

Microgeographic factors and patterns of aeroallergen sensitisation

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The known Socio-economic and geographic factors influence allergic sensitisation.

The new Coastal proximity, climate, and environmental (urban v regional) factors affect allergy patterns in the Greater Sydney area. A selected ten-aeroallergen skin prick test (SPT) panel identified 98.5% of atopic patients in our sample. 72.4% of grass-sensitised patients were co-sensitised to both temperate and subtropical grasses.

The implications The identified patterns of allergic sensitisation can inform more effective aeroallergen avoidance strategies. The high level of co-sensitisation to temperate and subtropical grasses suggests that existing immunotherapy is suboptimal in the Australian setting. Our selected Australian SPT panel may assist clinicians screening for allergy.

Allergic sensitisation is the first step in the pathogenesis of allergic disease. Socio-economic and geographic factors, including rural environment and climate, affect patterns of allergic sensitisation.^{1,2} Recognising sensitisation patterns may inform more effective allergen avoidance strategies, and help guide approaches to testing for and treating allergies. In particular, it may influence the choice of immunotherapy, a treatment modality that reduces symptoms and medication use, and which modifies disease in the long term.³

Our study explored airborne allergen (aeroallergen) sensitisation patterns in the Greater Sydney area, and their relationships with climate, proximity to the coast, and environment (urban or rural). As co-sensitisation patterns may be important when making choices about allergy testing and immunotherapy, we also explored patterns of co-sensitisation to temperate and tropical grasses, and to two house dust mite (HDM) species, *Dermatophagoides farinae* and *Dermatophagoides pteronyssinus*.

Methods

In a retrospective, cross-sectional, multicentre study, we analysed the electronic database records for patients of three Sydney allergy clinics who had undergone aeroallergen skin prick testing (SPT) during the period January 2001 – October 2014. Electronic records were available from January 2001 for one clinic, from January 2009 for the second, and from January 2013 for the third. One author (AWK) reviewed the records and extracted data on sex, date of birth, SPT results, and age and postcode at the time of testing. If patients had undergone multiple SPT, only the most recent results were analysed. Patients were excluded if their postcodes were outside the Greater Sydney area (hereafter: “Sydney”) as defined by the Australian Bureau of Statistics’ (ABS) Australian Statistical Geographic Standard⁴ (Box 1) or if their most recent SPT test was invalid.

Abstract

Objective: To examine patterns of airborne allergen (aeroallergen) sensitisation in the Greater Sydney area (Sydney), and their relationships with climate, coastal proximity and environment (urban v regional).

Design, setting, participants: Retrospective cross-sectional study of patients who underwent aeroallergen skin prick testing at three Sydney allergy clinics, January 2001 – October 2014.

Main outcome measurements: Proportions of patients sensitised to specific aeroallergen types; relationships between sensitisation patterns and climate and geography.

Results: Of 1421 patients who met the selection criteria (mean age, 28.3 years [SD, 21.3]; 53.3% were female), 1092 (76.8%) were sensitised to at least one aeroallergen. Those living less than 15 km from the coast were less commonly sensitised to cockroach (< 15 km, 15.1%; 15–30 km, 40.0%; > 30 km, 39.7%; $P < 0.001$) and grass aeroallergens (< 15 km, 36.5%; 15–30 km, 52.2%; > 30 km, 58.1%; $P < 0.001$) than patients further inland; the same applied to mould, weed and tree aeroallergens. Subtropical grass sensitisation was more common in temperate/warm summer climates (about 50%) than in temperate/hot summer (27.1%) or subtropical climates (15%) ($P < 0.001$), and less common in urban (36.7%) than in regional areas (54%; $P = 0.014$). 72.4% of grass-sensitised patients were co-sensitised to both temperate and subtropical grasses. A selected ten-aeroallergen skin prick test panel identified 98.5% of atopic patients in this Sydney sample.

