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Electronic Supplementary Material

House Dust Mite Extract Forms a Der p 2 Corona on Multi-Walled Carbon Nanotubes: Implications for Allergic Airway Disease

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Fig. S1 Removal of unbound HDM, referred to as washing, consisted of centrifugation (14,000 rpm, 15 minutes), removal of supernatant, and resuspension in PBS. A. Protein concentration of the supernatant was measured with a colorimetric assay (660 nm protein assay). Solid line shows the average absorbance spectrum. Shading shows standard deviation. (n=3 distinct samples). B. The majority of the unbound HDM is removed in the first wash. Three wash steps are used to ensure all unbound protein is removed. The background signal from the 660 nm reagent was subtracted from each sample prior to calculating protein concentrations. Significance was calculated using an ANOVA with a post-hoc Tukey test. *p < 0.001, ns = not significant.

Table S1. Spectral counts, reported as a percent of total signal, for the excised 15 kDa band of the HDM-MWCNT corona (Fig. 2B). Proteins with spectral counts >1% are listed.

Protein	MW (kDa)	15 kDa (%)
Der p 2 (NPC2 family)	14	81.6
Thioredoxin	12	9.70
Der p 36 (C2 domain containing protein)	25	3.51
Nucleoside diphosphate kinase	18	1.56

Table S2. Spectral counts, reported as a percent of total signal, for the excised 12 kDa band of the HDM-MWCNT corona (Fig. 2B). Proteins with spectral counts >1% are listed.

Protein	MW (kDa)	12 kDa (%)
Der p 30 (ferritin)	12	71.0
Fructose-bisphosphate aldolase	39	9.84
Der p 11 (paramyosin)	102	5.22
Uncharacterized protein LOC113797715	191	4.71
Glutathione peroxidase	25	1.88
Myosin heavy chain	112	1.73
Inosine-5'-monophosphate dehydrogenase	56	1.51



Fig. S2 Western blotting using an antibody against der p 2 confirms the band visible at ~15 kDa is der p 2 (14 kDa).

Table S3. Normalized abundance (% of total protein) of the top-10 proteins identified in HDM and the HDM corona formed on MWCNTs (HDM-MWCNT). Proteins present in a sample, but not in the top-10, are shown in italics. Proteins with normalized abundance < 0.1% are not shown. The most abundant protein in each sample is bolded. Mean and standard deviation are reported (n=3). A different lot (#390991) of HDM was used in comparison to the data in the main text (Table 2). For both lots, der p 2 is the most abundant protein in corona.

Protein	MW (kDa)	HDM (%)	HDM-MWCNT (%)
Der p 2 (NPC2 family)	14	24.5 ± 5.20	51.6 ± 15.6
Der p 1 (cysteine protease)	34	23.6 ± 0.98	4.72 ± 3.69
Der p 30 (ferritin)	12	8.50 ± 2.62	4.22 ± 1.17
Uncharacterized protein LOC113799616	25	7.28 ± 2.31	10.7 ± 9.76
Uncharacterized protein LOC113797715	191	7.04 ± 2.13	0.71 ± 0.29
Uncharacterized protein LOC113790193	27	3.48 ± 1.80	3.12 ± 1.67
Fatty acid-binding protein-like	15	2.19 ± 0.39	2.76 ± 2.48
Nucleoside diphosphate kinase	18	1.91 ± 0.82	3.92 ± 1.78
Uncharacterized protein LOC113796509	77	1.85 ± 1.80	-
Der p 10 (tropomyosin)	33	1.28 ± 1.07	0.74 ± 0.85
Der f 22 (Group 2-like)	18	0.47 ± 0.77	3.35 ± 3.32
Der p 11 (paramyosin)	102	0.25 ± 0.056	1.96 ± 1.57
Lysosomal aspartic protease-like	45	0.13 ± 0.19	1.09 ± 1.84