Figure S1

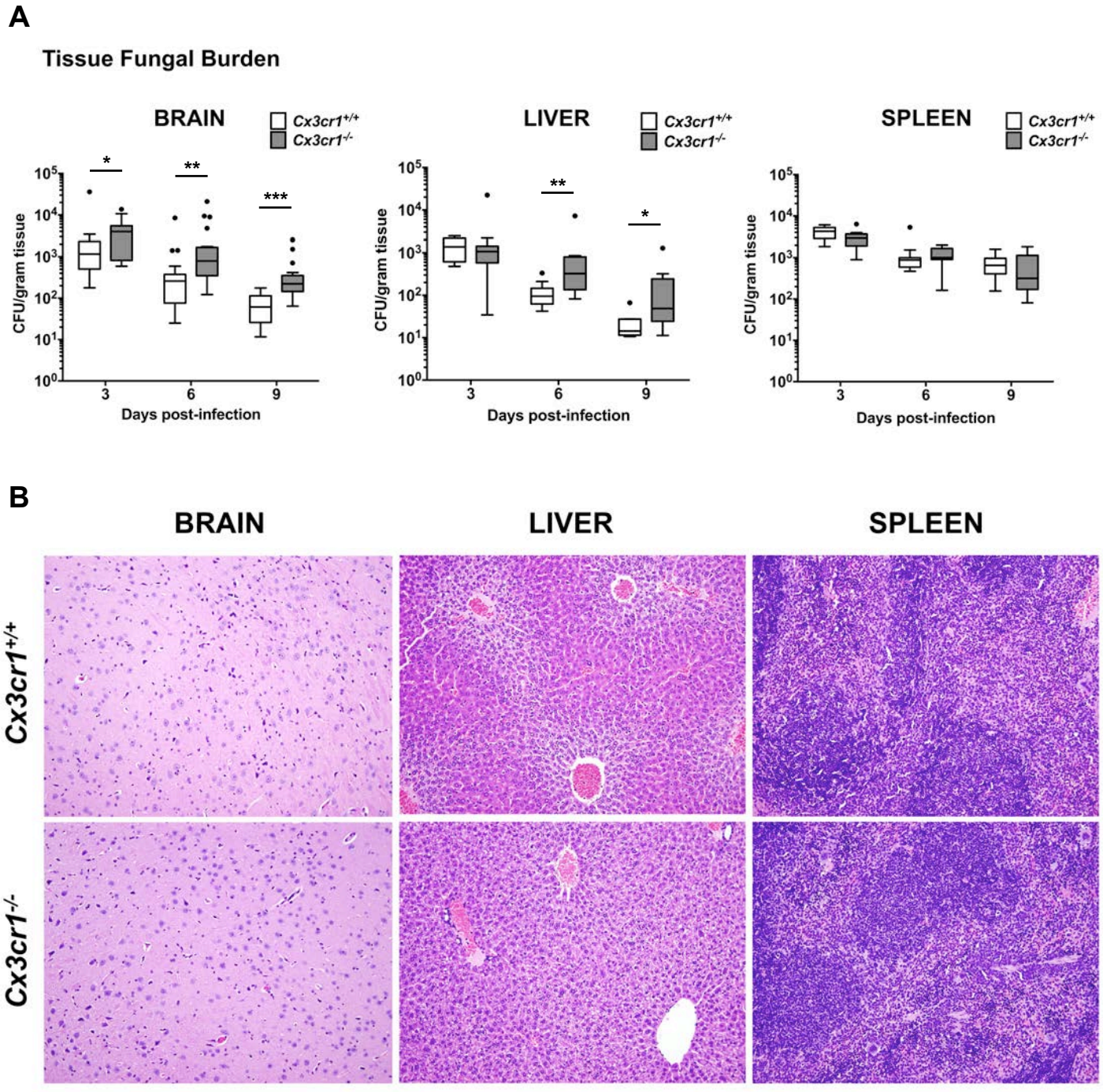

Figure S1. Tissue fungal burden and histology in brain, liver and spleen of *Candida*-infected WT and *Cx3cr1*$^{-/-}$ mice. (A) Fungal burden. $^{*}P < 0.05$; $^{**}P < 0.01$; $^{***}P < 0.0001$ (n = 10-21; two to three independent experiments). (B) Hematoxylin and Eosin staining (day 6 post-infection). Magnification, 200× (n = 8; two independent experiments).

