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ORIGINAL ARTICLE

Tay-Sachs disease preconception screening in Australia: self-knowledge of being an Ashkenazi Jew predicts carrier state better than does ancestral origin, although there is an increased risk for c.1421+1G>C mutation in individuals with South African heritage

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Abstract The Australasian Community Genetics Program provided a preconception screening for Tay-Sachs disease (TSD) to 4,105 Jewish high school students in Sydney and Melbourne over the 12-year period 1995–2007. By correlating the frequencies of mutant HEXA, MIM *606869 (gene map locus 15q23-q24) alleles with subjects' nominated ethnicity (Ashkenazi/Sephardi/Mixed) and grandparental birthplaces, we established that Ashkenazi ethnicity is a better predictor of TSD carrier status than grandparental ancestral origins. Screening self-identified Ashkenazi subjects detected 95% of TSD carriers (carrier frequency 1:25). Having mixed Ashkenazi and non-Ashkenazi heritage reduced the carrier frequency (1:97). South African heritage conveyed a fourfold risk of c.1421+1G>C mutation compared with other AJ subjects (odds ratio (OR), 4.19; 95% confidence interval (CI), 1.83–9.62, p=0.001), but this was the only specific case of ancestral origin improving diagnostic sensitivity over that based on determining Ashkenazi ethnicity. Carriers of c.1278insTATC mutations were more likely to have heritage from Western Europe (OR, 1.65 (95% CI, 1.04–2.60), p=0.032) and South Eastern Europe (OR, 1.77 (95% CI, 1.14–2.73), p=0.010). However, heritage from specific European countries investigated did not significantly alter the overall odds of TSD carrier status.

Keywords Tay-Sachs disease · Australia · South Africa · Jewish · Screening

Abbreviations
AJ Ashkenazi Jewish
TSD Tay-Sachs Disease

Nonstandard abbreviations are used in this paper.

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Introduction

Tay-Sachs disease (TSD) is a fatal neurodegenerative lysosomal sphingolipid storage disorder caused by mutations of *HEXA* MIM *606869 (gene map locus 15q23-q24). The *HEXA* gene product is the α-subunit of β-hexosaminidase, a dimeric enzyme involved in the lysosomal degradation of GM2 gangliosides. TSD carrier frequency is approximately 1:25 in individuals of Ashkenazi Jewish (AJ) descent (Kolodny 2009). TSD incidence in Jewish people is one in 3,900 births, compared to one in 320,000 births in the general population (Triggs-Raine et al. 2001). Several mutant *HEXA* alleles have been demonstrated in the AJ population (Arpaia et al. 1988).

The Australian Jewish community (estimated population 104,000) mainly reside in Melbourne and Sydney (Rubinstein 1995). The majority of Australian Jews have Ashkenazi heritage (Rutland 2005).

The Australasian Community Genetics Program (Laboratory and Community Genetics Department, Pacific Laboratory Medicine Services [PaLMS, Pathology North], Royal North Shore Hospital, Sydney) facilitates senior high school student and young adult outreach preconception genetic screening for TSD and other recessive diseases common in the AJ community (Gaucher disease, cystic fibrosis, mucolipidosis type IV, Fanconi anemia, familial dysautonomia, Canavan disease, Bloom syndrome) (Barlow-Stewart et al. 2003). Over 50% of Jewish adolescents in Melbourne and Sydney attend Jewish high schools that access TSD screening programs (Australian Bureau of Statistics 2006).

From 1995 to 2007, we conducted in Sydney TSD genetic screening tests for 4,105 Jewish high school students from Sydney and Melbourne. The Melbourne subgroup have previously been reported in part (Gason et al. 2005), and the design of the Sydney program has also been described (Barlow-Stewart et al. 2003). These screening programs are based on established best-practice principles, and the program design draws on the experience of overseas TSD screening programs (Ekstein and Katzenstein 2001; Bach et al. 2007; Lowden and Davidson 1977; Kaback et al. 1977). However, over time, individuals screened are becoming drawn from a population that is demographically different from any preceding generation. Social change such as intermarriage, both within the Jewish community (Ashkenazi/Sephardi) and within the general Australian community (Jewish/non-Jewish) may be changing their risk profiles. It is therefore possible that strategies of offering screening to the entire Jewish community through high school and adult preconception access points may become less effective over time due to lack of identification of subjects as being at risk for TSD carrier status.

Our study has reviewed key demographic and genealogical parameters of subjects and correlated them with the encountered TSD carrier frequencies of different mutant *HEXA* alleles in the modern Australian AJ community, with respect to self-recognition of being AJ, and with grandparental country of origin. We separately compared findings from Australia's two largest cities, Sydney and Melbourne, to maximize the likelihood of detecting local community trends or differences. From these studies, we sought to clarify the risk profiles of the current generation of Jewish youth choosing preconception TSD carrier screening in the context of planning screening strategies for the future.

Materials and methods

The study was approved by the Hawkesbury Human Research Ethics Committee of the Northern Sydney Central Coast Area Health Service of the New South Wales Government Department of Health.