Conclusions: Environmental and geographic factors are associated with different patterns of allergic sensitisation in Sydney. Extensive co-sensitisation to subtropical and temperate grasses has implications for immunotherapy in Australia, where most currently available therapies are based on formulations directed at temperate grasses only.

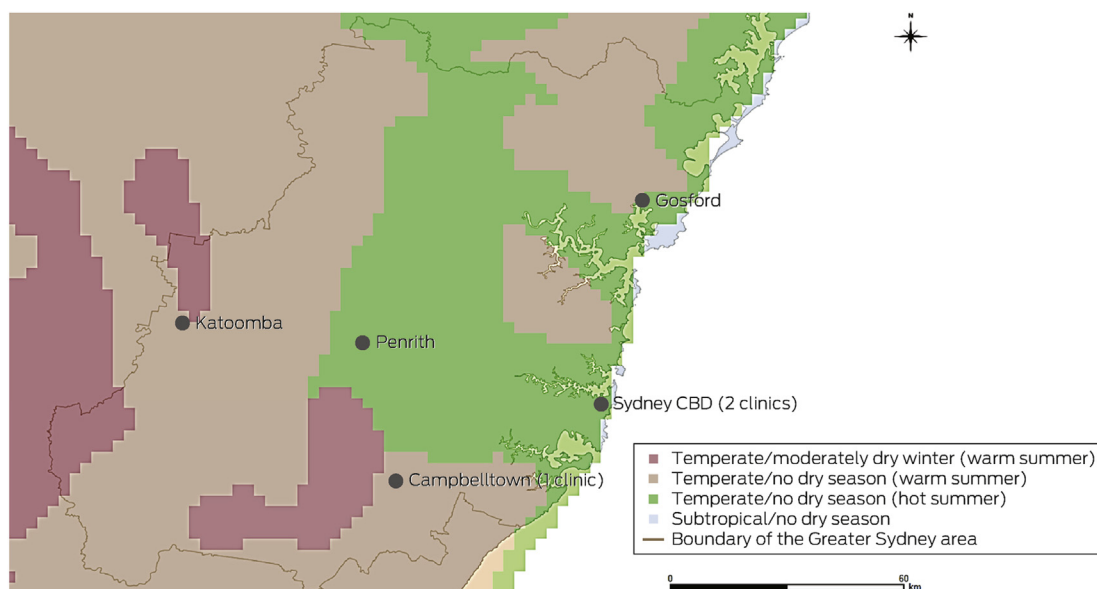
Skin prick testing

Each clinic performed SPT according to the Australasian Society of Clinical Immunology and Allergy guidelines.⁵ SPT results were reported as the mean diameter (mm) of the wheal reaction to the testing reagent. Criteria for valid tests were that the mean diameter of the wheal induced by the negative (ie, non-allergenic) control, phenolated glycerol saline, was no greater than 3 mm, and that the wheal induced by the positive control, 10 mg/mL histamine dihydrochloride (directly elicits a cutaneous wheal and flare response), was more than 4 mm wider than that of the negative control.⁵ Patients were deemed sensitised to an allergen if the mean diameter of the induced wheal was at least 3 mm if there was no reaction to the negative control, or more than 3 mm larger than that of the negative control if there was a reaction to the negative control.⁵

The allergens tested were those known to be present in Sydney. We grouped these allergens according to shared characteristics: HDM (*D. farinae*, *D. pteronyssinus*); animals (cat, dog); cockroach mix;

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1 The Köppen climate zones of Greater Sydney^{4,9}



moulds (*Aspergillus*, *Alternaria*, *Cladosporium*, *Penicillium*); weeds (plantain, *Parietaria*, *Oleaceae* mix); trees (plane tree, pine tree mix [*Pinus contorta* and *P. ponderosa*], birch tree); temperate grasses (five grass mix [timothy, rye, meadow, sweet vernal, cocksfoot], rye grass, timothy grass); and tropical grasses (Bermuda grass, Bahia grass). HollisterStier Allergy and Stallergenes (Alyostal) reagents were used during testing. Some patients were exposed to different SPT panels, as clinically indicated at presentation.