Figure S2

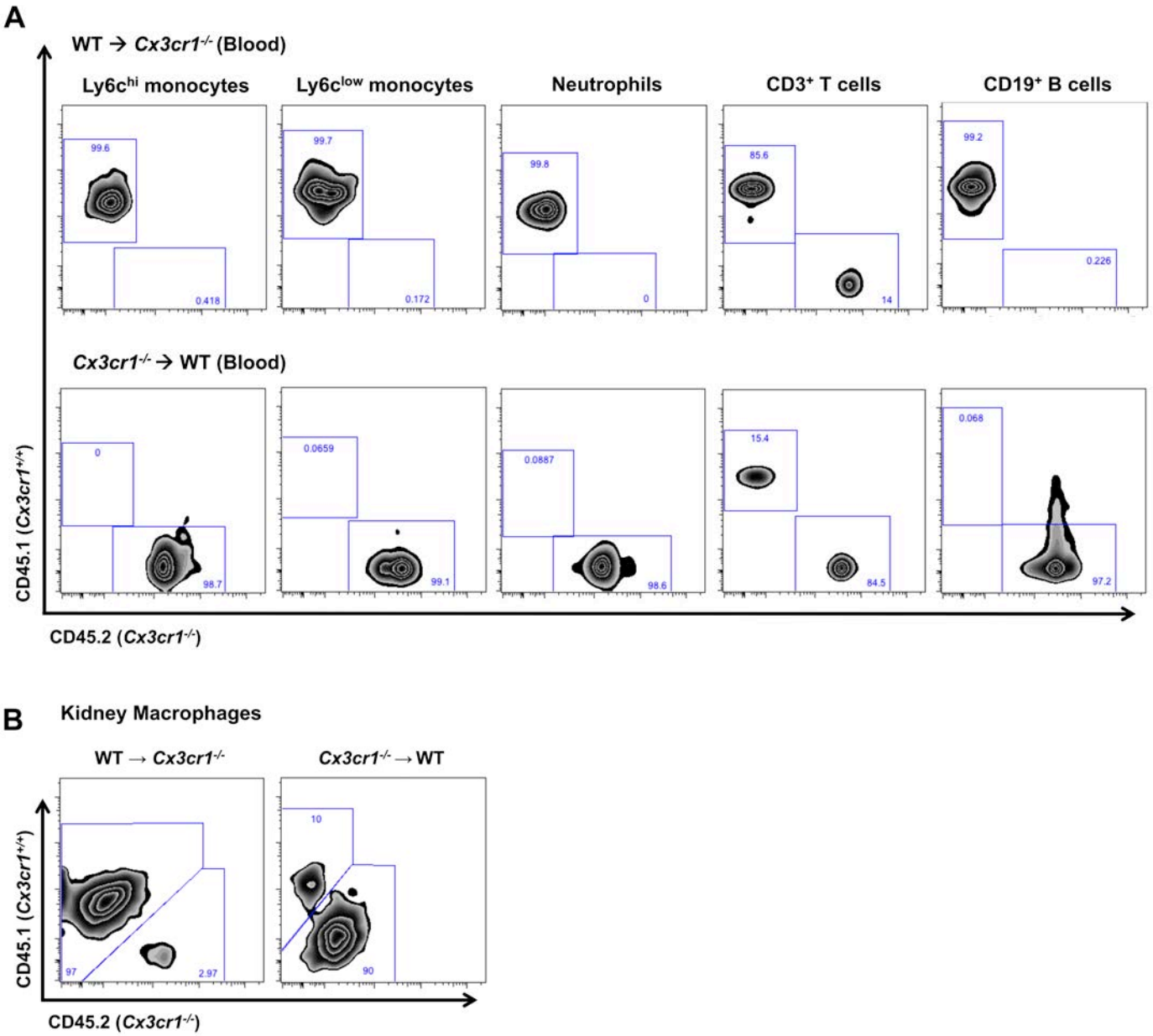

Figure S2. Engraftment of bone-marrow radiation chimeras before *Candida* infection. Whole bone marrow cells from WT (CD45.1$^+$) and Cx3cr1$^{-/-}$ (CD45.2$^+$) congenic mice were cross-transferred to irradiated recipients and evaluated for engraftment 8 weeks later. (A) Blood. Each column of FACS plots corresponds to the subpopulation labeled at the top, analyzed for CD45.1 and CD45.2 expression. WT donor→Cx3cr1$^{-/-}$ recipient mice (upper panel); Cx3cr1$^{-/-}$ donor→WT recipient mice (lower panel). Data are representative