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ORIGINAL ARTICLE

The prevalence of IgE positivity, environmental factors, and clinical implications associated with mite species in allergic patients in Taiwan

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Abstract

Background: Allergic diseases are a growing public health concern with increasing prevalence and severity. Allergens play significant roles in triggering immune responses and the development of allergic reactions.

Objective: Investigate the presence and clinical significance of dust mites, storage mites, and predatory mite *Cheyletus eruditus* (Ce) in household environments.

Methods: A survey of household dust was performed to determine mite occurrence and analyze influencing factors, an analysis of the correlation between mite species and allergic symptoms, and basophil activation triggered by mite allergens. Cross-reactivity between Ce and house dust mites was assessed.

Results: The high appearance rate of mite species in households of Taiwan was *Dermatophagoides pteronyssinus* (Dp) and *D. farinae* (Df). Environmental factors such as pet keeping, vacuum cleaner usage, air conditioner usage, proximity to the kitchen, cleaning frequency, and protein concentration in beds were shown to influence mite prevalence. The appearance of Dp and Df significantly increased the occurrence of airway and nasal symptoms, while the presence of Ce was strongly correlated with skin symptoms. The activation of basophils and the correlation between specific IgE levels and allergic symptoms in response to Ce exposure were demonstrated. The presence of Ce was associated with elevated levels of allergens in bedding. The IgE adsorption between mite species was demonstrated suggesting cross-reactivity between the Ce and Dp was limited. Presence of Ce is associated with elevated levels of major mite allergens in beddings.

Conclusions: Allergenicity of Ce was confirmed by IgE reactivity and basophil activation regarding mite infestation as a potential cause of skin-related allergy.

Key words: Predatory mite, *Cheyletus eruditus*, mite allergy, allergic dermatitis, mite specific IgE, basophil activation

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Introduction

Allergy results from an exaggerated immune response to harmless environmental substances called allergens, leading to a range of allergic reactions from mild to severe.[1] Allergic diseases vary in severity, from mild symptoms to life-threatening reactions,[2] and their increasing prevalence in recent decades poses a significant public health concern.[3] Allergic diseases are showing an alarming trend of increasing severity,[4] with conditions like asthma, rhinitis, and dermatitis becoming more prevalent and severe.[5] The impact of severe allergies on individuals, healthcare systems, and economies is substantial, requiring emergency medical intervention, hospitalizations, and expensive medications.[6]

Allergic diseases develop due to allergen exposure, allergens can induce immune responses, resulting in allergic reactions.[7] Inhalant allergens like mites are associated with respiratory allergies such as allergic rhinitis and asthma.[8] These inhaled allergens triggered immune responses and caused symptoms like nasal congestion, sneezing, wheezing, and coughing. Allergens can also cause allergic conditions like contact dermatitis through direct skin contact.[9] Development of allergic diseases is influenced by allergen exposure, genetic predisposition, immune system regulation, and environmental factors.[10]

Mite exposure allergy involves complex immunological mechanisms and IgE production.[1] These reactions result from immune activation processes, including allergen sensitization, cytokine production, B cell activation, and IgE production.[11] Allergenic components found in mite proteins, feces, dead bodies, saliva, and excrement initiate immune activation upon initial exposure.[12] Subsequent exposure leads to IgE cross-linking, triggering immediate hypersensitivity reactions mediated by mast cells and basophils.[13] Basophil activation plays a crucial role in allergic diseases and can provide insights into allergic reactions.[14] Understanding these immunological mechanisms is vital for developing effective diagnostic and therapeutic approaches for individuals with mite allergies.[15]

Different mite categories exist in residential environments, including house dust mites, storage mites, and predatory mites, each with unique characteristics and allergenic roles.[16,17] House dust mites, *Dermatophagoides pteronyssinus* (Dp), *D. farinae* (Df) and *Blomia tropicalis* (Bt), are the most significant indoor allergens associated with allergy.[17] Storage mites, *Tyrophagus putrescentiae* (Tp), commonly contaminate stored food and grains, causing allergic reactions in exposed individuals. House dust mites and storage mites can induce respiratory and skin symptoms and systemic allergic responses.[18] Predatory mites, *Cheyletus eruditus* (Ce), feed on other small insects or mites, primarily affecting workers in industries like grain handling and pest control, causing occupational allergies.[19]

Predatory mites-Ce have been found infesting birds and sheep in Brazil,[20] and they are commonly found in various dusty environments. Predatory mites flourish in unclean surroundings and can cause allergies and dermatitis in humans and animals through contact or contaminated food.[21] Allergic symptoms have been reported from ingesting Ce-contaminated foods,[18] as well as skin reactions from their bites.[22] Dermatitis caused by Ce has been documented in humans, especially in veterinary settings.[22] Ce acts as a respiratory allergen, leading to persistent nonoccupational allergic rhinitis in subtropical environments.[23] Their presence in residential environments was correlated with the severity and persistence of allergic rhinitis symptoms.[23] These findings emphasize the significance of Ce in triggering allergies and highlight emerging healthcare concerns in humans.

This study aims to comprehensively investigate the presence and clinical significance of Predatory mite Ce in household environments. It includes a survey of mite appearance, analysis of factors influencing mite occurrence, correlation with allergic symptoms, IgE response analysis, evaluation of cross-reactivity, basophil activation assessment, and analysis of the relationship with other mite allergens. The study aims to understand the attributes of allergic symptoms induced by Predatory mite Ce and the degree of cross-reactivity with house dust mite.

Materials and Methods

Dust sample collection, Mite Detection and Species Characterization

Dust samples from bedding of mattresses were collected belonging to 202 allergic patients to investigate the mite species prevalence in households. Dust samples were collected by a corded vacuum cleaner with replaceable dust bag. The sampling area is determined based on the dimensions of a double mattress, which measures 1.52 m × 1.88 m. Each sampling area spent a total of 10 minutes going back and forth to collect dust from the mattress. Samples was mixed with saturated NaCl to separate mites for mite detection and species characterization. Supernatant was placed on a glass slide containing Hoyer's medium for observation. Morphological characteristics from Colloff's textbook were used for mite identification by microscopic examination.[24]

Environmental Condition and Living Habit Survey

A survey was conducted to assess household environmental conditions and their correlation with mite presence. Parameters including pet keeping, vacuum cleaner usage, air conditioner usage, bedroom location, and cleaning frequency were recorded. The survey questionnaire listed in the supplementary material as an appendix. Cleaning habits were categorized into three groups. Protein content on beds was measured to explore its association with mite presence.

Correlation between Mite Presence and Allergic Symptoms

Serum samples were collected from 120 allergic patients, with symptoms including upper and lower airway, and skin symptoms at Taichung Veterans General Hospital. The Institutional Review Board and Ethics Committee approved the study (TCVGH IRB No. CE15141A) and informed consent was obtained. These patients, diagnosed by allergy specialists, exhibited one or more allergic symptoms, including airway symptoms, nasal symptoms, and skin symptoms. Airway symptoms included wheezing, coughing, chest tightness, respiratory distress, and shortness of breath. Nasal symptoms included sneezing, runny or itchy nose, nasal swelling, nasal congestion, and sinus pressure. Skin symptoms included rash, itching, redness and swelling, and skin discoloration.

Basophil Activation Analysis by Flow Cytometry

The KU812 cell line, a human basophilic leukemia cell line, was obtained from Bioresource Collection and Research Center (Hsinchu, Taiwan) and cultured in RPMI 1640 medium. Cells were stimulated with crude extracts of Dp and Ce or normal saline for 4 hours. Diamine oxidase (Dao^+) was used to visualize and quantify histamine release, while anti-CD63 FITC-conjugated antibody detected activated basophils. CD203c, a glycoprotein involved in inflammatory responses, was measured using anti-CD203c PE-conjugated antibody. The percentage of cells positive for CD63 and CD203c determined basophil activation.

Dynamic Monitoring of Intracellular Calcium in Basophils

The fluorescent dye Fluo-4 enables real-time monitoring of calcium concentration. The effects of mite allergens on calcium signaling and basophil activation were examined. KU812 cells were stimulated with mite allergens, and intracellular calcium levels were monitored using FITC-labeled flow cytometry with Fluo-4 staining. Before stimulation, culture medium was removed, and cells were loaded with Fluo-4 calcium dye. Fluorescence at 506 nm was collected using the FITC channel after exciting the Fluo-4 dye at 488 nm.

Mite-Specific IgE Measurement with ImmunoCap System

120 allergic patients and 5 nonallergic individuals were enrolled for mite-specific IgE measurement. Blood samples were obtained from participants at Taichung Veterans General Hospital (TCVGH) in Taiwan. Stored serum samples were analyzed using the ImmunoCap system, a quantitative assay that measures specific IgE levels.

Measurement of Major Allergens Using ELISA

The levels of four major allergens (Der p 1, Der p 2, Blo t 5, and Tyr p 2) were determined using ELISA kits provided by INDOOR Biotechnologies, Inc. (UK). Each allergen-specific ELISA kit (Der p 1, Der p 2, Blo t 5, and Tyr p 2) was used according to the manufacturer's instructions. The optical density of the ELISA data was measured at 450 nm.

Cross-Reactivity Evaluation of Dust Mites and Predatory Mites

Cross-reactivity among dust mites and predatory mites was assessed using an IgE adsorption assay. Serum samples from 97 allergic patients with positive IgE responses against Dp and 25 allergic patients with positive IgE responses against Ce were selected. In the IgE adsorption assay, positive sera (specific IgE > 0.35 KU/L) were incubated with 50 μg crude extracts of mites. The IgE responses in the serum samples before and after adsorption were detected using ELISA, following the same procedures as previously described.

