**Supplementary Methods**

## **NOD mice infection**

NOD and NOD.Cg-*PrkdcscidIl2rgtm1Sug*/Shi.Jic (NOD/*scid-IL2rg–/–*)mice were purchased from CLEA Japan (Tokyo, Japan). All mice were maintained under specific-pathogen free conditions with gamma ray-sterilized diet and acidified tap water (0.002 N HCl) *ad libitum*. Animals were infected intravenously in the lateral caudal vein with 5 x105 *A. fumigatus* conidia resuspended in 100 μL of PBS. Animals were monitored daily for body weight loss and survival evaluation.

**Neutrophil depletion**

For assays with neutrophil depletion, wt or *Clec7a*–/– mice were treated intraperitoneally with *InVivo*Plus anti-mouse Ly6G antibody (clone 1A8) or *InVivo*Mab rat IgG2a isotype control (clone 2A3) (Bio X Cell, Vermont, USA) as described in online suppl. Figure 7.

## **Intratracheal infection**

Animals were sedated and the trachea was accessed through the mouth opening with the help of a 20Gx 1 1/4” indwelling needle intravenous cannula (Terumo, Tokyo, Japan). 1 x107 conidia cells were resuspended in 30 μL of PBS and animals passively inhaled the fungal inoculum. The residual liquid inside the needle was flushed into the trachea by the application of 1mL of air. Animal weight and survival was monitored daily.

 For BALF collection, animals were euthanized by sedation and injected with 1 mL of ice-cold PBS through the trachea. Recovered fluid was centrifuged at 14 000xg / 5minutes and supernatants were used for measurements. For organ analysis, mice were euthanized by cervical dislocation and tissues were also perfused with ice-cold PBS. Organs were harvested, weighted, and macerated in PBS through mesh sieves. Dilutions of the macerates were plated on PDA plates (Eiken Chemical, Tokyo, Japan), incubated at 30°C for two days and recovered colonies-forming units (CFU) were counted. Fungal burden was expressed as CFU per gram of organ. Lung macerates were centrifuged at 14 000xg / 5 minutes and supernatants were used for cytokine analysis.

## **Fc chimeras and binding assay**

The extracellular domain of murine dectin-1 and dectin-2 were cloned into a pIRES bleo3 vector containing the Fc portion of human IgG2 and constructs were expanded by transforming into *E. coli* JM109 competent cells. The plasmids were transfected into HEK293 cells, and the Fc chimeras were purified from culture supernatants by affinity chromatography on Protein A Sepharose columns (GE Healthcare, Uppsala, Sweden). As a control for binding assays, “empty” Fc proteins (without insertion of CLR domains) were expressed and used in parallel.

 For binding assessment, 100 μg of swollen conidia was coated on a 96 well-plate. Wells were blocked with BSA and incubated with Fc chimeras (dilution range: 10~0.005 μg/mL) in 1 mM CaCl2, 150 mM NaCl, 2 mM MgCl2 Tris-buffered solution (pH 7.0) overnight at 4°C. Fc proteins were detected by using a peroxidase-conjugated anti-human Fcγ IgG antibody (Jackson ImmunoResearch, Pennsylvania, USA) and TMB colorimetric substrate (SeraCare, Massachusetts, USA). Optical density was measured at 450 nm.

**Reporter Cells**

Dectin-1 and dectin-2 reporters were 43-1 cells (NFAT-GFP reporter gene) engineered to express the murine extracellular portion of the corresponding receptor fused with the FcRγ chain. Control reporter cells (FcRγ chain without fusion) were run in parallel. Until used, cells were maintained in 10% inactivated Fetal Bovine Serum (Biosera, Nuaillé, France) / RPMI-1640 medium (Fujifilm Wako, Osaka, Japan). For the assay, cells were plated in a 96 well-plate at a density of 3 x104/100 μL medium per well and stimulated with formalin-fixed swollen conidia. After overnight incubation, GFP expression was assessed by flow cytometry.

## **Peritoneal macrophages**

Peritoneal macrophages were induced by injection of 3% thioglycolate solution (Nissui, Tokyo, Japan) into the peritoneal cavity of wt or *Clec7a*–/–- *Clec4n*–/– mice. After 4 days, peritoneal lavage was collected by injection of 5 mL of cold PBS and recovered cells were stained for flow cytometry analysis.

**R analysis**

Gene expression data was gathered from the public database Expression Atlas (data under Creative Commons Attribution 4.0 International License). Gene expression levels were expressed as transcripts per kilobase million (TPM) and were presented as heatmaps, constructed in the R software (v. 3.6.0 for Mac OS X, with the package plots).