Between 1995 and 2007, TSD carrier screening was offered through a community-funded program to 16–18-year-old students attending Jewish high schools in both Sydney and Melbourne. Participation was voluntary and informed through pretest genetic counseling. Participants provided written consent for TSD genetic testing, with optional additional consent for involvement in further research and development. In Sydney, all students also answered a demographic questionnaire at the time of testing. Data collected included nominated ethnicity (Ashkenazi/Sephardi/mixed Jewish/non-Jewish), the country of birth of the participants, of their two parents, and their four grandparents. In Melbourne, this questionnaire was answered only by participants screened between 1997 and 2002. Data from all subjects who answered the questionnaire have been included in the study. Further follow-up contact with individual subjects was limited to only those subjects who had consented to involvement in research and development.

TSD carrier screening was performed using DNA extracted either from venous blood samples (1995–2004) or from buccal cell wash sample (2005–2007). Results of testing were entered into a secure database system and de-identified prior to this study. Our laboratory methods were as described previously (Warren et al. 2005); note that our laboratory protocol requires HEXA mutations identified by enzyme analyses to be confirmed by *HEXA* DNA analysis before inclusion in our database. HEXA enzyme testing was replaced by *HEXA* DNA-only testing from 2005 onwards. DNA-only-based testing is designed to detect the three common *HEXA* mutations in the AJ population (Table 1). DNA-only testing has been shown to be a highly sensitive and cost-effective method at detecting heterozygotes in an orthodox AJ cohort of 38,197 individuals in Israel (Bach et al. 2001), with equivalent specificity compared to enzyme-based testing. In Bach's cohort, enzyme testing had variable sensitivity (93.1–99.1%) and specificity (88.1–98.8%) and, amongst 151 obligate carriers tested, no low-prevalence mutations were found. Enzyme testing in theory affords the possibility of detecting low-prevalence *HEXA* DNA mutations (Triggs-Raine et al. 1990). In contrast to the findings of Bach et al., we identified in our cohort two low-prevalence HEXA DNA mutations by enzyme testing and confirmed by DNA sequencing. However, due to the low prevalence of these mutations, our group agrees with the finding of Bach et al.

Table 1 Common HEXA mutations and their frequencies in Jewish populations previously studied (National Centre for Biotechnology Information gene and protein sequence reference NM_000520.4: NP_000511.2)

Common HEXA mutations in AJ populations	Description of mutation	Myerowitz and Costigan 1988 (20 AJ carriers)	Peleg et al. 1994 (152 AJ carriers)	Grebner and Tomczak 1991 (148 AJ carriers)	Paw et al. 1990 (156 AJ carriers)	Average of these results
c.1278insTATC	4-base pair insertion in exon 11→ frame shift→stop codon (Myerowitz and Costigan 1988)	14 (70%)	125 (82%)	108 (73%)	114 (73%)	361/476 (76%)
c.1421+1G>C	G→C transversion in the donor splice site of intron 12 (Arpaia et al. 1988; Ohno and Suzuki 1988)	–	15 (10%)	26 (18%)	24 (15%)	65/456 (14%)
p.Gly269Ser	Missense mutation (Ohno et al. 1988)	–	6 (4%)	5 (3%)	6 (4%)	17/456 (4%)
Other		–	6 (4%)	9 (6%)	12 (8%)	27/456 (6%)

that DNA-only testing is currently the most cost-effective method in a heterozygote AJ population.

Published analysis of this screening program found it to be effective, with high uptake, low negative perceptions, and high knowledge levels amongst participants. (Ioannou et al. 2010a, b)

Demographic classification as to whether the subjects considered themselves to be AJ, Sephardi, or mixed was by self-declaration from the questionnaire responses provided.

Grandparents' country of birth was also obtained from the subjects' questionnaires. Responses were stratified into regions (Table 2), based on political geographic boundaries and language to reflect Jewish community life in Europe prior to World War II (WWII), with additional reference to the Australian Bureau of Statistics Standard Australian Classification of Countries (second edition) (Harper 2008). The resultant geographic groupings we used were comparable to those used in past published papers investigating the frequencies of TSD carriers amongst AJ and non-AJ groups during the premolecular and molecular era (Risch et al. 2003; Peleg et al. 1994; Myrianthopoulos and Melnick 1977).

We also regrouped the data on grandparents' birthplaces so that individuals were categorized into only two groups: having no grandparent or else having at least one grandparent from any specific region/country. Using this classification schema, note that individuals could belong to more than one ancestral group.

All statistical analyses were conducted using the SPSS v15.0 (IBM SPSS, Inc., Somers, NY, www.spss.com). *P* values less than 0.05 were considered statistically significant.