Statistical analysis

Differences between two groups were analyzed using the Mann-Whitney U-test for non-parametric analysis. Statistical analyses were conducted using SPSS, Version 12 (SPSS, Inc., Chicago, IL, USA). Sample Power 2.0 was utilized for power calculation analysis. Differences among three groups were assessed via Analysis of Variance (ANOVA), followed by Tukey's multiple comparisons tests ($\alpha = 0.05$). Graphical representations of the statistical analyses were generated using GraphPad Prism 9 (GraphPad Software, San Diego, CA, USA). A *P*-value of < 0.05 was considered statistically significant. Results with $P < 0.01$ are even more statistically significant than those with $P < 0.05$, indicating a higher level of confidence in the comparisons.

Results

Mite Species Appearance Rate in Household

To determine the prevalence of mite species in households, we conducted a dust survey of allergic patients in Taiwan. Out of the 202 survey samples analyzed, the most common mite species found were Dp (82.7%), Df (72.8%), and *B. tropicalis* (Bt) (46.0%) (**Figure 1A**). House dust mites (Dp, Df, and Bt) were the predominant species in Taiwanese households, accounting for a higher prevalence compared to storage mites (Tp). Additionally, 24.8% (50/202) of the samples contained Ce mites.

Correlation of Environmental Conditions and Mite Disturbance

To understand the factors that cause mites to appear, surveys were conducted in a total of 202 households. The prevalence of pet keeping was significantly associated with higher frequencies of Bt ($p = 0.023$) and Ce ($p = 0.018$), while other mites showed no significant association (**Figure 1B**). The exact *P* values for the detailed statistical analysis were listed in **Supplemental Table 1A**. Among the households with pet keeping where Bt mites were present, the highest frequency was observed in those with keeping dogs, accounting for 19.8% (40/202), followed by households with keeping cats at 7.4% (15/202). Among the pet keeping where Ce mites were detected, the highest frequency was observed among those with dogs, comprising 10.9% (22/202), followed by households with cats at 5.0% (10/202), and finally households with birds at 0.5% (1/202).

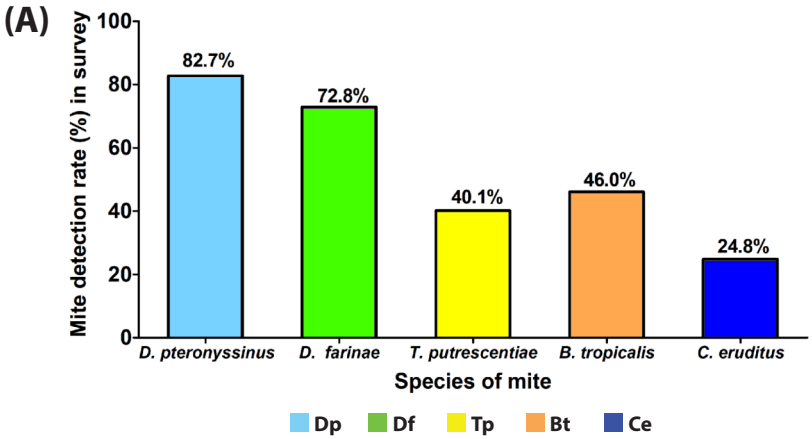


Figure 1. Prevalence of Mite Species in Household Dust Samples from Allergic Patients in Taiwan. (A) A total of 202 dust samples were collected from households of allergic patients in Taiwan. The prevalence rates of these species are shown as percentages. Three categories of mites were observed including house dust mites, storage mites, and predatory mites.

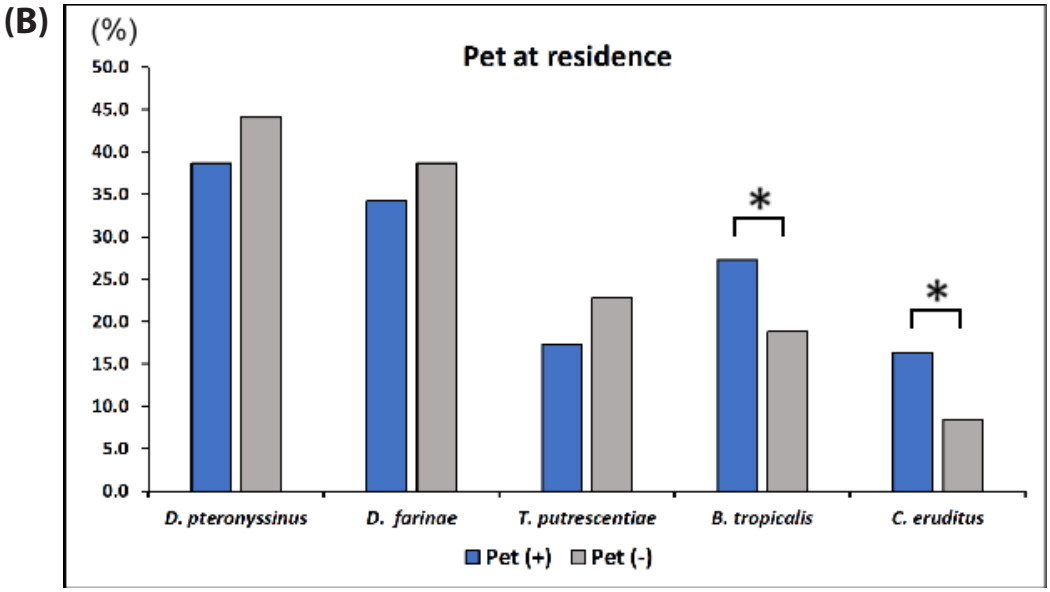
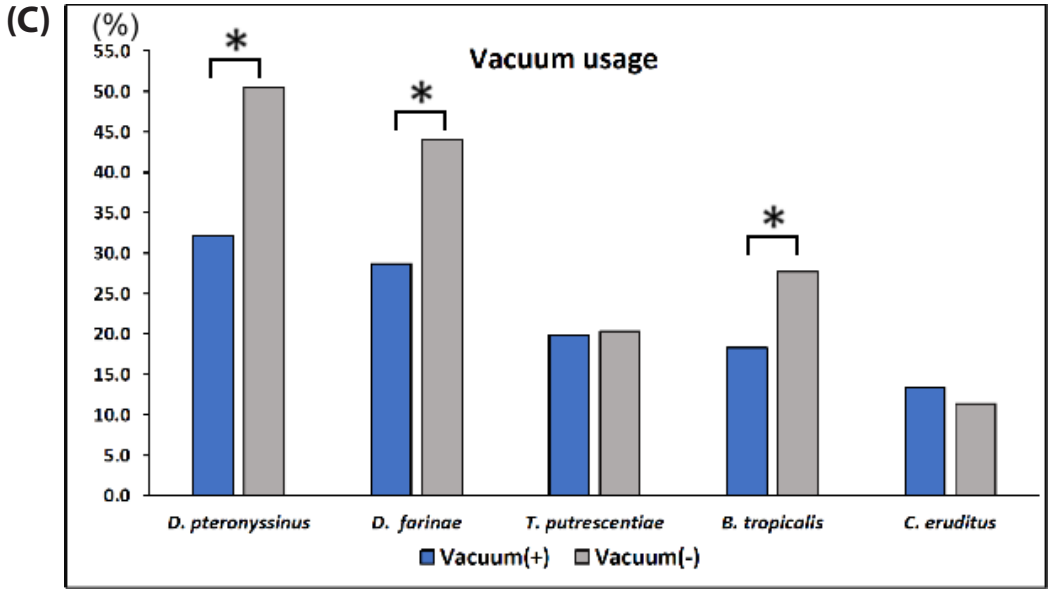
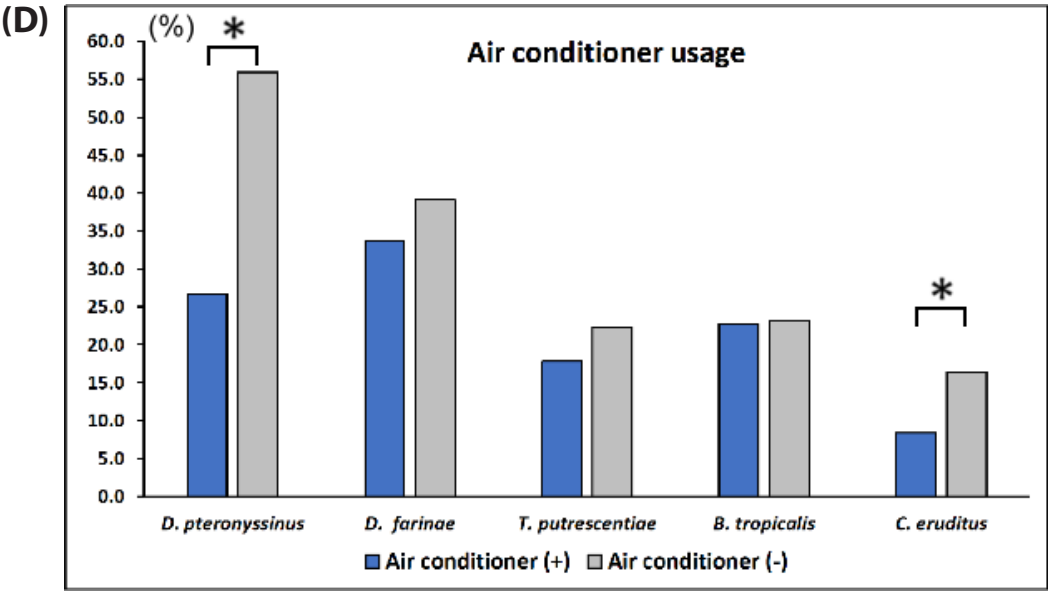


Figure 1B-1G. Correlation of Environmental Conditions and Mite Disturbance in Household Surveys. (B) Relationship between pet keeping in residences and occurrence frequencies of mite species. (C) Influence of vacuum cleaner usage in the bedroom on occurrence frequencies of mite species. (D) Impact of air conditioner usage in the bedroom on occurrence frequencies of mite species. (E) Association of bedroom location on occurrence frequencies of mite species. (F) Influence of cleaning cycle on occurrence frequencies of mite species. (G) Relationship between protein concentrations in the bed and occurrence frequencies of mite species. The p-values less than 0.05 were considered statistically significant among the group comparisons. When significance $p < 0.05$ presented as "*"; $p < 0.01$ presented as "**".