Definition of coastal habitation

Each patient was assigned to one of three coastal habitation groups according to the distance of their postcode from the coastline (< 15 km, 15–30 km, > 30 km). To assign these groups, a map of ABS postcode areas⁶ (vector format) was opened in ArcGIS (ESRI), a geographic information system program. Each postcode area was converted to a point on the map determined by its geometric centroid (a single coordinate representing the average of all points in the postcode area). The distance of the centroid of each postcode from the nearest point on the coastline was then determined.

Climate zone classification

Climate zones were defined according to the Australian Köppen climate classification of the Australian Bureau of Meteorology (Appendix 1, Box 1).⁷ Four climate zones were defined in Sydney: subtropical/no dry season; temperate/no dry season (hot summer); temperate/no dry season (warm summer), and temperate/moderately dry winter (warm summer).^{7,8}

To determine the climate zone of each postcode, a digitised map (vector format) of ABS postcode areas⁶ was overlaid with a digitised map (raster format) of the Australian Köppen climate zones in ArcGIS (ESRI).⁹ Postcodes were assigned the predominant climate zone (by area) within their boundaries.

Definition of urban and regional areas

Urban and regional areas were defined according to the Accessibility/Remoteness Index of Australia+ (ARIA+), an Australian government-endorsed geographic measure of remoteness.¹⁰ Two

remoteness categories were defined in the Sydney area, “Major cities of Australia” (in this article: “urban”) and “Inner regional Australia” (“regional”).

Data analysis

The proportions of patients sensitised and co-sensitised to the tested aeroallergens were calculated. To determine the ten-aeroallergen panel that provided the highest detection rate of atopic individuals, descriptive analyses assessed all possible aeroallergen combinations. These analyses were performed in SPSS 22.0 (IBM). Confidence intervals for proportions of patients exhibiting aeroallergen sensitisation were calculated by the Clopper and Pearson method in GraphPad Prism 6.04 (GraphPad Software).

Differences in the proportions of positive SPT results between climate zones, coastal and inland areas, and urban and regional areas were analysed in χ^2 tests. Z-tests (with Bonferroni correction) identified significant pairwise differences between specific climate zones or coastal habitation areas. Analysis of data for coastal habitation areas by χ^2 tests was adjusted for climate zone to assess the interaction of their effects. Sub-analysis of the 563 patients aged 16 years or less evaluated the influence of changes of address on results; it was assumed that children were less likely to have moved house as often as adults. These analyses were performed in SPSS 22.0.

Ethics approval

Ethics approval was granted by the St Vincent’s Hospital Human Research Ethics Committee (reference, LNR/14/SVH/88).

Results

A total of 1421 patients met the selection criteria. The mean age at testing was 28.3 years (SD, 21.3); 757 patients were female (53.3%). As expected of tertiary allergy services, there was a high proportion of sensitised patients, with 1092 (76.8%; 95% confidence interval [CI], 74.6–79.0%) sensitised to at least one aeroallergen. The

distribution of patients between climatic and geographic zones is summarised in [Appendix 2](#).

Across Sydney, the most common sensitising aeroallergens were HDM (63.2% of tested patients; 95% CI, 60.6–65.7%) and grasses (46.3%; 95% CI, 43.6–49.0%). Sensitisation to temperate grasses (44.5%; 95% CI: 41.7–47.4%) was more common than to subtropical grasses (37.6%; 95% CI, 34.7–40.4%; $P < 0.001$). The most common sensitising aeroallergens are listed in [Box 2](#).

Among the 1092 patients sensitised to at least one aeroallergen, 1057 (96.8%) had undergone both HDM and grass pollen SPT; of these, 995 (94.1%) were sensitised to at least one of these allergen groups, and 484 (45.8%) were sensitised to both HDM and grass pollen.