Logistic regression analyses were performed to determine if country groups or individual countries were significant predictors of carrier status. In the series of logistic regression analyses, the dependent variables were either c.1278insTATC mutation status (carrier/noncarrier) or c.1421+1G>C mutation status (carrier/noncarrier). The small sample size and number of cases of p.Gly269Ser

Table 2 Geographic country groupings (constructed to study differential HEXA allele carrier frequencies by region of grandparents' birthplace)

Country groupings	Member countries (for purposes of this table)
Western Europe	Austria, Belgium, Denmark, Finland, France, Germany, Holland/Netherlands, Italy, Norway, Portugal, Spain, Sweden, Switzerland
North Eastern Europe	Belarus, Estonia, Latvia, Lithuania, Moldova, Poland, Russia, Ukraine, Siberia
South Eastern Europe	Bulgaria, Croatia, Czechoslovakia, Hungary, Romania, Serbia, Slovakia, Yugoslavia
UK	England, Ireland, Scotland, Wales, UK (unspecified)
Southern Africa	Mozambique, Namibia, South Africa, Zambia, Zimbabwe
North Africa and the Middle East	Egypt, Iran, Iraq, Lebanon, Libya, Morocco, Syria, Tunisia, Turkey, Yemen, Middle East (unspecified)
Australia/New Zealand	Australia, New Zealand
Israel/Palestine	Israel, Palestine
Other countries	Afghanistan, Argentina, Armenia, Azerbaijan, Barbados, Bolivia, Brazil, Burma, Canada, Chile, China, Cyprus, Fiji, Greece, India, Indonesia, Japan, Kazakhstan, Malaysia, Mexico, Mongolia, Pakistan, Peru, Philippines, Singapore, Sri Lanka, Trinidad and Tobago, USA, Uzbekistan, West Indies
Unknown	Unknown, entry blank

Table 3 Nominated ethnicity amongst Australian Jewish subjects studied

Ethnic group	Sydney, *N*=2,846% (*n*)	Melbourne, *N*=1,259% (*n*)	Combined, *N*=4,105% (*n*)
Ashkenazi	76.9 (2,186)	81.3 (1,024)	78.2 (3,210)
Sephardi	3.2 (92)	1.8 (23)	2.8 (115)
Mixed	13.7 (391)	7.5 (94)	11.8 (485)
Unknown	6.2 (177)	9.4 (118)	7.2 (295)

mutations prevented a similar analysis for p.Gly269Ser mutation status. Either a country group or else an individual country was entered as a covariate using a forced method of entry. The logistic regression analyses generated odds ratios with 95% confidence intervals, and absence of a grandparent from the major country group or individual country was considered the reference category. Data from the Sydney and Melbourne groups were initially analyzed separately and, where no statistically significant difference of results was found, results were then pooled and reanalyzed.

Results

Nominated ethnicity amongst AJ subjects studied is expressed in Table 3. Table 4 describes the proportion of subjects studied with at least one grandparent from each predefined geographical region (Table 2) and from the nine most common countries of origin. Table 5 describes the relative proportions of common and de novo *HEXA* mutations found in our study population. The two most prevalent mutations seen, c.1278insTATC and c.1421+1G>C, were further classified into ancestral regional groupings (Table 6).

AJ ethnicity is a good predictor of being a TSD mutation carrier (X^2=69.07, *df*=1, p<0.001). Students with European ancestry were more likely to be TSD carriers (X^2=2,247.24, *df*=9, p<0.001); however, further analysis by individual European country of origin did not increase the predictive power. Melbourne had a significantly higher proportion of AJ subjects, compared with Sydney (X^2=10.48, *df*=1, p=0.001) (Table 3), and the two cities also had differences in the relative proportions of subjects whose ancestors were from different European geographic origins (Table 4). South African ancestry conveyed a fourfold increased likelihood of carrying the mutation c.1421+1G>C (OR, 4.19 (95% CI, 1.83–9.62), p=0.001) compared with other AJ subjects. Odds generated from logistic regression analysis comparing c.1278insTATC and c.1421+1G>C carrier status and grandparents' birthplace for AJ subjects are summarized in Table 7.

Discussion

The most likely explanation for the origin of multiple HEXA mutations in AJ populations is that they arose around 1100 AD by founder effect and genetic drift (Slatkin 2004; Durst et al. 2001; Risch et al. 1995; Goldstein et al. 1999; Niell et al. 2003; Frisch et al. 2004). Four independent sphingolipid storage diseases have arisen in the AJ population (TSD, Niemann-Pick disease, Gaucher disease, and mucolipidosis type IV) leading some investigators to hypothesize a heterozygote advantage (Zlotogora et al. 1988; Motulsky 1995; Myrianthopoulos and Melnick 1977).

The geographic ancestral origins of the Jewish population screened for TSD carrier status in our study were

Table 4 Individuals with at least one grandparent from defined geographic groupings (regions/countries)

Region	Sydney, *N*=2,846% (*n*)	Melbourne, *N*=1,259% (*n*)	Country	Sydney, *N*=2,846% (*n*)	Melbourne, *N*=1,259% (*n*)
Europe (unspecified)	68.6 (1,953)	87.2 (1,098)	Ukraine	3.8 (107)	4.7 (59)
Western Europe	19.5 (554)	19.8 (249)	Lithuania	9.7 (276)	4.4 (56)
North Eastern Europe	47.0 (1,337)	71.2 (897)	Czechoslovakia	8.0 (227)	10.0 (126)
South Eastern Europe	21.8 (620)	22.0 (277)	Germany	11.7 (332)	12.6 (159)
South Africa	32.2 (916)	11.0 (138)	England	16.3 (464)	9.8 (124)
UK	18.3 (522)	11.0 (139)	Hungary	11.8 (337)	10.1 (127)
North Africa+Middle East	7.8 (221)	5.6 (71)	Russia	12.9 (366)	17.2 (217)
Australia/New Zealand	22.7 (646)	23.6 (297)	Poland	26.2 (747)	54.6 (687)
Israel/Palestine	4.7 (133)	6.8 (86)	South Africa	32.0 (910)	11.0 (138)
Other	9.8 (279)	7.0 (88)			