of 25 mice per group. (B) Kidney macrophages. FACS plots of kidney leukocytes purified from the indicated chimeric mice and gated on MHCII^{hi}F4/80^{hi}CD11b$^+$ cells. Data are representative of 4 mice.

Figure S3

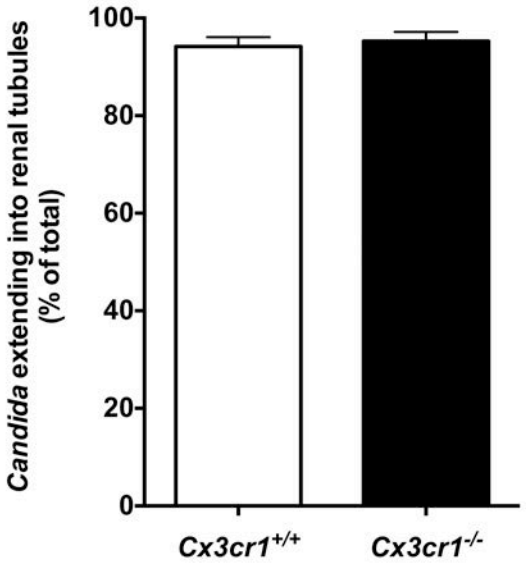

Figure S3. *Candida* pseudohyphae rapidly extend into the renal collecting system by 24 hours after infection. Percent of *Candida* pseudohyphal structures that have extended into renal tubules at day 1 post-infection in *Cx3cr1*$^{+/+}$ and *Cx3cr1*$^{-/-}$ mice (n = 11-13; two independent experiments; P = 0.68).