Of the 901 patients who had undergone testing to all eight allergen groups, 124 (13.8%) were sensitised to only one allergen group (mono-sensitised), of whom 72% were mono-sensitised to HDM aeroallergen ([Box 3](#)).

Climate zone influence

There were no climate zone differences in the patterns of HDM and animal aeroallergen sensitisation. A temperate/warm summer climate, however, was associated with a higher proportion of patients sensitised to cockroach, mould, weed, tree, and temperate or subtropical grass allergens than were temperate/hot summer and subtropical climates ([Box 4](#)).

A sub-analysis of patients aged 16 years or younger found that HDM sensitisation was more common in temperate/hot summer climates (72.9%) than in temperate/warm summer climates (moderately dry winter, 51.8%; no dry season, 59.6%; $P < 0.001$). There were no significant differences in patterns of sensitisation between climate zones for other allergen groups (data not shown).

Coastal habitation

There was no relationship between coastal proximity and patterns of HDM and cat sensitisation. Lower proportions of patients residing less than 15 km from the coast were sensitised to cockroach, mould, weed, tree, temperate grass and subtropical grass aeroallergens than those further inland ([Box 5](#), [Appendix 3](#)).

All participants living in temperate/moderately dry winter, warm summer zones also lived more than 30 km from the coast, while all in subtropical climate zones lived more than 15 km from the coast. In both temperate/no dry season zones, the pattern of increasing sensitisation further from the coastline remained significant after adjustment for climate zone, with two exceptions: there was no relationship between coastal proximity and sensitisation to cockroach and mould in patients living in temperate/no dry season, warm summer climates (data not shown).

A sub-analysis of tested patients aged 16 years or younger indicated that those residing less than 15 km from the coast were less commonly sensitised to mould, weed, tree and subtropical grass aeroallergens than patients further inland ($P < 0.05$ for each). Similar patterns were observed for cockroach and temperate grass sensitisation, but were not statistically significant ($P = 0.063$ and $P = 0.055$ respectively) (data not shown).

Urban and regional residence

Lower proportions of patients in urban areas were sensitised to cockroach, mould and subtropical grass aeroallergens than in regional areas. There were no differences for other allergen groups ([Box 6](#)). Although the differences in a sub-analysis of patients aged 16 years or younger were similar, they were not statistically

2 Proportion of patients exhibiting sensitisation to specific aeroallergens

Allergen	Number of patients tested	Number sensitised	Proportion sensitised (95% CI)
House dust mite	1404	887	63.2% (60.6–65.7)
<i>D. farinae</i>	496	269	54.2% (49.7–58.7)
<i>D. pteronyssinus</i>	1404	878	62.5% (59.9–65.1)
Animals	1303	517	39.6% (37.2–42.1)
Cat	1274	452	35.5% (32.8–38.2)
Dog	1132	256	22.4% (20.2–24.7)
Cockroach mix	983	295	30.0% (27.2–33.0)
Moulds	1295	347	26.8% (24.4–29.3)
<i>Alternaria</i>	1242	255	20.5% (18.3–22.9)
<i>Aspergillus</i>	1213	154	12.7% (10.9–14.7)
<i>Cladosporium</i>	872	137	15.7% (13.4–18.3)
<i>Penicillium</i>	628	76	12.1% (9.7–15)
Weeds	1207	387	32.1% (29.4–34.8)
Plantain	1204	340	28.2% (25.7–30.9)
<i>Parietaria</i>	636	27	4.2% (2.8–6.1)
<i>Oleaceae</i> mix	634	68	10.7% (8.4–13)
Trees	972	155	15.9% (13.7–18.4)
Plane tree	932	65	7.0% (5.4–8.8)
Pine mix	564	33	5.9% (4.1–8.1)
Birch tree	616	116	18.8% (15.8–22.1)
Temperate grass	1209	538	44.5% (41.7–47.4)
Five grass mix	188	68	36.2% (29.3–43.5)
Rye grass	1103	490	44.4% (41.5–47.4)
Timothy grass	605	328	54.2% (50.1–58.2)
Subtropical grass	1137	427	37.6% (34.7–40.4)
Bermuda grass	1129	383	33.9% (31.2–36.8)
Bahia grass	755	331	43.8% (40.3–47.5)
All grasses	1350	625	46.3% (43.6–49.0)
Any aeroallergen	1421	1092	76.8% (74.6–79.0)