Table 5 Australian AJ mutation profile; *HEXA* mutations identified in our study population

	Sydney	Melbourne	Combined
Sample size	2,186	1,024	3,210
Total TSD carriers	95	42	137
Noncarriers	2,091	982	3,073
Mutation-specific carrier absolute number/frequencies:			
Total TSD carriers	95 (43:1,000)	42 (41:1,000)	137 (43:1,000)
c.1278insTATC	67 (31:1,000)	34 (33:1,000)	101 (32:1,000)
c.1421+1G>C	20 (9:1,000)	5 (5:1,000)	25 (8:1,000)
p.Gly269Ser	4 (2:1,000)	2 (2:1,000)	6 (2:1,000)
p.Arg24Tryp	2 (1:1,000)	0	2 (1:1,000)
p.Phe304del	0	1 (1:1,000)	1 (<1:1,000)
Private mutations			
p.Arg499Cys	1 (1:2,000)	0	1 (<1:1,000)
p.His204Pro	1 (1:2,000)	0	1 (<1:1,000)

extremely diverse. When grandparents' country of birth was examined, 86 countries and five continents were represented. North American demographic studies of AJ TSD carrier frequency have identified varying carrier frequencies between AJ communities founded by immigrants from different regions of Europe (Toronto, 1:14; Baltimore, 1:22; and Washington D.C., 1:28; Average USA, 1:30) (Lowden and Davidson 1977). However, these studies were based on results of enzyme-based carrier testing and therefore made no distinction between individual AJ HEXA allele frequencies among the subpopulations studied.

Although a small AJ community has existed in Australia since the time of European colonization of Sydney in 1788, the majority of the Australian AJ population was founded by immigration subsequent to WWII. Similar to American AJ immigration patterns, focussed communities of immigrants with shared recent language and heritage settled separately in both Melbourne and Sydney, resulting in slightly different subpopulation profiles. Significantly, more subjects from Melbourne identified as AJ than from Sydney (81.3% vs. 76.8%), correlating to reports of more grandparents born in Russia (17.2% vs. 12.9%) and Poland

Table 6 AJ origin of grandparents by region, carrier status, and allele frequency of two common *HEXA* mutations (N=12,840)

Regional groups	Total (N=12,840), n (% of N)	Non-carrier (N_1=12,332), n (% of N_1)	Carrier (N_2=508), n (% of N_2)	Absolute carrier frequency ratio (%)	c.1278insTATC (N_3=376), n (% of N_3)	c.1278insTATC carrier frequency ratio (%)	c.1421+1G>C (N_4=96), n (% of N_4)	c.1421+1G>C carrier frequency ratio (%)
Europe (all)	7,005 (54.5)	6,740 (54.7)	265 (52.1)	1: 26 (3.8)	217 (57.8)	1:32 (3.1)	22 (22.9)	1:318 (0.3)
North Eastern Europe	4,310 (33.6)	4,166 (33.8)	144 (28.3)	1:30 (3.3)	113 (30.1)	1:39 (2.6)	20 (20.8)	1:215 (0.5)
Southern Africa	2,672 (20.8)	2,534 (20.5)	138 (27.2)	1:19 (5.2)	75 (19.9)	1:36 (2.8)	55 (57.3)	1:49 (2.1)
South Eastern Europe	1,489 (11.6)	1,428 (11.6)	61 (12.0)	1:24 (4.1)	59 (15.7)	1:25 (4.0)	2 (2.1)	1:745 (0.1)
Western Europe	969 (7.5)	919 (7.5)	50 (9.8)	1: 19 (5.2)	38 (10.1)	1:26 (3.9)	0 (0.0)	Unknown (0)
Australia/New Zealand	1,172 (9.1)	1,134 (9.2)	38 (7.5)	1:31 (3.2)	34 (9.0)	1:34 (2.9)	4 (4.2)	1:293 (0.3)
UK	785 (6.1)	762 (6.2)	23 (4.5)	1:34 (2.9)	17 (4.5)	1:46 (2.2)	4 (4.2)	1:196 (0.5)
Uncertain	606 (4.7)	590 (4.8)	16 (3.1)	1:38 (2.6)	9 (2.4)	1:67 (1.5)	7 (7.3)	1:87 (1.2)
Other	350 (2.7)	335 (2.7)	15 (3.0)	1:23 (4.3)	13 (3.5)	1:27 (3.7)	2 (2.1)	1:175 (0.6)
Unspecified Europe	237 (1.8)	227 (1.8)	10 (2.0)	1:24 (4.2)	7 (1.9)	1:34 (3.0)	0 (0.0)	Unknown (0)
North Africa/ Middle East	53 (0.4)	45 (0.4)	8 (1.6)	1:7 (15.1)	8 (2.1)	1:7 (15.1)	0 (0.0)	Unknown (0)
Israel/Palestine	197 (1.5)	192 (1.6)	5 (1.0)	1:39 (2.5)	3 (0.8)	1:66 (1.5)	2 (2.1)	1:99 (1.0)