Figure S4

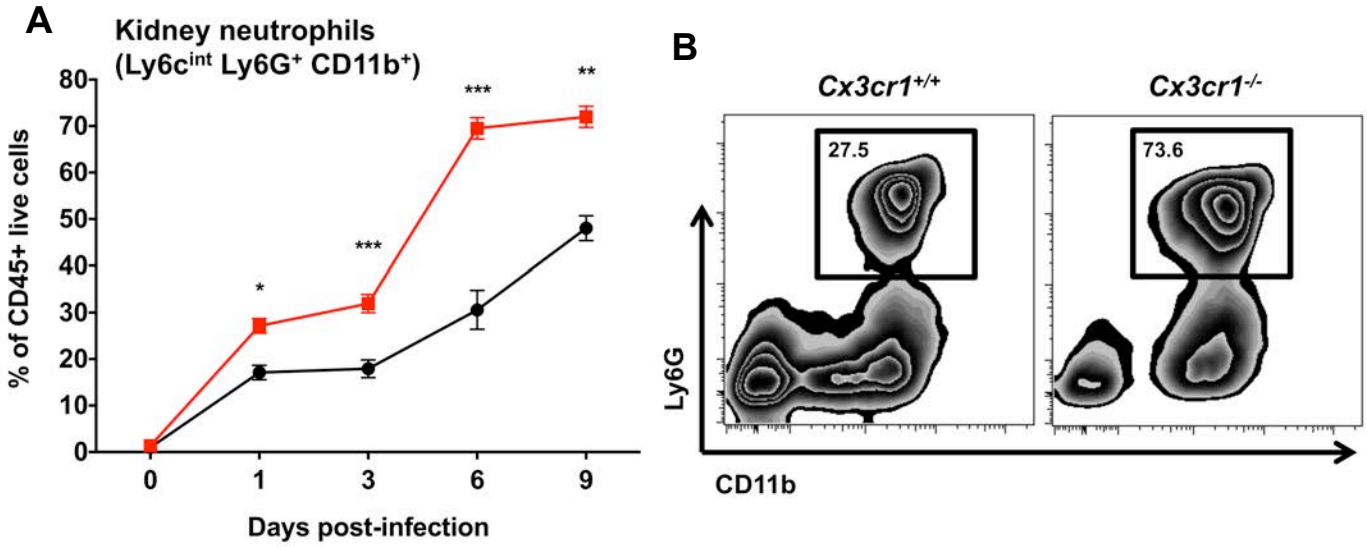

Figure S4. Cx3cr1 deficiency results in increased accumulation of neutrophils in the kidney after *Candida* infection. (A) Percent of total CD45$^+$ leukocytes represented by Ly6C^{int}Ly6G$^+$CD11b$^+$ neutrophils that accumulated in *Cx3cr1*$^{+/+}$ and *Cx3cr1*$^{-/-}$ kidneys at different time-points after infection. $^*P < 0.01$; $^{**}P < 0.001$; $^{***}P < 0.0001$ (n = 9-15; three to four independent experiments). (B) Representative FACS plots demonstrating increased frequency of Ly6C^{int}Ly6G$^+$CD11b$^+$ neutrophils in *Cx3cr1*$^{-/-}$ kidneys at day 6 post-infection (n = 9-15; three to four independent experiments).