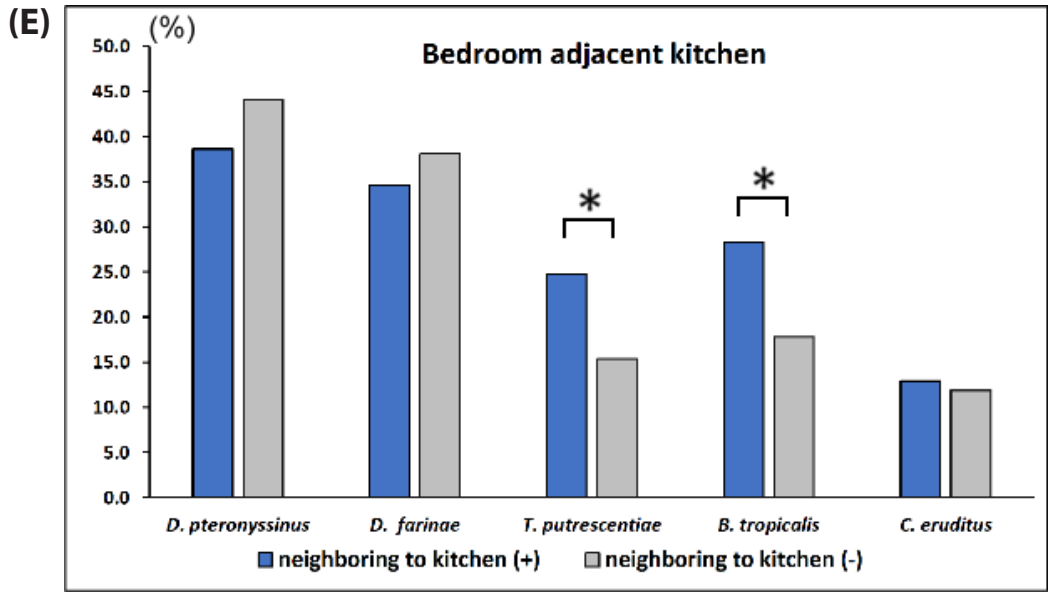


Figure 1. (Continued)

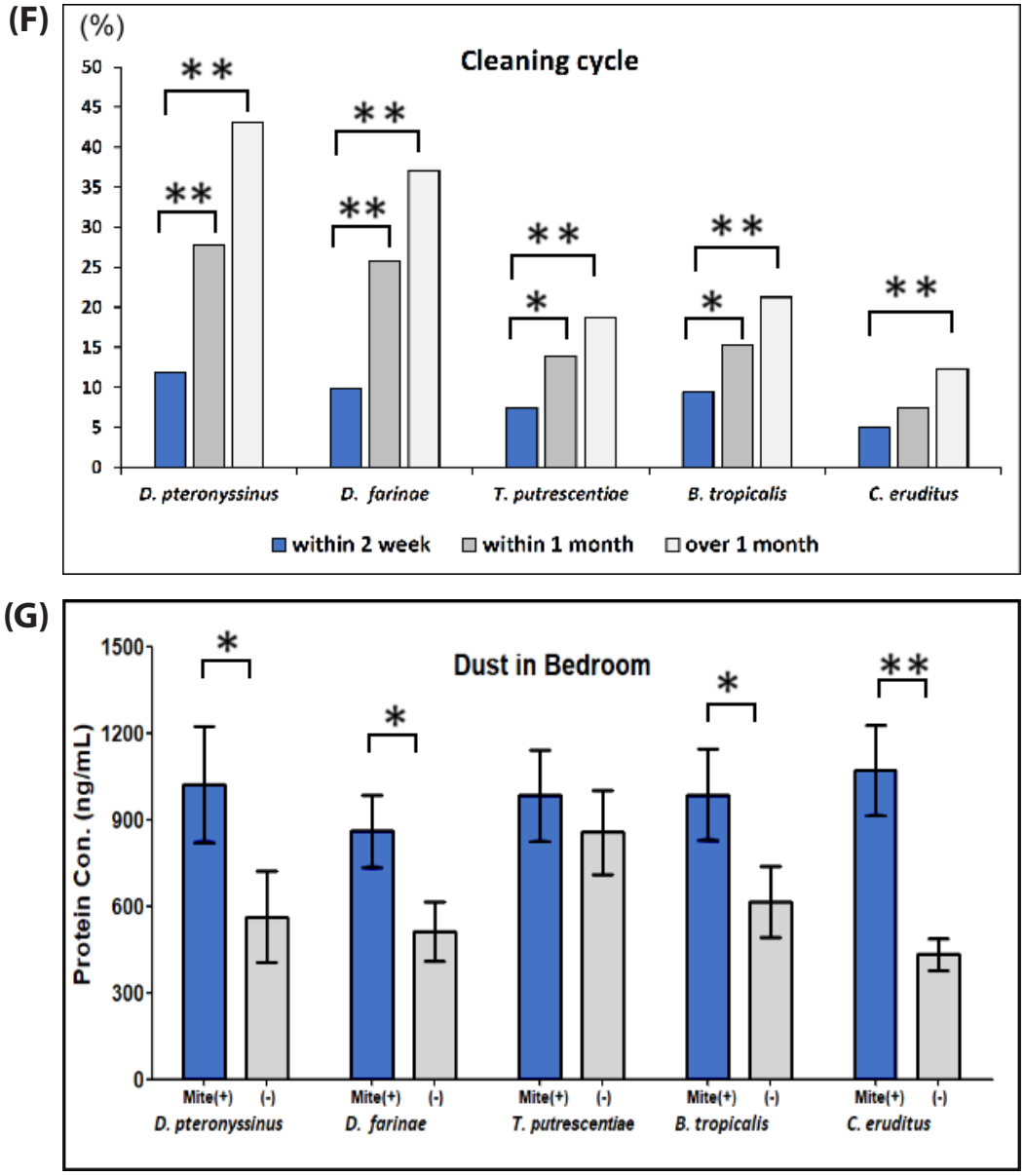


Figure 1. (Continued)

Vacuum cleaner usage in the bedroom was linked to lower frequencies of Dp (p = 0.022), Df (p = 0.031), and Bt (p = 0.042) (**Figure 1C**). The exact P values were listed in **Supplemental Table 1B**. The usage of air conditioners in the bedroom was associated with lower frequencies of Dp (p = 0.032) and Ce (p = 0.022) (**Figure 1D**), the exact P values listed in **Supplemental Table 1C**. Mites appeared more frequently in bedrooms adjacent to the kitchen, particularly house dust mite-Bt (p = 0.023) and storage mite-Tp (p = 0.029) (**Figure 1E**) (**Supplemental Table 1D**). Cleaning frequency also had an impact, with significant differences observed between cleaning cycles of within 2 weeks and over 1 month (the exact P values listed in **Supplemental Table 1E-a**). House dust mites (Dp, Df, and Bt) and storage mites (Tp) were less prevalent with a cleaning cycle of two weeks compared to one month (**Figure 1F**) (the exact P values listed in **Supplemental Table 1E-b**). The presence of mites correlated with higher protein concentration on the bed for species Dp, Df, Bt, and Ce (**Figure 1G**), (the exact P values listed in **Supplemental Table 1F**). The protein concentration in the floor dust showed minimal variation regardless of the presence or absence of mites (data not shown).

Correlation of Mite Appearance and Allergic Symptoms

To explore the potential link between mite presence and allergic symptoms, correlation analyses were conducted. Among the 202 survey samples, a higher percentage of samples with mite appearance exhibited allergic symptoms compared to those without allergic symptoms for Dp (55.4% vs. 27.2%) (p = 0.017), Df (p = 0.027), Bt (p = 0.001), and Ce (p = 0.001) (**Figure 2A**) (**Supplemental Table 2A**).