3 Aeroallergen groups to which 124 mono-sensitised patients were sensitised*

Allergen group	Number mono-sensitised (%)
House dust mite	89 (72%)
Animals	8 (6%)
Cockroach	3 (2%)
Moulds	11 (9%)
Weeds	4 (3%)
Trees	1 (1%)
Temperate grasses	8 (6%)
Subtropical grasses	0

*This table includes only patients who had undergone skin prick testing for all eight allergen groups (901 patients) and who were sensitised to aeroallergens of only one allergen group. ♦

4 Aeroallergen sensitisation according to climate zone of residence

Allergen	Temperate/moderately dry winter, warm summer			Temperate/no dry season, warm summer			Temperate/no dry season, hot summer			Subtropical			P ⁱ
	Sensitised (%)	Number tested	Group*	Sensitised (%)	Number tested	Group*	Sensitised (%)	Number tested	Group*	Sensitised (%)	Number tested	Group*	
House dust mite	58%	116	—	63.2%	476	—	64.4%	781	—	52%	31	—	0.29
Animals	33%	106	—	42.1%	416	—	39.6%	750	—	32%	31	—	0.30
Cockroach	38%	84	α	38.5%	361	α	24.1%	514	β	0	24	γ	< 0.001
Mould	29%	102	α, β	36.3%	419	β	22.0%	744	α, γ	3%	30	γ	< 0.001
Weeds	37%	83	α, β	42.6%	371	β	26.6%	723	α	20%	30	α, β	< 0.001
Trees	13%	85	α, β	24.6%	362	β	11.0%	501	α	0	24	α	< 0.001
Temperate grasses	46%	109	α, β	53.6%	435	β	39.3%	638	α, γ	15%	27	γ	< 0.001
Subtropical grasses	50%	93	α	52.3%	398	α	27.1%	619	β	15%	27	β	< 0.001
All grasses	50%	111	α, β	55.2%	440	β	41.9%	768	α	16%	31	γ	< 0.001
Total patients tested		119			483			788			31		

* Z-test (with Bonferroni corrections) for pairwise comparison of proportions of patients sensitised to each allergen group in each climate zone. Significant differences ($P < 0.05$) are indicated by allocating climate zones different Greek letters: α zones are significantly different from β and γ zones, but not from other α zones. For example, the proportion of patients sensitised to grass allergens in temperate/moderately dry winter, warm summer zones (50%) was significantly different to the proportion for subtropical zones (16%), but not the proportions in the other two temperate zone categories (55.2%, 41.9%). † χ^2 test. ◆

5 Aeroallergen sensitisation in according to coastal proximity of residence

Allergen	< 15 km from coastline			15–30 km from coastline			> 30 km from coastline			P ⁱ
	Sensitised (%)	Number tested	Group*	Sensitised (%)	Number tested	Group*	Sensitised (%)	Number tested	Group*	
House dust mite	62.6%	621	—	66.0%	468	—	60.0%	315	—	0.22
Animals	40.5%	612	—	36.8%	410	—	42.0%	281	—	0.33
Cockroach	15.1%	391	α	40.0%	355	β	39.7%	237	β	< 0.001
Mould	17.4%	610	α	37.2%	406	β	32.3%	279	β	< 0.001
Weeds	23.2%	611	α	36.9%	355	β	47.3%	241	γ	< 0.001
Trees	24.8%	613	α	39.2%	380	β	50.0%	256	γ	< 0.001
Temperate grass	33.9%	496	α	49.9%	421	β	54.8%	292	β	< 0.001
Subtropical grass	20.4%	491	α	46.1%	384	β	57.3%	262	γ	< 0.001
All grasses	36.5%	619	α	52.2%	435	β	58.1%	296	β	< 0.001
Total number of patients tested		676			472			323		