Figure S5

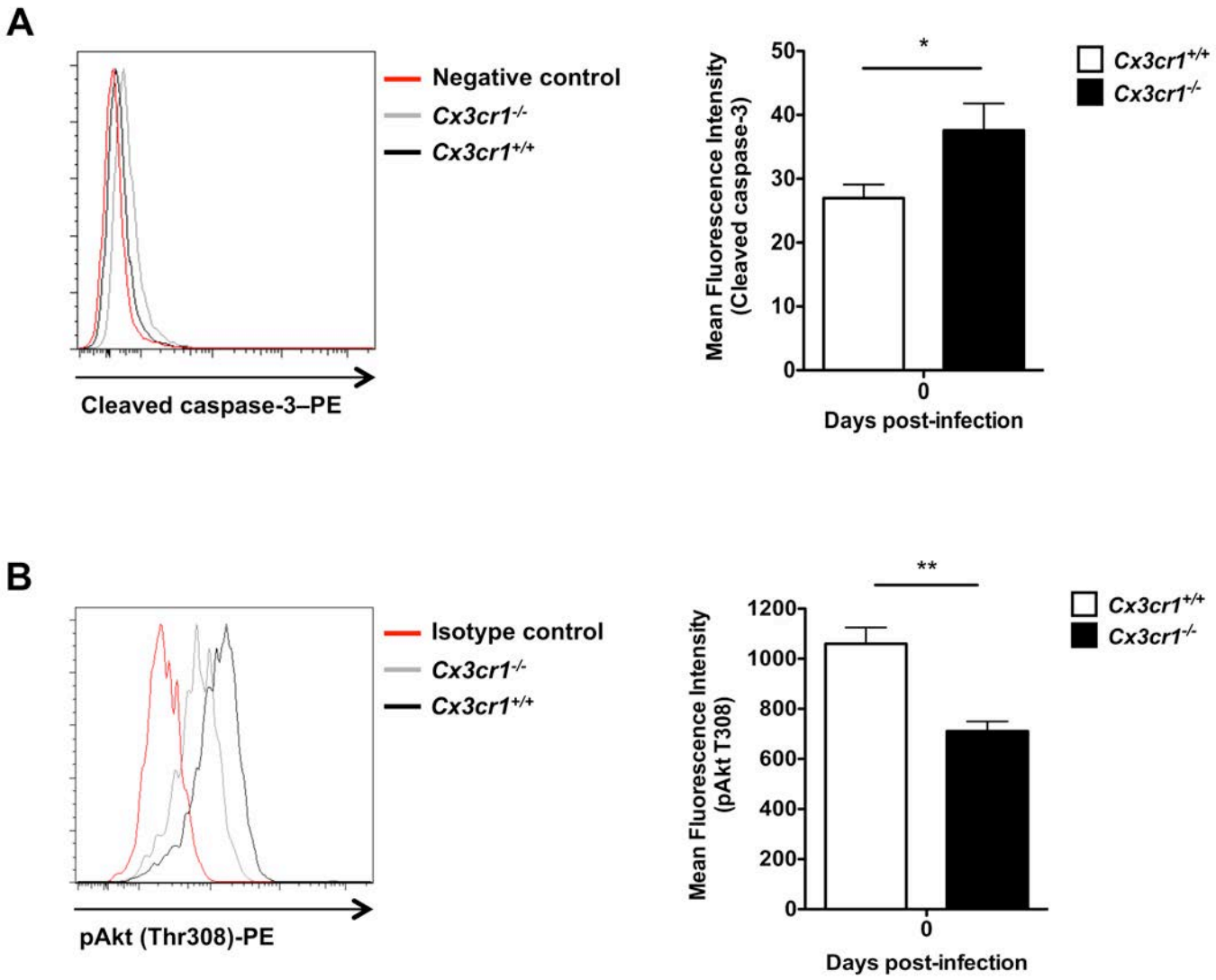

Figure S5. Cx3cr1 deficiency is associated with increased caspase-3 cleavage and decreased Akt phosphorylation in kidney resident macrophages. (A) Decreased expression of cleaved caspase-3 in *Cx3cr1*$^{+/+}$ kidney resident macrophages from uninfected mice. Left, representative FACS histogram. Right, summary mean fluorescence intensity data obtained by FACS. $^{*}P<0.05$ (n = 9; three independent experiments). (B) Increased expression of pAKT (Thr308) in *Cx3cr1*$^{+/+}$ kidney resident macrophages from uninfected mice. Left, representative FACS histogram. Right, mean fluorescence intensity data obtained by FACS. $^{**}P<0.01$ (n = 6). Data are shown from one of two independent experiments with similar pattern of results. All quantitative data are mean ± SEM.

Table S1. Relative abundance of leukocyte subsets and their Cx3cr1 expression in the kidney at steady state and after *Candida* infection.

Leukocyte subset*	% of total $CD45^+$ leukocytes**					% of total $Cx3cr1^+$ leukocytes***		
	Day post-infection							
	0	1	3	6	9	0	3	6
Neutrophils	0.5-1	12-21	14-28	17-61	25-60	N/A	N/A	N/A
Monocytes	3-5	35-39	12-39	4-11	4-10	6-15	21-30	11-17
Macrophages	53-67	16-21	25-47	26-47	15-33	77-87	55-65	62-69
Dendritic cells	9-12	7-14	4-9	3-11	2-5	3-7	9-13	10-14

N/A, not applicable

* Neutrophils were defined as $Ly6C^{int}Ly6G^+CD11b^+$ cells, monocytes were defined as $MHCII^-F4/80^{int}CD11c^-CD11b^+$ cells, macrophages were defined as $MHCII^{hi}F4/80^{hi}CD11c^{low}CD11b^+$ cells, and dendritic cells were defined as $MHCII^{hi}CD11c^{hi}F4/80^{low}$ cells.