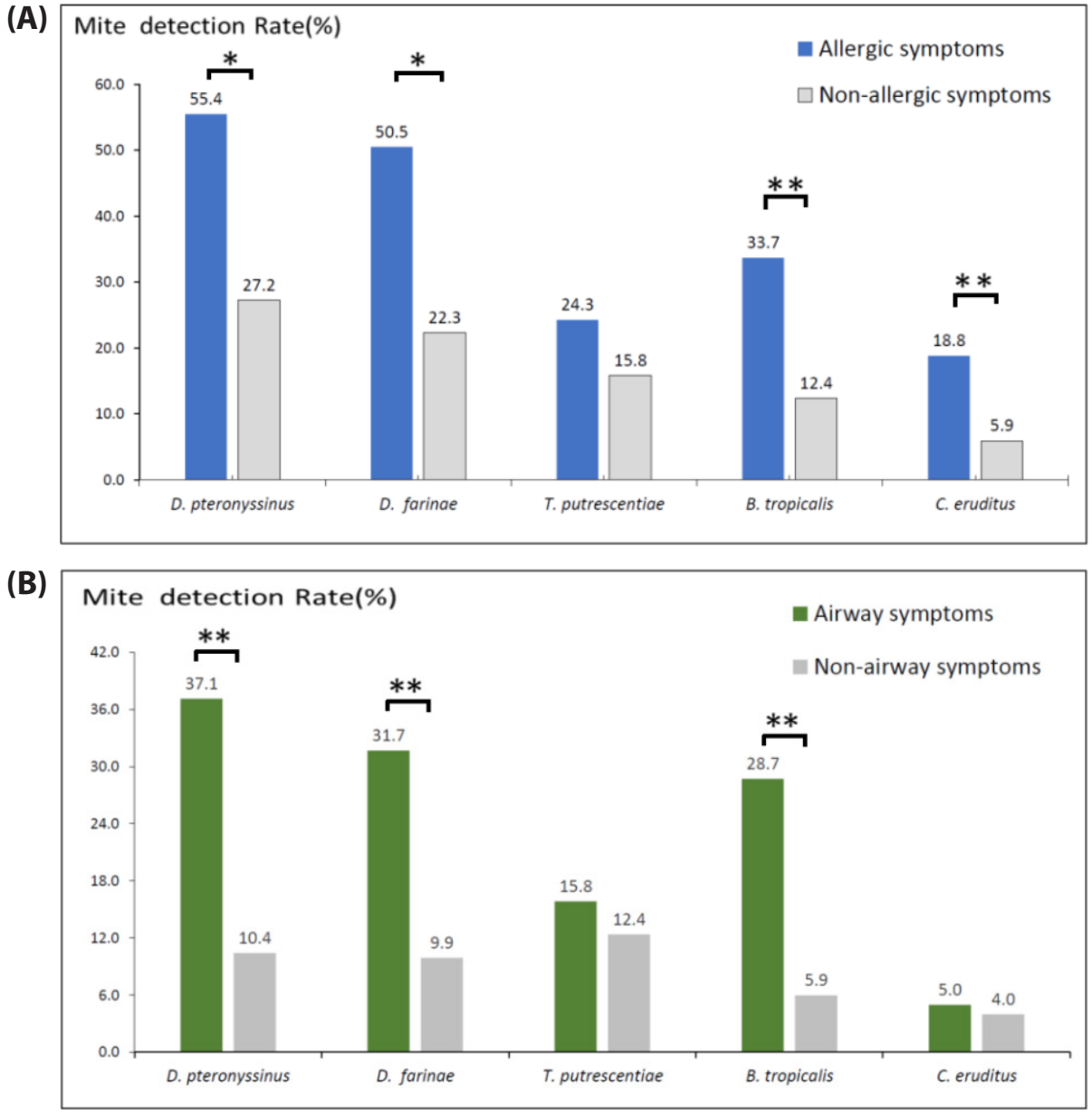


Figure 2. Correlation between mite appearance and allergic symptoms. (A)The detection rate of five mite species in the group with allergic or non-allergic symptoms. (B) Association between mite appearance and airway symptoms. (C) Association between mite appearance and nasal symptoms. (D) Association between mite appearance and skin symptoms. The p-values less than 0.05 were considered statistically significant among the group comparisons. When significance $p < 0.05$ presented as "*"; $p < 0.01$ presented as "**".

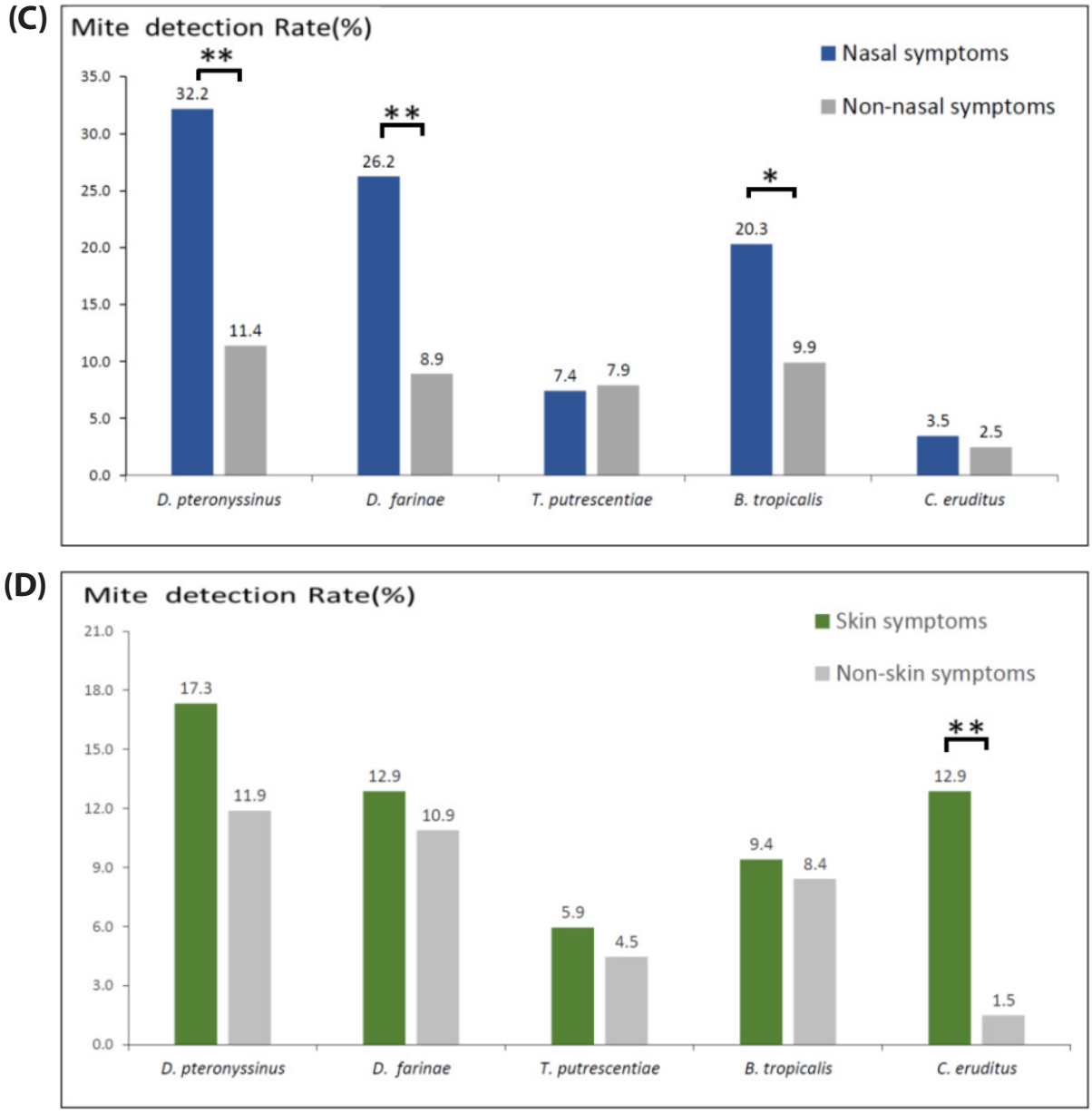


Figure 2. (Continued)

This indicates a significant correlation between mite presence in the household environment and the provocation of allergic symptoms. Further analysis revealed that the appearance of Dp, Df, and Bt significantly increased the occurrence of airway symptoms, while Dp (p = 0.001), Df (p = 0.002), and Bt (p = 0.007) were significantly associated with nasal symptoms for Dp (p = 0.007), Df (p = 0.008), and Bt (p = 0.019) (**Figure 2B and 2C**) (**Supplemental Table 2B and 2C**). The presence of Ce showed a significantly higher percentage of skin symptoms (p = 0.001) (**Figure 2D**) (**Supplemental Table 2D**), indicating a strong correlation between Ce appearance and skin symptoms.

Evaluation of Basophil Activation Triggered by Mites through Histamine and Calcium Release

The effects of mite allergens on basophil activation were studied using KU812 cells stained with Dao^+, anti-CD63 and anti-CD203C antibodies, analyzed by flow cytometry. Upon mite stimulation, the percentage of double-positive cells ($Dao^+/CD63^+$) significantly increased in Ce (25.24%) and Dp (29.27%) compared to saline (**Figure 3A**). Additionally, the levels of intracellular calcium ions (Ca^{2+}) markedly rose up to 10^4 at 100 seconds after mite allergen challenge, while minimal changes were observed in the saline (**Figure 3B**). Flow cytometry analysis revealed a significant increase in basophils expressing both CD63 and CD203C markers (82.88% for Ce and 73.40% for Dp) compared to the saline group (11.68%) (**Figure 3C**). It indicates the activation of basophils upon exposure to Ce and Dp mites.