* Z-test (with Bonferroni corrections) for pairwise comparison of proportions of patients sensitised to each allergen group in each climate zone. Significant differences ($P < 0.05$) are indicated by allocating climate zones different Greek letters: α zones are significantly different from β and γ zones, but not from other α zones. † χ^2 test. ◆

significant (cockroach, $P = 0.153$; moulds, $P = 0.456$; subtropical grasses, $P = 0.190$) (data not shown).

Co-sensitisation

A total of 1135 patients underwent both temperate and subtropical grass SPT. Of the 554 sensitised to any grass, 401 (72.4%) were sensitised to both temperate and subtropical grasses; 128 patients (23.1%) were sensitised to temperate grasses only, and 25 (4.5%) to subtropical grasses only.

A total of 496 patients underwent both *D. pteronyssinus* and *D. farinae* SPT. Of the 301 sensitised to HDM, 260 (86.4%) were sensitised to both species, while 32 patients (11%) were sensitised to *D. pteronyssinus* only and 9 (3%) to *D. farinae* only.

Testing panel

A ten-aeroallergen testing panel consisting of *D. pteronyssinus*, cat, dog, cockroach mix, *Alternaria*, *Aspergillus*, plantain, rye grass, timothy grass, and either Bermuda or Bahia grass (the two grasses yielded the same results) identified 98.5% of all sensitised patients (1076 of 1092).

Discussion

This study explored patterns of aeroallergen sensitisation in Sydney. Other authors have hypothesised that *D. pteronyssinus* is the predominant HDM allergen in Sydney;¹¹ indeed, it has been found that *D. farinae* comprises only 5.1% of HDMs found in house dust in Sydney.¹² Other studies of HDM species

6 Aeroallergen sensitisation in urban and regional areas

Allergen	Urban		Regional		P*
	Sensitised (%)	Number tested	Sensitised (%)	Number tested	
House dust mite	63.1%	1341	65%	63	0.79
Animals	39.5%	1242	43%	61	0.69
Cockroach	29.1%	932	47%	51	0.011
Mould	26.3%	1238	39%	57	0.047
Weeds	31.6%	1154	42%	53	0.14
Trees	15.8%	923	18%	49	0.69
Temperate grass	44.0%	1150	54%	59	0.14
Subtropical grass	36.7%	1083	54%	54	0.014
Grasses	45.8%	1289	56%	61	0.15
Total number of patients tested		1358		63	

* χ^2 test. ♦

prevalence reported that *D. farinae* is rarely found in Australian cities, suggesting the predominance of *D. pteronyssinus* across the nation,¹³⁻¹⁵ an interpretation supported by our results. Only a small proportion of our Sydney sample were sensitised solely to *D. farinae* (3%) and a comparatively higher proportion to *D. pteronyssinus* alone (11%). *D. farinae* and *D. pteronyssinus* allergen cross-reactivity probably explains the high proportion of HDM-sensitised patients who were co-sensitised to both species (86.4%).

A panel of ten aeroallergens — *D. pteronyssinus*, cat, dog, cockroach mix, *Alternaria*, *Aspergillus*, plantain, rye grass, timothy grass, and either Bermuda or Bahia grass — may be a useful screening panel in Sydney, having identified 98.5% of atopic patients in our study. This panel, modified by adding or subtracting aeroallergens according to regional variations in the predominance of allergens, may also be useful elsewhere in Australia.