** Range based on 9-15 animals tested (three to four independent experiments).

*** Range based on 4-6 animals tested (two independent experiments).

Table S2. Number of candidemic subjects and non-infected controls from the two patient cohorts analyzed in the present study.

	Candidemia Cases n = 281	Non-infected Controls n = 384	Total
RUNMC			
Caucasian	37	167	204
DUMC			
Caucasian	166	156	322
African American	78	61	139
			665

RUNMC, Radboud University Nijmegen Medical Center; DUMC, Duke University Hospital

Table S3. Demographic and clinical characteristics of DUMC subjects

	No Systemic Candidiasis n = 217	Systemic Candidiasis n = 244	p value
Male gender	112 (51.6%)	136 (55.7%)	0.376
Mean Age (years) adult (n = 443) pediatric (n = 18)	 58.7 (56.4-60.9) --	 52.5 (54.0-58.3) 6.7 (3.6-9.9)	0.11
Solid organ transplant	6 (2.8%)	35 (14.3%)	<0.0001
Malignancy	49 (22.6%)	69 (28.3%)	0.162
Chemotherapy	26 (12.0%)	37 (15.2%)	0.321
Neutropenia	6 (2.8%)	23 (9.4%)	0.0033
Surgery within last 30 days	121 (55.8%)	111 (45.7%)	0.0310
Receipt of total parenteral nutrition	7 (3.2%)	50 (21.1%)	<0.0001
HIV	0	3 (1.2%)	0.251*
Dialysis-dependent	16 (7.4%)	29 (12.1%)	0.089
Acute renal failure	49 (22.6%)	84 (34.6%)	0.0047
Immunocompromised state	95 (43.8%)	140 (57.6%)	0.0031
ICU admission within past 14 days	67 (30.9%)	118 (49.6%)	<0.0001
Liver disease	9 (4.3%)	58 (23.8%)	<0.0001
Candida spp. *albicans* *glabrata* *parapsilosis* *tropicalis* *krusei* other >1 *Candida* spp.	N/A	 109 (44.7%) 64 (26.2%) 40 (16.4%) 27 (11.1%) 9 (3.7%) 12 (4.9%) 14 (5.8%)	

*Fishers Exact p value

DUMC, Duke University Hospital; ICU, intensive care unit

Table S4. Association of the mutant CX_3CR1-M280 allele with susceptibility to systemic candidiasis in African-American subjects

	No Systemic Candidiasis n = 61	Systemic Candidiasis n = 78	p value	OR (95% CI)
CC	47(77.1%)	65 (83.3%)	0.354	0.671 (0.289-1.560)
CT + TT	14 (23.9%)	13 (16.7%)		

Table S5. Sequences of the primers used for qPCR with SYBR Green in the present study.

Gene product	Primer name*	Primer sequence (5' → 3')	Amplicon length (bp)
Gapdh	Gapdh F	aactttggcattgtggaagg	223
	Gapdh R	acacattgggggtaggaaca	
Cxcr2	Cxcr2 F	ggtggggagttcgtgtagaa	201
	Cxcr2 R	cgaggtgctaggatttgagc	
Cxcl1	Cxcl1 F	gctgggattcacctcaagaa	180
	Cxcl1 R	tctccgttacttggggacac	
Cxcl2	Cxcl2 F	aagtttgccttgaccctgaa	180
	Cxcl2 R	aggcacatcaggtacgatcc	
Il-4	Il-4 F	cctcacagcaacgaagaaca	155
	Il-4 R	atcgaaaagcccgaaagagt	

*F, forward; R, reverse.
Taqman primers and probes for Gapdh, Il-1β, Il-6, Ccr2, Cx3cr1 and Cx3cl1 were purchased from Applied Biosystems and their sequences are not available.