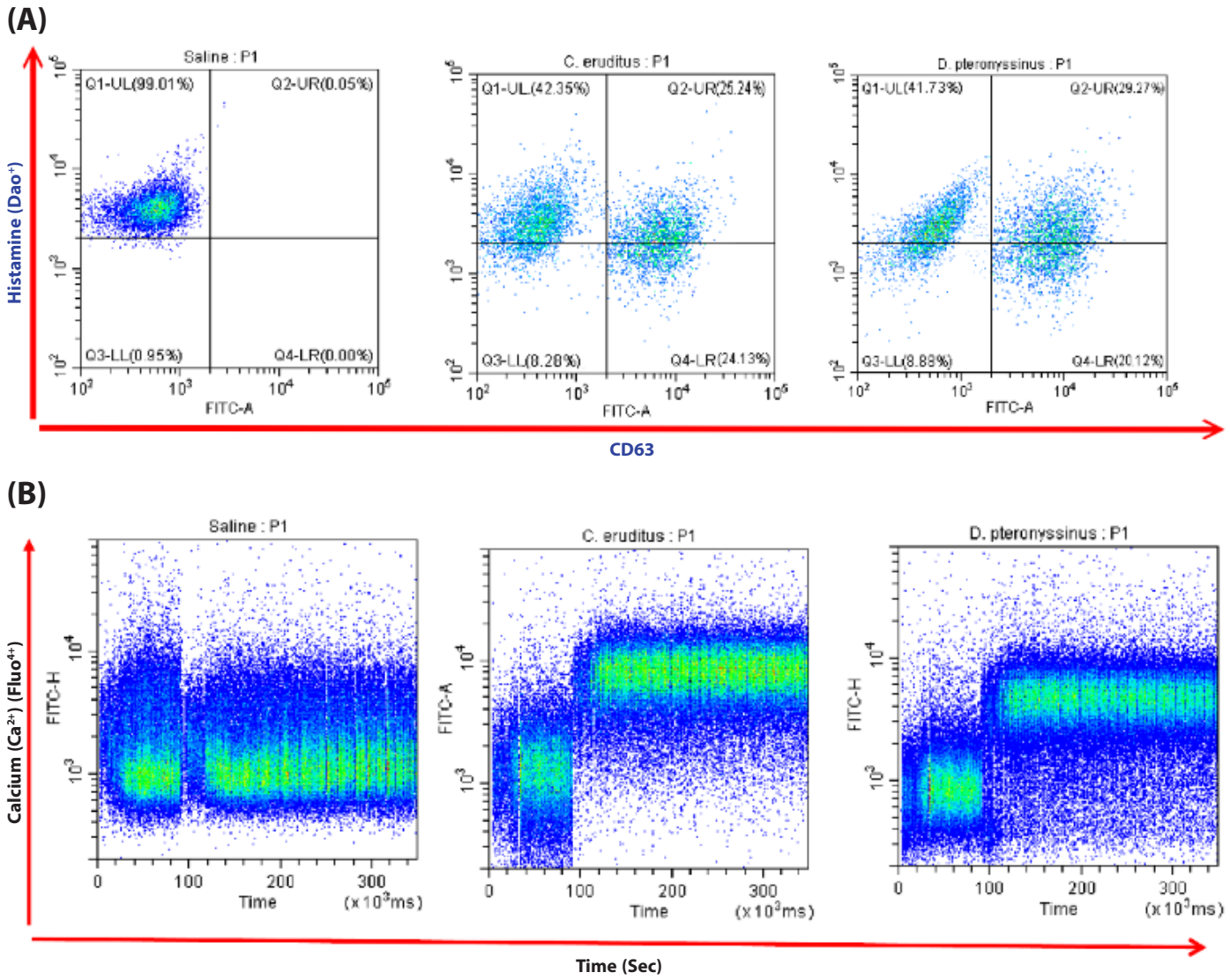


Figure 3. Evaluation of basophil activation triggered by mites through histamine, calcium release, and surface marker expression. (A) Flow cytometry analysis of basophil activation using Dao⁺ (histamine) and anti-CD63 antibody staining in basophil cell line KU812. Two-dimensional scatter plots showing the percentage of double-positive cells in response to saline, the mites of Ce and Dp stimulation. (B) Monitoring of intracellular calcium ion concentrations using fluo-4 staining and FITC-labeled flow cytometry in basophil cells. The fluorescence intensity, reflecting changes in intracellular calcium ion levels, was measured over time. (C) Flow cytometry analysis of CD63 and CD203C expression on the surface of basophil cells after exposure to mite allergens. Two-dimensional scatter plots showing the percentage of basophils expressing both CD63 and CD203C markers. (D) Prevalence of IgE sensitization to house dust mite, storage mite, and predatory mite. The prevalence of IgE sensitization to various mite species in Taiwanese households was investigated among 120 participants with allergic symptoms. Mite-specific IgE > 0.35 IU/ml was considered positive. The prevalence of IgE sensitization to mites is presented as a percentage.

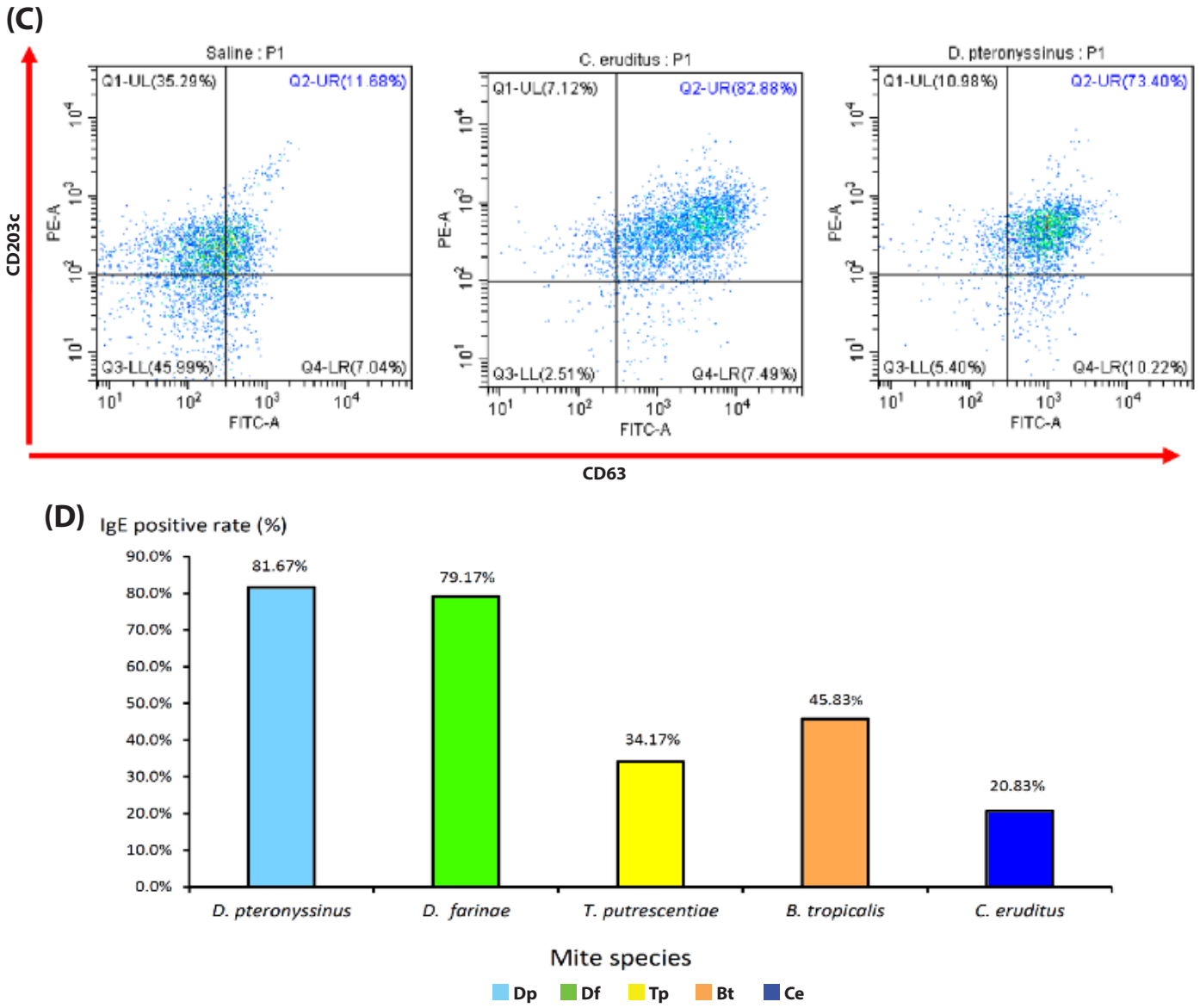


Figure 3. (Continued)

Prevalence of IgE Sensitization to Mite Species in Taiwan

To determine the mite allergens responsible for allergy, we investigated IgE sensitization to mite species in Taiwanese households. A total of 120 participants with allergic symptoms were recruited. Results revealed high prevalence of IgE sensitization to house dust mites Dp (81.67%), followed by Df (78.17%), and Bt (45.83%) and storage mites Tp (34.17%) (***Figure 3D***). Although Ce was less common, 20.83% of allergy patients exhibited positive IgE reactions to this mite. House dust mites and storage mites were identified as dominant allergens in Taiwanese households. Further investigation into the prevalence and distribution of Ce is warranted.

Correlation between Mite-Specific IgE and Total IgE in Allergic Patients

The correlation between mite-specific IgE and total IgE in 120 allergic patients with symptoms was examined. Results showed a significant positive correlation between Ce-specific IgE and Total IgE (R^2 = 0.723, p = 0.001) (**Figure 4A**) (**Supplemental Table 4A**). Similar findings were observed for Dp-specific IgE and total IgE (R^2 = 0.753, p = 0.001) (**Figure 4B**) (**Supplemental Table 4B**). Monitoring total IgE levels may help assess the severity of allergic symptoms in patients with positive IgE response to mites. Total IgE levels were slightly higher in patients with positive IgE to Ce but not significantly different (**Figure 4C**) (**Supplemental Table 4C**). However, patients with positive IgE to Dp (> 0.35 KU/L) had significantly higher total IgE levels (p = 0.0003) (**Figure 4D**) (**Supplemental Table 4D**). Further analysis revealed a significant increase in Ce-specific IgE (p = 0.0001) and Dp-specific IgE (p = 0.0001) in the group with total IgE levels > 160 IU/ml (**Figure 4E and 4F**) (**Supplemental Table 4E and 4F**). It suggests a strong correlation between total IgE levels and mite-specific IgE, which may provide important diagnostic strategies for clinical diagnosis.

