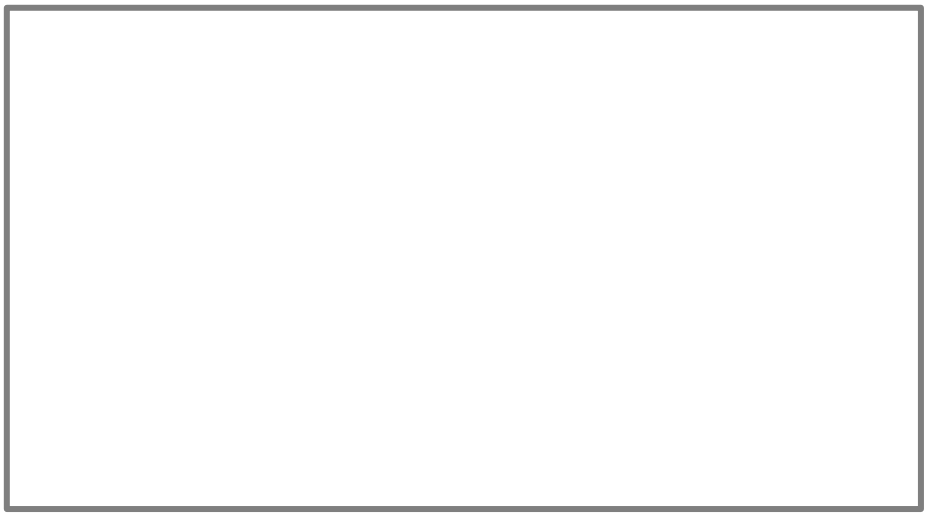


)LJ ,&3 2(6 TXDQWLILFDWLRQ RI &G DQG)H DIWHU



)LJ ,&3 2(6 TXDQWLILFDWLRQ RI &XWes3 PZDWDSURSDJDWHG LQ K\GURSRQLF DQG FG VROXWLRQV

)RU]LQF FRQWHQW ZH REVHUYHG PRUH]Ves3 ODWKHV &R XQGHU FRQWURO FRQGLWLRQV FRPSDUHG WR FG GLG QRW VWDWL ERWK SODQWV ZHUH H[SRVHG WR FDGPLXP 1RQHWKHOHV]LQFves3 URRWV WKDQ LQ &RO URRWV XQGHU FRQWURO SODQWV ZHUH H[SRVHG WR FDGPLXP ZH REVHUYHG D GHF



)LJ ,&3 2(6 TXDQWLILFDWLRQ RI &XWes3 PZDWDSURSDJDWHG LQ K\GURSRQLFV F VROXWLRQV

Other outputs

3. Mizzou events - Dr. Marshall Keyster seminar

H L $rU % øNbËì1 Nì@ 4(r)-.er y-mersn-6-3(.)]TJ EMC /P <</MCI5 0 >>BDC /C2 0 12
K W W S F D O H 9 d` @U 5 F 5cV5$€ DU LF 0 T p D HEW 6F WBD O 0 cBDV0 Â€ •F 0