N.B. The carrier category contains n=376 grandparents of a c.1278insTATC carrier, 96 grandparents of a c.1421+1G>C carrier and 40 grandparents of subjects with other mutations

Table 7 Odds of being a c.1278insTATC/c.1421+1G>C carrier considering grandparents' birthplace for Ashkenazi subjects: none versus at least one

Regional groups	Zero grandparents			≥1 Grandparent					
	Total	c.1278insTATC carrier	c.1421+1G>C carrier	Total	c.1278insTATC carrier	c.1278insTATC carrier OR (95% CI)	*P* value	c.1421+1G>C carrier OR (95% CI)	*P* value
Europe (all)	732	17	14	2,478	77	1.35 (0.79–2.30)	0.270	0.21 (0.09–0.47)	<0.001
North Eastern Europe	1,327	40	14	1,883	54	0.95 (0.63–1.44)	0.808	0.50(0.22-1.13)	0.096
Southern Africa	2,286	70	9	924	24	0.84 (0.53–1.35),	0.480	4.18 (1.82–9.57)	0.001
South Eastern Europe	2,473	62	22	737	32	1.77 (1.14–2.73)	0.010	0.30 (0.071–1.29)	0.11
Western Europe	2,570	67	24	640	27	1.65 (1.04–2.60)	0.032	0.00 (0.00)	0.99
Australia/New Zealand	2,511	70	22	699	24	1.24 (0.77–1.99)	0.37	0.33 (0.076–1.38)	0.128
UK	2,698	82	21	512	12	0.77 (0.42–1.41)	0.393	0.75 (0.22–2.53)	0.64
North Africa/ Middle East	3,172	89	24	38	5	5.24 (2.00–13.77)	0.001	0.00 (0.00)	0.99
Israel/Palestine	3,068	92	23	142	2	0.46(0.11–1.90)	0.284	0.94 (0.13–7.00)	0.95

(54.6% vs. 24.2%). A larger proportion of Sydney compared with Melbourne AJ subjects had grandparents from South Africa (32.0% vs. 11.0%).

Demographic differences within the Sydney and Melbourne communities did not affect overall TSD carrier frequency in our study, which was 3.3% in both cities with no significant difference between the cities (Table 5). AJ TSD carrier frequencies were also comparable (Melbourne, 4.1%; Sydney, 4.3%) and similar to other AJ populations worldwide (Kaback et al. 1977), suggesting participants correctly identified their AJ origins. Self-identification of AJ ethnicity correlated statistically with a higher proportion of grandparents from North Eastern Europe, South Africa, and South Eastern Europe (X^2=2,247.24, df=9, p<0.001). Confirmation of subjects' correct self-identification of AJ heritage and TSD carrier risk has not been previously reported in a screened Jewish population outside of Israel and USA in the current at-risk generation.

Variation in the frequencies of the c.1278insTATC and c.1421+1G>C mutations was seen in the Sydney and Melbourne subpopulations, mirroring demographic differences by grandparents' country of birth demonstrated in these cities.

Uptake of screening in individuals approached by our group in Sydney and the Melbourne branch has been very high (approaching 100%) since the advent of cheek brush/mouthwash sampling. Ioannou et al. published a paper in 2010 evaluating TSD screening in Melbourne using a purpose-designed questionnaire exploring student knowledge (disease and genetics), reasons for screening, anxiety, and predicted negative feelings if found to be a carrier. Two hundred seventy-three students were offered screening and 272 (99.6%) completed the questionnaire. Only two students chose not to have screening (Ioannou et al. 2010a, b). Unfortunately, demographic information on our questionnaire has only been collected from study participants. Although it is therefore not possible to determine the ethnicity of any students who may have refused testing, given the very small number of students declining testing the potential for a negative ascertainment bias on this basis is extremely unlikely to have influenced our findings.

Frisch et al. (2004) identified a conserved c.1278insTATC haplotype in 55 unrelated AJ individuals, suggesting the occurrence of a common founder in Central Europe. The c.1278insTATC mutation was diagnosed in 73.2% of Australian carriers (Sydney, 69.8%; Melbourne, 81.0%), comparable to the figure of 70–82% reported in other AJ populations (Peleg et al. 1994; Grebner and Tomczak 1991; Paw et al. 1990; Myerowitz and Costigan 1988). However, grandparents' birthplace in specific European countries or regions showed no significant relationship with grandchildren's risk of c.1278insTATC carrier status (Table 7).

The c.1421+1G>C mutation was diagnosed in 18.9% of Australian TSD carriers (Sydney, 21.1%; Melbourne, 11.9%), compared with 13% of American AJ TSD carriers (Arpaia et al. 1988).The c.1421+1G>C mutation carrier frequency was 1:49 in subjects with grandparents from South Africa (OR, 4.19 (95% CI, 1.83–9.62), p=0.001), compared to 1:129 in all Australian AJ subjects. The increased proportion of Sydney AJ subjects with grandparents from South Africa (32.0% vs. 11.0%) mirrored c.1421+1G>C increased frequency in Sydney vs. Melbourne subpopulations (0.00915, 1:109 vs. 0.00488, 1:204).