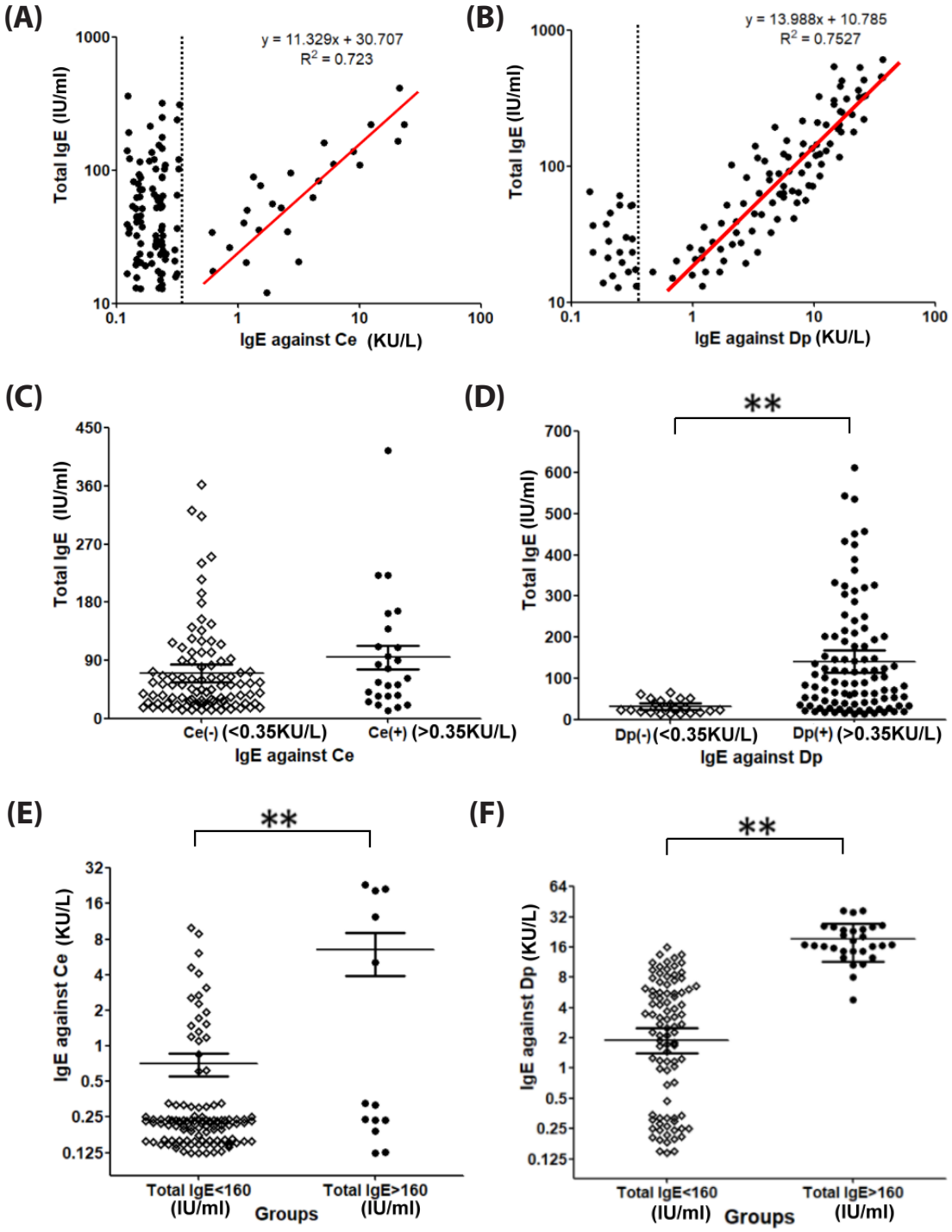


Figure 4. Correlation between mite-specific IgE and total IgE in allergic patients. A total of 120 allergic patients were recruited for analysis. (A) Scatter plot showing the significant positive correlation between Ce-specific IgE (IgE > 0.35) and total IgE in allergic patients (R^2 = 0.723, p < 0.05). (B) Scatter plot demonstrating a significant positive correlation between Dp-specific IgE (IgE > 0.35) and total IgE in allergic patients (R^2 = 0.753, p < 0.05). (C) Comparison of total IgE levels between allergic patients with positive and negative IgE to Ce. (D) Comparison of total IgE levels between allergic patients with positive and negative IgE to Dp. (E) Comparison of Ce-specific IgE between allergic patients with total IgE levels > 160 IU/ml and those with ≤ 160 IU/ml. (F) Comparison of Dp-specific IgE between allergic patients with total IgE levels > 160 IU/ml and those with ≤ 160 IU/ml. The p-values less than 0.05 were considered statistically significant among the group comparisons. When significance p < 0.01 presented as "**".

Evaluation of Cross-Reactivity Among Dust Mites and Predatory Mites

The cross-reactivity of allergens between Ce and house dust mite in allergic patients was assessed using allergen-specific IgE adsorption. Among 120 participants, 97 allergic patients had positive responses to Dp (> 0.35 KU/L). Adsorption with Df significantly decreased Dp IgE levels (p = 0.003), whereas adsorption with Ce did not show a significant decrease (**Figure 5A**) (**Supplemental Table 5A**). Of the 120 participants, 25 had positive IgE reactions to Ce. Adsorption with Dp or Df did not significantly reduce Ce IgE levels (**Figure 5B**) (the exact P values listed in **Supplemental Table 5B**). Our findings indicate high cross-reactivity between Dp and Df, while cross-reactivity between Dp and Ce is limited. Dp and Ce have distinct allergens, highlighting their differences in allergenicity.

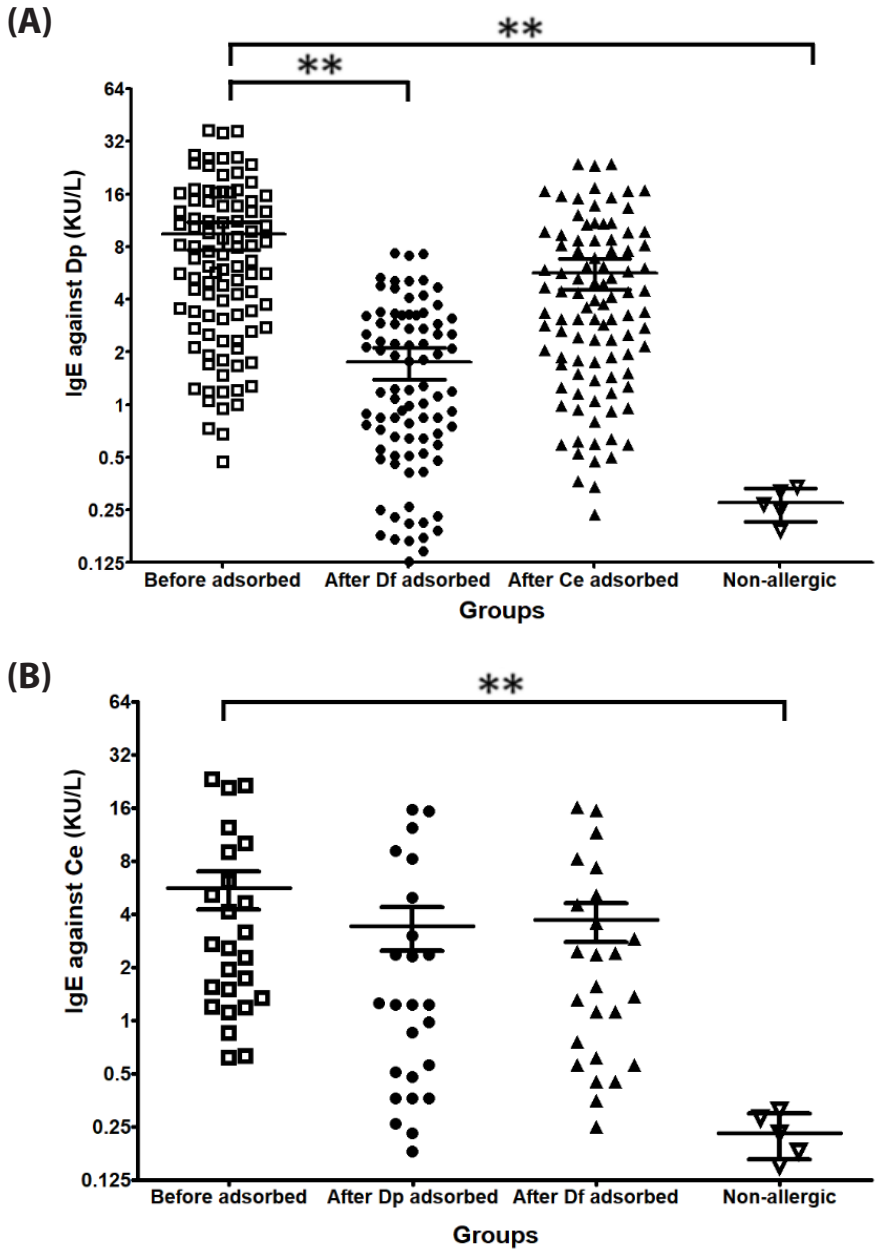


Figure 5. Evaluation of cross-reactivity among dust mites and predatory mites. (A) Comparison of Dp-specific IgE levels before and after adsorption in allergic patients. Serum adsorption was performed with Df or Ce. A total of 97 allergic patients with positive IgE against Dp with were recruited for analysis. (B) Comparison of Ce-specific IgE levels before and after adsorption in allergic patients. Serum adsorption was performed with Dp or Df. A total of 25 allergic patients with positive IgE against Ce with were recruited. The p-values less than 0.05 were considered statistically significant among the group comparisons. When significance $p < 0.01$ presented as "**".