We found a high proportion of co-sensitisation to temperate and subtropical grasses in the Sydney area (72.4%), consistent with findings by studies in subtropical Australian regions.¹⁶ This differs from findings in Europe and North America, where temperate grasses are predominantly responsible for grass pollen sensitisation.^{17,18} Temperate and subtropical grass pollens have both shared and distinct immunological properties, and the IgE reactivity of residents of temperate and subtropical regions is higher to the grass group of the corresponding climate.^{16,19,20} This suggests that differences in temperate and subtropical grass immunological reactivity are clinically relevant. As such, the high proportion of patients co-sensitised to temperate and subtropical grasses which we found indicates that both temperate and subtropical grass testing and immunotherapy may be important in Sydney and other temperate and subtropical regions. As most immunotherapeutic agents, particularly newer sublingual immunotherapy tablets, are directed against temperate grass aeroallergens alone, they may not provide adequate coverage in Australia.^{16,21} Treatments directed against both temperate and subtropical grasses may be required. Bermuda grass testing and immunotherapy may be particularly relevant, as it is less cross-reactive with temperate grasses than Bahia grass.²⁰

Our results confirmed that sensitisation to *Alternaria*, grass, weed and tree is less common in residents near the coast.^{11,22,23} Further, cockroach, *Aspergillus* and *Cladosporium* sensitisation is less common in areas less than 15 km from the sea. The climatic

conditions of the coast (including increased humidity, thermal stability, and coastal breezes) may offer some protection against these aeroallergens.

Interestingly, regional patients were more frequently sensitised to cockroach than were urban residents. A possible explanation is that our "regional" areas lay on the outskirts of Sydney, and were areas with lower household incomes and socio-economic status,²⁴ both of which are risk factors for sensitisation to cockroach aeroallergens.^{25,26}

The strengths of our study included the fact that it was the first to examine associations between sensitisation to specific allergen groups and climatic and geographic factors in an Australian setting over distances of tens of kilometres. It is also the first to report the prevalence of aeroallergen sensitisation in the Sydney area in a cohort of this size. Further, by separately analysing sensitisation for those under 16 years of age, who were unlikely to have changed addresses as often as older people, the study provides strong evidence of the effect of coastal habitation on reducing sensitisation rates to mould, weed, tree and subtropical grass aeroallergens. Finally, this study is more likely to reflect patterns of clinically relevant sensitisation because it used SPT results to define sensitisation, rather than allergen-specific IgE blood tests; further, it was restricted to patients who had presented to allergy specialists.

Our study, however, has limitations. Firstly, as postcodes and not exact addresses defined places of residence, microclimate and geographic location could not be precisely allocated for each person. Secondly, the proportion of people with allergic sensitisation in our investigation would be higher than in the general population, as we studied patients attending tertiary allergy clinics. Finally, as some patients were tested with different SPT panels, according to clinical indication at presentation, the prevalence of sensitisation we report may be higher than if all patients had undergone SPT for each of the allergens. This limitation was unavoidable, given the retrospective nature of our study. We also note that clinical reactivity requires both allergen sensitisation and subsequent allergen exposure. As such, sensitisation is clinically relevant only when there is also a risk of exposure to the allergen.

In conclusion, our study yields three major insights. Firstly, allergic sensitisation to a variety of different aeroallergens was less

common in patients attending allergy clinics who resided less than 15 km from the coast, in temperate/hot summer or subtropical climates, or in urban parts of Sydney. These relationships may affect decisions about allergen reduction and avoidance measures. Secondly, a ten-aeroallergen SPT panel identified more than 98% of atopic patients in Sydney; this panel may assist clinicians when screening for allergy. Finally, currently available immunotherapeutic options, based on northern hemisphere temperate grass allergens, may be inappropriate in the Australian setting in view of the high degree of co-sensitisation to

temperate and subtropical grasses; regimens directed at both grass types would be more suitable for treating grass allergy in subtropical regions.

Competing interests: Richard Harvey is a consultant for Medtronic, Olympus and Neilmed, and received grant support from Meda Pharmaceuticals, Stallergenes, ENT Tech and Neilmed. Janet Rimmer has received honoraria from Novartis, Sanofi Aventis, Mundipharma, BioCSL and Stallergenes. Constance Katelaris has received honoraria from Novartis, BioCSL, Stallergenes and Sanofi Aventis.

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