The Jewish community in South Africa is of Eastern European AJ origin (Meiner et al. 1991; Levin 2001), overwhelmingly originally from Lithuania (Tatz et al. 2007). The AJ population in Lithuania plummeted from the 755,000 recorded in Lithuania's 1897 census to 153,743

in its 1923 census (Lane et al. 1985), caused by emigration to South Africa, USA, and Canada. From 1941, Nazi genocide achieved near complete annihilation of the Jews of Kaunas and Vilnius provinces in Lithuania, the relatively small area from which more than half of the South African AJ population trace their ancestry (Lane et al. 1985). As a result, there is now no surviving European reference population. In a published survey conducted between 2003 and 2004, 608 Australian and New Zealand recent South African AJ immigrants made 697 mentions of ancestral homes in Kaunas (Kovno) province and 65 in the Vilnius (Vilna) province (Tatz et al. 2007). The South African AJ community expanded from 10,000 members in 1890 (Jenkins et al. 1977a, b; Lane et al. 1985) to 120,000 in the early 1980s. By 2007, an estimated 40% of South Africa's AJ population (47,000) had emigrated for social and political reasons (Tatz et al. 2007).

It has been postulated that c.1421+1G>C mutations may have existed at a higher allele frequency in a relatively small area of Lithuania by founder effect and might be preserved in individuals with grandparents from South Africa. In data analyzed from the Dor Yeshorim TSD screening program in New York and Jerusalem; Risch et al. (2003), demonstrated a higher c.1421+1G>C allele frequency in subjects with at least one grandparent from Lithuania (0.0113) compared with mixed AJ group (0.0041), where 3,718 of 249,372 grandparents with Lithuanian origins. Risch et al. (2003) included an unspecified number of subjects with South African ancestry in the Lithuanian group but did not report of a significant result in the subjects with South African heritage as a subgroup. In our study, c.1421+1G>C mutation carrier frequency was not increased among the small sample of Australian subjects identifying at least one grandparent from Lithuania (1:300).

In 1985, the TSD carrier frequency among the Jews of South Africa was estimated to be 1:23 (Lane et al. 1985). The National Health Laboratory service (previously the South African Institute for Medical Research) has offered a genetic screening service for TSD since the 1970s which can be availed by individuals at risk (personal communication Dr. Amanda Krause and Ms. Fahmida Essop 2009). Six of 43 AJ individuals screened by them for TSD were c.1421+1G>C carriers (14% of individuals tested, 18% of 33 TSD carriers identified) a proportion greater than the 13% of TSD carriers expected for American AJ populations. This result is not directly comparable to our data, given the different mode of patient selection and small sample size. Further evaluation is undoubtedly required; however, the trend is in the same direction as our Australian data. AJ ancestry in our cohort was a risk factor for TSD inheritance (X^2=69.07, df=1, p<0.001). Mixed heritage (AJ/Sephardi/non-Jewish) was shown to dilute this risk.

One hundred thirty out of 137 of the HEXA mutation carriers detected through screening in our study were in AJ self-identifying individuals. A policy of screening individuals with AJ ethnicity would detect the majority of HEXA mutation carriers with sensitivity of 95%. Amongst 895 individuals tested who did not self-identify as AJ, but as "Sephardi," "uncertain," "Jewish," or "mixed" ethnicity, seven carriers were identified. The majority of these non-AJ carriers (five of seven) were found in individuals with "mixed" ethnicity (carrier frequency, 1:97; 1%) representing 3.6% of all carriers. All five mixed ethnicity carriers were found to express the c.1278insTATC mutation. One carrier of the c.1278insTATC mutation identified origins as uncertain, and one carrier of the c.1421+1 G>C identified as Jewish.

Two AJ individuals screened by enzyme analysis prior to 2005 were found to carry private mutations. One individual carried the p.Arg499Cys mutation with grandparents from Poland. The p.Arg499Cys mutation has been previously seen in several ethnic groups including Polish (Mules et al. 1992).

The second mutation p.His204Pro has not been previously described, and we are in the process of characterizing this (unpublished data). Grandparental ancestry of this subject was Dutch and English.

Our research strategy involved the use of a de-identified database of results. De-identification of data resulted in the possibility of non-acknowledgment of familial relationships within the database for both carriers and noncarriers. c.1278insTATC carrier status was observed in five individuals with at least one grandparent from the North African/Middle Eastern region (OR, 5.25; 95% CI, 2.00–13.76, p=0.001). These individuals all nominated their ethnicity as AJ, suggesting European ancestry. The five subjects, all of whom consented for their results to contribute to research and development, were re-identified at arm's length by a secondary investigator post-analysis, and a familial relationship was confirmed in two of the five cases.

A strategy of investigating the relationship between grandparent's birthplace and TSD carrier status has some inherent weaknesses. Without family pedigree testing, the grandparent from whom an identified mutation was inherited remains uncertain. Grandparent and parent carriers with noncarrier offspring are undetected.