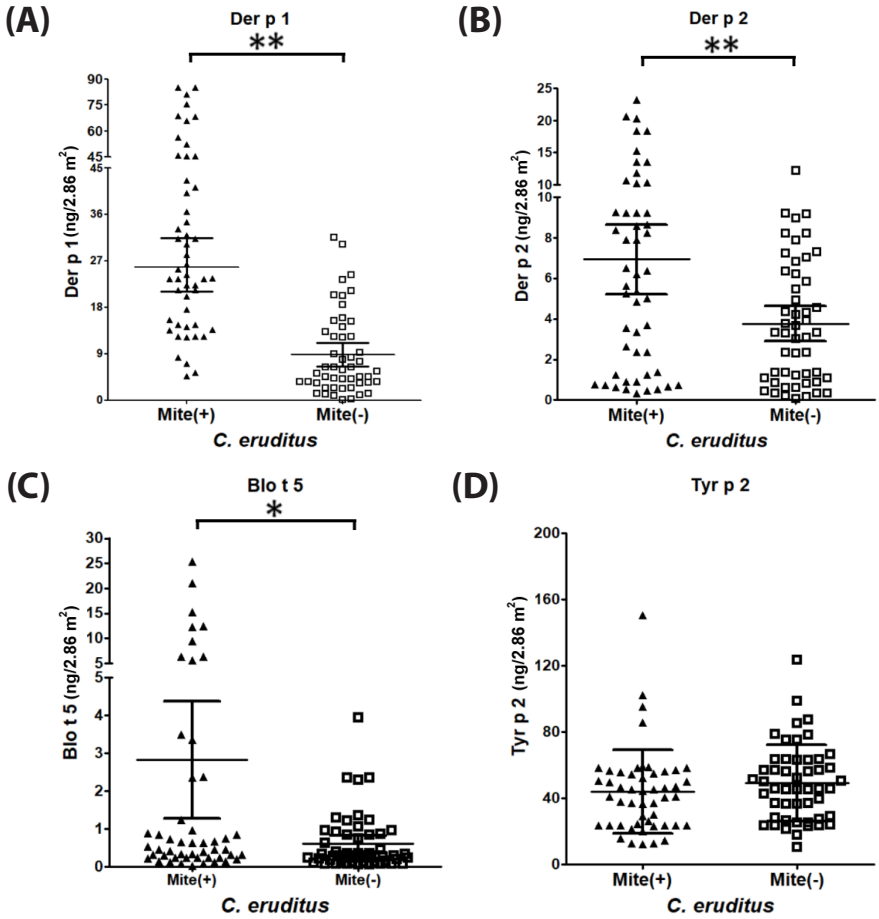


Figure 6. Association between the presence of predatory mites Ce and residual allergens in household beddings. The levels of major allergens, including: (A) Der p 1, (B) Der p 2, (C) Blo t 5, and (D) Tyr p 2, were compared between households with the presence of Ce and households without Ce in bedding dust samples. Allergen concentration presented as ng/ml. The p-values less than 0.05 were considered statistically significant among the group comparisons. When significance $p < 0.05$ presented as "*"; $p < 0.01$ presented as "**".

Association Between Predatory Mites Ce and Residual Allergens on Household Beddings

The presence of predatory mites Ce in household environments and its association with residual allergens in beddings was investigated. A total of 100 bedding dust samples were collected from 50 households with Ce and 50 households without Ce. The allergen levels were presented as ng/2.86 m^2, representing the allergen content per unit area of a double mattress size of 1.52 m × 1.88 m. ELISA analysis detected significantly higher levels of Der p 1 ($p = 0.0001$), Der p 2 ($p = 0.0013$), and Blo t 5 ($p = 0.0460$) allergens in beddings of households with Ce compared to those without Ce (**Figure 6**). However, there was no significant difference in the level of Tyr p 2, the Tp major allergen, between the two groups (**Figure 6**) (the exact *P* values listed in **Supplemental Table 6**). These findings suggest that high levels of residual allergens in beddings may be associated with the presence of predatory mites Ce.

Discussion

Predatory mites-Ce exhibits predatory behavior towards other arthropods and can cause injuries in animals and occasionally induce pruritic rashes in humans.[25] House dust mites Df and Dp are widespread in human habitations and commonly cause allergic reactions. We investigated mite species in Taiwanese households by conducting a dust survey among allergic patients. House dust mites, especially Dp and Df were prevalent in the samples. Additionally, 24.8% of the samples contained Ce, which can contribute to allergic symptoms. The high prevalence of house dust mites underscores their role as common allergenic sources, while the presence of Ce adds to the overall mite burden and potential allergic reactions.

The correlation between environmental conditions and mite presence in households was investigated to identify several factors that contribute to mite appearance and frequency of mite species. Pet ownership was associated with higher frequencies of Bt and Ce mites, highlighting pets as potential carriers or disseminators of mites. Pet owners should take measures for pet hygiene and control to reduce the risk of mite-related allergies. Vacuuming the bedroom was associated with lower frequencies of Dp, Df, and Bt,

as it effectively removes allergens. Regular vacuuming in mite-prone areas can minimize allergen exposure and alleviate symptoms. The usage of an air conditioner in the bedroom was linked to lower frequencies of Dp and Ce, potentially due to its impact on humidity levels. Air conditioners can create an unfavorable environment for mite growth and survival. Maintaining optimal humidity conditions is crucial for mite control in households.

Higher frequencies of mites in bedrooms adjacent to the kitchen were found, particularly storage mites of Tp, indicating that food storage areas may influence mite presence. Proper storage and cleanliness in the kitchen are crucial for reducing storage mite populations and preventing allergies. The frequency of cleaning cycles also affected mite prevalence. Shorter cleaning cycles were associated with lower frequencies of house dust mites and storage mites. Regular and frequent cleaning helps remove dust and allergens, creating an unfavorable environment for mite survival. Furthermore, our study revealed that the protein content in beds was significantly higher when mites were present, suggesting that mites contribute to higher levels of allergenic proteins, triggering allergic reactions in susceptible individuals.

We explored the potential correlation between mite presence and allergic symptoms. Significant associations were found between mites and the occurrence of allergic symptoms. Samples with mite appearance had a higher incidence of allergic symptoms compared to those without. Mite species showed distinct associations with specific symptoms: Dp, Df, and Bt were associated with airway and nasal symptoms, while Ce was linked to skin symptoms. It suggests that mites in the household environment can trigger allergic reactions, with different species contributing to different symptom types.

Basophil activation measurement is crucial for understanding allergic reactions to allergens. The use of intracellularly labeled DAO, along with surface expression analysis of CD203c and CD63 on basophils, provides a simple method to assess basophil histamine release and monitor immunotherapy.[26] In this study, basophil activation triggered by mite allergens was evaluated using histamine and calcium release as markers. Results showed a significant increase in basophil activation, evidenced by increased histamine release and CD63 expression, upon stimulation with Ce and Dp mite allergens. The study revealed an increase in basophils expressing both CD63 and CD203C after exposure to Ce and Dp mites, suggesting mite exposure activates basophils.[27] Monitoring basophil expression levels of CD203c may be a useful tool in monitoring asthma in patients.[28] Results revealed a substantial increase in the percentage of basophils expressing both CD63 and CD203C after exposure to Ce and Dp mites. This finding suggests that mite exposure induces the activation of basophils, as indicated by the upregulation of both CD63 and CD203C.

Results showed high prevalence of house dust mites Dp, Df, and Bt among patients with allergies, emphasizing their significance as key allergens. These findings align with global studies on house dust mite sensitization in individuals with allergies.[29] House dust mites are recognized as the primary source of indoor allergens, contributing to allergic rhinitis and allergic asthma globally.[29] Our study also identified the involvement of house dust mites and storage mites in allergic reactions among Taiwanese households, specifically sensitization to Bt and Tp of patients, highlighting their significance as relevant allergens in allergic population. Previous research has shown that Bt and Tp play a role in sensitization, allergenicity, IgE-related immunologic responses, and allergic diseases.[30,31] Although Ce is not commonly found in general household environments, 20.83% of allergy patients showed positive IgE reactions to this mite. This indicates that Ce may have an underestimated role as a potential allergen in households. Ce should be recognized as a relevant airborne allergen, not only in occupational respiratory medicine but also in persistent nonoccupational allergic rhinitis, particularly among populations living in humid tropical and/or subtropical climates.[23]

Results revealed high cross-reactivity between house dust mite allergens Dp and Df, indicating shared allergenic components. It aligns with previous studies highlighting the close relationship between these two dust mite species.[16] However, the adsorption of predatory mite Ce did not significantly decrease Dp IgE levels, suggesting low cross-reactivity between their allergens. These results emphasize the distinct allergenicity of predatory mites compared to house dust mites. Similar findings have been observed with the predatory mite *Amblyseius cucumeris*.[32] Results indicate species-specific allergens and common allergens that cross-react with Dp, it provides insights into the molecular basis underlying mite-induced allergic responses.

We found that households with Ce had higher levels of mite major allergens in their beddings compared to households without Ce. It suggests a potential association between Ce presence and increased levels of common household allergens, specifically Der p1, Der p2, and Blo t5. Der p1 and Der p2 are major allergens from the fecal pellets and bodies of the house dust mite.[33] Blo t5, is a major allergen from the house dust mite Bt.[30] The Ce presence is significantly associated with higher levels of allergens in households, indicating that increased allergen accumulation in bedding could contribute to the presence of these predatory mites. Several potential reasons for this association, Ce is known to feed on other mites. As the populations of these mites increase, the presence of Ce may also increase due to the availability of food sources. It leads to higher levels of these major allergens in bedding. Secondly, the consumption of dust mites by Ce may result in increased shedding of allergenic particles in the form of fecal pellets and body fragments, which can raise the levels of these major allergens in the environment.

This study provides insights into mite species prevalence in Taiwanese households, with the most common species being Dp, Df, and Bt. Environmental factors such as pet keeping, vacuum cleaner usage, air conditioner usage, proximity to the kitchen, cleaning frequency, and protein concentration in beds influence mite prevalence. Understanding these correlations can guide preventive measures to reduce mite-related allergies and improve indoor air quality. The correlation between mite appearance and allergic symptoms, including airway, nasal, and skin symptoms, is significant. Ce presence is associated with a higher percentage of skin symptoms. Controlling mite populations through cleaning, dust mite covers, and humidity maintenance can alleviate allergic symptoms. Basophil activation triggered by Ce is similar to Dp, with significant histamine and calcium release and upregulation of CD63 and CD203C expression. Specific IgE against Ce or Dp positively correlates with total IgE levels in mite-related allergies. The presence of Ce is associated with elevated levels of major mite allergens in beddings, emphasizing the role of predatory mites and complex interactions with other mite species.