In countries like Australia which have become adopted homes to immigrant Jewish communities, new generations of AJ descent are experiencing continuing evolution of cultural history and identity. Demography of the generation we studied revealed the effect of the Australian "melting pot," with a large proportion of Jewish subjects identifying as having mixed ethnicity. Our program was successful in identifying its target population as 78.6% of subjects screened identified as AJ. However, students attending

non-Jewish high schools were not able to be offered cohort TSD screening, representing a significant proportion of young people remaining without access to screening and thus at risk for being TSD carriers with decreased opportunity to gain that knowledge.

Despite postulated demographic changes that have been occurring since earlier studies of HEXA mutation frequencies of AJ populations were undertaken (Lowden and Davidson 1977; Kaback et al. 1977), the risk profile of AJ individuals in our study regarding TSD carrier status remains undiluted (carrier frequency, 1/25).

Conclusions

Nominated AJ ethnicity was the single best predictor of TSD carrier risk in our study. Screening only those individuals identified as having AJ heritage would identify 95% of all Australian Jewish TSD carriers.

Individuals of mixed AJ and non-AJ heritage have reduced risk of being TSD carriers (1:97). Individuals with one or more grandparents from South Africa had a fourfold greater risk of being a carrier of c.1421+1G>C mutation, compared with other AJ subjects.

The TSD carrier frequency for all mutations in Australian Jewish subjects is 1:30. The TSD carrier frequency for all mutations in Australian AJ subjects is 1:25. This proportion of AJ subjects who are TSD carriers is unchanged from previous international studies despite widespread demographic change and social influences such as intermarriage in the wider community.

These findings suggest that the policy approach remains sound in encouraging access to high school and preconception TSD carrier testing for all members of the Jewish community. However, should funding or resources limit the ability to undertake full community screening, then the alternative of screening only those subjects who identified themselves as being AJ would identify 95% of carriers.

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References

Arpaia E, Dumbrille-Ross A, Maler T, Neote K, Tropak M, Troxel C, Stirling JL, Pitts JS, Bapat B, Lamhonwah AM et al (1988) Identification of an altered splice site in Ashkenazi Tay-Sachs disease. Nature 333(6168):85–86

Australian Bureau of Statistics (2006) Census of Population and Housing; Department of Employment, Education and Training Census of Non-government Schools, http://abs.gov.au/websitedbs/D3310114.nsf/home/Census+data;2009. Accessed 17 June 2009

Bach G, Tomczak J, Risch N, Ekstein J (2001) Tay-Sachs screening in the Jewish Ashkenazi population: DNA testing is the preferred procedure. Am J Med Genet 15(1):70–75, 99

Bach G, Zeigler M, Zlotogora J (2007) Prevention of lysosomal storage disorders in Israel. Mol Genet Metab 90(4):353–357

Barlow-Stewart K, Burnett L, Proos A, Howell V, Huq F, Lazarus R, Aizenberg H (2003) A genetic screening programme for Tay-Sachs disease and cystic fibrosis for Australian Jewish high school students. J Med Genet 40(4):e45

Durst R, Colombo R, Shpitzen S, Avi LB, Friedlander Y, Wexler R, Raal FJ, Marais DA, Defesche JC, Mandelshtam MY, Kotze MJ, Leitersdorf E, Meiner V (2001) Recent origin and spread of a common Lithuanian mutation, G197del LDLR, causing familial hypercholesterolemia: positive selection is not always necessary to account for disease incidence among Ashkenazi Jews. Am J Hum Genet 68(5):1172–1188

Ekstein J, Katzenstein H (2001) The Dor Yeshorim story: community-based carrier screening for Tay-Sachs disease. Adv Genet 44:297–310

Frisch A, Colombo R, Michaelovsky E, Karpati M, Goldman B, Peleg L (2004) Origin and spread of the 1278insTATC mutation causing Tay-Sachs disease in Ashkenazi Jews: genetic drift as a robust and parsimonious hypothesis. Hum Genet 114(4):366–376

Gason AA, Metcalfe SA, Delatycki MB, Petrou V, Sheffield E, Bankier A, Aitken M (2005) Tay Sachs disease carrier screening in schools: educational alternatives and cheekbrush sampling. Genet Med 7(9):626–632

Goldstein DB, Reich DE, Bradman N, Usher S, Seligsohn U, Peretz H et al (1999) Age estimates of two common mutations causing factor XI deficiency: recent genetic drift is not necessary for elevated disease incidence among Ashkenazi Jews. Am J Hum Genet 64(4):1071–1075

Grebner EE, Tomczak J (1991) Distribution of three alpha-chain beta-hexosaminidase A mutations among Tay-Sachs carriers. Am J Hum Genet 48(3):604–607

Harper P (2008) Standard Australian Classification of Countries (SACC), 2nd Edn. Australian Bureau of Statistics Catalogue No. 1269.0. Australian Bureau of Statistics, Australia

Ioannou L, Massie J, Lewis S, Petrou V, Gason A, Metcalfe S, Aitken MA, Bankier A, Delatycki MB (2010) Evaluation of a multi-disease carrier screening programme in Ashkenazi Jewish high schools. Clin Genet 78(1):21–31

Jenkins T, Lane AB, Kromberg JG (1977a) Tay-Sachs disease screening and prevention in South Africa. S Afr Med J 51(4):95–98

Jenkins T, Lane AB, Kromberg JGR, Nurse GT (1977b) Tay-Sachs screening: current status in South Africa. Prog Clin Biol Res 18:61–67

Kaback MM, Nathan TJ, Greenwald S (1977) Tay-Sachs disease: heterozygote screening and prenatal diagnosis—U.S. experience and world perspective. Prog Clin Biol Res 18:13–36

Kolodny E (2009) Tay-Sachs Disease. Encyclopedia of Neuroscience, pp. 895–902

Lane AB, Dunn DS, Jenkins T (1985) A genetic profile of the South African Ashkenazi Jewish population. S Afr Med J 68(13):935–939

Levin D (2001) The Litvaks: a short history of the Jews in Lithuania. Yad Vashem Publications, Jerusalem

Lowden JA, Davidson J (1977) Tay-Sachs screening and prevention: the Canadian experience. Prog Clin Biol Res 18:37–46

Meiner V, Landsberger D, Berkman N, Reshef A, Segal P, Seftel HC, van der Westhuyzen DR, Jeenah MS, Coetzee GA, Leitersdorf E (1991) A common Lithuanian mutation causing familial hypercholesterolemia in Ashkenazi Jews. Am J Hum Genet 49(2):443–449

Motulsky AG (1995) Jewish diseases and origins. Nat Genet 9(2):99–101

Mules EH, Hayflick S, Miller C, Reynolds L, Thomas G (1992) Six novel deleterious and three neutral mutations in the gene encoding the alpha-subunit of hexosaminidase A in non-Jewish individuals. Am J Hum Genet 50(4):834–841

Myerowitz R, Costigan FC (1988) The major defect in Ashkenazi Jews with Tay-Sachs disease is an insertion in the gene for the alpha-chain of beta-hexosaminidase. J Biol Chem 263 (35):18587–18589

Myrianthopoulos NC, Melnick M (1977) Tay-Sachs disease: a genetic-historical view of selective advantage. Prog Clin Biol Res 18:95–106

Niell BL, Long JC, Rennert G, Gruber SB (2003) Genetic anthropology of the colorectal cancer-susceptibility allele APC I1307K: evidence of genetic drift within the Ashkenazim. Am J Hum Genet 73(6):1250–1260

Ohno K, Suzuki K (1988) A splicing defect due to an exon-intron junctional mutation results in abnormal beta-hexosaminidase alpha chain mRNAs in Ashkenazi Jewish patients with Tay-Sachs disease. Biochem Biophys Res Commun 153(1):463–469

Paw BH, Moskowitz SM, Uhrhammer N, Wright N, Kaback MM, Neufeld EF (1990) Juvenile GM2 gangliosidosis caused by substitution of histidine for arginine at position 499 or 504 of the alpha-subunit of beta-hexosaminidase. J Biol Chem 265 (16):9452–9457

Peleg L, Karpati M, Gazit E, Raas-Rothschild A, Goldman B (1994) Mutations of the hexosaminidase A gene in Ashkenazi and non-Ashkenazi Jews. Biochem Med Metab Biol 52(1):22–26

Risch N, de Leon D, Ozelius L, Kramer P, Almasy L, Singer B, Fahn S, Breakefield X, Bressman S (1995) Genetic analysis of idiopathic torsion dystonia in Ashkenazi Jews and their recent descent from a small founder population. Nat Genet 9(2):152–159

Risch N, Tang H, Katzenstein H, Ekstein J (2003) Geographic distribution of disease mutations in the Ashkenazi Jewish population supports genetic drift over selection. Am J Hum Genet 72(4):812–822

Rubinstein WD (1995) Judaism in Australia: religious community profiles Bureau of Immigration. Australian Government Publication Service, Canberra

Rutland SD (2005) The Jews in Australia. Cambridge University Press, Port Melbourne

Slatkin M (2004) A population-genetic test of founder effects and implications for Ashkenazi Jewish diseases. Am J Hum Genet 75 (2):282–293

Tatz C, Arnold P, Heller G (2007) Worlds apart: the re-migration of South African Jews. Publishing Pty Ltd., Australia

Triggs-Raine BL, Feigenbaum AS, Natowicz M, Skomorowski MA, Schuster SM, Clarke JT, Mahuran DJ, Kolodny EH, Gravel RA (1990) Screening for carriers of Tay-Sachs disease among Ashkenazi Jews. A comparison of DNA-based and enzyme-based tests. N Engl J Med 323(1):6–12

Triggs-Raine B, Mahuran DJ, Gravel RA (2001) Naturally occurring mutations in GM2 gangliosidosis: a compendium. Adv Genet 44:199–224

Warren E, Anderson R, Proos AL, Burnett LB, Barlow-Stewart K, Hall J (2005) Cost-effectiveness of a school-based Tay-Sachs and cystic fibrosis genetic carrier screening program. Genet Med 7 (7):484–494

Zlotogora J, Zeigler M, Bach G (1988) Selection in favor of lysosomal storage disorders? Am J Hum Genet 42(2):271–273