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Declaration of competing interest

No conflicts of interest, financial or otherwise, are declared by the authors.

Author's contributions

- Chun-Hsien Hsu was responsible for the planning of the experimental architecture and the execution of the basophil activation experiment.
- Ding-Kuo Chien collected and analyzed the experimental data.
- Jaw-Ji Tsai was responsible for recruiting allergic patients in the clinics and assisted with the IgE adsorption assay.
- Chung-Yang Yen was responsible for recruiting allergic patients and identifying the allergic symptoms.
- En-Chih Liao designed experiments, supervised the experiment and project progress, and wrote and revised the manuscript.

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Supplementary Tables

Table 1A. The association between pet keeping and mite appearance.

	Pet (+)	Pet (-)	*P* Value
D. pteronyssinus	78 (38.6%)	89 (44.1%)	0.678
D. farinae	69 (34.2%)	78 (38.6%)	0.336
T. putrescentiae	35 (17.3%)	46 (22.8%)	0.667
B. tropicalis	55 (27.2%)	38 (18.8%)	**0.023**
C. eruditus	33 (16.3%)	17 (8.4%)	**0.018**

Total n = 202

Table 1B. The association between vacuum usage and mite appearance.

	Vacuum (+)	Vacuum (-)	*P* Value
D. pteronyssinus	65 (32.2%)	102 (50.5%)	**0.022**
D. farinae	58 (28.7%)	89 (44.1%)	**0.031**
T. putrescentiae	40 (19.8%)	41 (20.3%)	0.886
B. tropicalis	37 (18.3%)	56 (27.7%)	**0.042**
C. eruditus	27 (13.4%)	23 (11.4%)	0.514

Total n = 202

Table 1C. The association between air conditioner usage and mite appearance.

	Air conditioner (+)	Air conditioner (-)	*P* Value
D. pteronyssinus	54 (26.7%)	113 (55.9%)	**0.032**
D. farinae	68 (33.7%)	79 (39.1%)	0.294
T. putrescentiae	36 (17.8%)	45 (22.3%)	0.196
B. tropicalis	46 (22.8%)	47 (23.3%)	0.786
C. eruditus	17 (8.4%)	33 (16.3%)	**0.022**

Total n = 202

Table 1D. The association between bedroom adjacent kitchen and mite appearance.

	adjacent kitchen (+)	adjacent kitchen (-)	*P* Value
D. pteronyssinus	78 (38.6%)	89 (44.1%)	0.678
D. farinae	70 (34.7%)	77 (38.1%)	0.784
T. putrescentiae	50 (24.8%)	31 (15.3%)	**0.023**
B. tropicalis	57 (28.2%)	36 (17.8%)	**0.029**
C. eruditus	26 (12.9%)	24 (11.9%)	0.806

Total n = 202

Table 1E-a. The association between cleaning cycle and mite appearance.

	within 2 weeks	within 1 month	*P* Value
D. pteronyssinus	24 (11.9%)	56 (27.7%)	**0.004**
D. farinae	20 (9.9%)	52 (25.7%)	**0.002**
T. putrescentiae	15 (7.4%)	28 (13.9%)	**0.026**
B. tropicalis	19 (9.4%)	31 (15.3%)	**0.023**
C. eruditus	10 (5.0%)	15 (7.4%)	0.163

Total n = 202

Table 1E-b. The association between cleaning cycle and mite appearance.

	within 2 weeks	within 1 month	*P* Value
D. pteronyssinus	24 (11.9%)	87 (43.1%)	**0.0001**
D. farinae	20 (9.9%)	75 (37.1%)	**0.0001**
T. putrescentiae	15 (7.4%)	38 (18.8%)	**0.0002**
B. tropicalis	19 (9.4%)	43 (21.3%)	**0.0001**
C. eruditus	10 (5.0%)	25 (12.4%)	**0.0002**

Total n = 202

Table 1F. The association between protein concentration in the bed and mite appearance.

	Mite appearance (+)	Mite appearance (-)	*P* Value
D. pteronyssinus	1023.52#	563.51	**0.017**
D. farinae	862.51	512.36	**0.022**
T. putrescentiae	985.85	856.89	0.416
B. tropicalis	987.63	615.63	**0.026**
C. eruditus	1071.36	433.36	**0.003**

#Mean of protein concentration: ng/mL

Table 2A. The association between allergic symptoms and mite appearance.

	Allergic symptoms (+)	Allergic symptoms (-)	*P* Value
D. pteronyssinus	112 (55.4%)	55 (27.2%)	**0.017**
D. farinae	102 (50.5%)	45 (22.3%)	**0.027**
T. putrescentiae	49 (24.3%)	32 (15.8%)	0.159
B. tropicalis	68 (33.7%)	25 (12.4%)	**0.001**
C. eruditus	38 (18.8%)	12 (5.9%)	**0.001**

Total n = 202

Table 2B. The association between airway symptoms and mite appearance.

	Airway symptoms (+)	Airway symptoms (-)	*P* Value
D. pteronyssinus	75 (37.1%)	21 (10.4%)	**0.001**
D. farinae	64 (31.7%)	20 (9.9%)	**0.002**
T. putrescentiae	32 (15.8%)	25 (12.4%)	0.414
B. tropicalis	58 (28.7%)	12 (5.9%)	**0.007**
C. eruditus	10 (5.0%)	8 (4.0%)	0.511

Total n = 202

Table 2C. The association between nasal symptoms and mite appearance.

	Nasal symptoms (+)	Nasal symptoms (-)	*P* Value
D. pteronyssinus	75 (37.1%)	21 (10.4%)	**0.007**
D. farinae	64 (31.7%)	20 (9.9%)	**0.008**
T. putrescentiae	32 (15.8%)	25 (12.4%)	0.841
B. tropicalis	58 (28.7%)	12 (5.9%)	**0.019**
C. eruditus	10 (5.0%)	8 (4.0%)	0.463

Total n = 202

Table 2D. The association between skin symptoms and mite appearance.

	Skin symptoms (+)	Skin symptoms (-)	*P* Value
D. pteronyssinus	35 (17.3%)	24 (11.9%)	0.238
D. farinae	26 (12.9%)	22 (10.9%)	0.329
T. putrescentiae	12 (5.9%)	9 (4.5%)	0.569
B. tropicalis	19 (9.4%)	17 (8.4%)	0.888
C. eruditus	26 (12.9%)	3 (1.5%)	**0.001**

Total n = 202

Table 4A and 4B. The correlation between mite-specific IgE and total IgE.

X axis	Y axis	Correlation R^2	*P* Value
IgE to *C. eruditus*#	Total IgE	0.723	**0.0001**
IgE to *D. pteronyssinus*#	Total IgE	0.753	**0.0001**

#Mite specific IgE > 0.35 recruited for analysis.

Table 4C. The correlation between positive Ce IgE and total IgE.

Group	Ce (-) (< 0.35 KU/L)	Ce (+) (> 0.35 KU/L)	*P* Value
Total IgE	69.95 ± 7.18# (n = 95)	94.36 ± 18.09 (n = 25)	0.1480

#mean ± SEM of mite specific IgE

Table 4D. The correlation between positive Dp IgE and total IgE.

Group	Dp (-) (< 0.35 KU/L)	Dp (+) (> 0.35 KU/L)	*P* Value
Total IgE	31.40 ± 3.51[#] (n = 22)	140.9 ± 13.76 (n = 98)	**0.0003**

[#]mean ± SEM of mite specific IgE

Table 4E and 4F. The association between Ce-specific IgE and total IgE.

	Total IgE < 160	Total IgE > 160	*P* Value
IgE to *C. eruditus*	0.71 ± 0.15 (n = 107)[#]	6.48 ± 2.59 (n = 13)	**0.0001**
IgE to *D. pteronyssinus*	3.91 ± 0.41 (n = 91)	19.36 ± 1.49 (n = 29)	**0.0001**

[#]mean ± SEM of mite specific IgE

Table 5A. Evaluation of cross-reactivity among dust mites and predatory mites.

		IgE to Dp	Comparison	*P* Value
Group A	Before adsorbed	9.40 ± 0.86[#]		
Group B	After Df adsorbed	1.75 ± 0.18	Group A vs. B	**0.003**
Group C	After Ce adsorbed	5.68 ± 0.56	Group A vs. C	0.604
Group D	Non-allergic	0.27 ± 0.03	Group A vs. D	**0.001**

Group A-C: n = 97 with positive IgE to Dp; Group D: n = 5
[#]mean ± SEM of mite specific IgE

Table 5B. Evaluation of cross-reactivity among predatory mites and dust mites.

		IgE to Ce	Comparison	*P* Value
Group A	Before adsorbed	5.62 ± 1.36[#]		
Group B	After Dp adsorbed	3.43 ± 0.96	Group A vs. B	0.194
Group C	After Df adsorbed	3.70 ± 0.92	Group A vs. C	0.248
Group D	Non-allergic	0.23 ± 0.03	Group A vs. D	**0.001**

Group A-C: n = 25 with positive IgE to Ce; Group D: n = 5
[#]mean ± SEM of mite specific IgE

Table 6. Association between the presence of predatory mites Ce and residual allergens.

	Ce (+)	Ce (-)	*P* Value
Der p 1	32.26 ± 3.06	8.82 ± 1.13	**0.0001**
Der p 2	6.94 ± 0.85	3.76 ± 0.44	**0.0013**
Blo t 5	2.83 ± 0.78	0.62 ± 0.11	**0.0460**
Tyr p 2	43.77 ± 3.57	49.21 ±3.25	0.2624

[#]mean ± SEM of allergen content ng/2.86 m^2 of unit area
Ce (+): n = 50; Ce (-): n = 50