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Breast and prostate cancer risk are not associated with increased homozygosity in outbred populations

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ABSTRACT

Regions of restricted genetic heterogeneity due to identity by descent (autozygosity) are known to confer susceptibility to a number of diseases. Regions of germline homozygosity (ROH) of 1-2Mb, the result of autozygosity, are detectable at high frequency in outbred populations. Recent studies have reported that ROH, possibly through exposing recessive disease-causing alleles or alternative mechanisms, are associated with an increased cancer risk. To examine whether homozygosity is associated with breast or prostate cancer risk we analysed 500K SNP data from two genome-wide association studies conducted by Cancer Genetics Markers of Susceptibility initiatives (<http://cgems.cancer.gov/>). Six common ROH were associated with breast cancer risk and four with prostate cancer ($P < 0.01$). Intriguingly, one of the breast cancer ROH maps to 6q22.31-6q22.3 a region that has been previously shown to confer breast cancer risk. While none of the ROH remained significantly associated with cancer risk after adjustment for multiple testing, a number of the ROHs merit further interrogation. Our findings do however provide no strong evidence that levels of measured homozygosity, whatever their aetiology (autozygosity, uniparental isodisomy or hemizyosity) confer an increased risk of developing breast or prostate cancer in predominantly outbred populations.

INTRODUCTION

Breast and prostate cancer are among the most common cancers in western populations.

Twin studies indicate that inherited genetic factors contribute to the development of approximately 27% of breast and 42% of prostate cancer¹ and both cancers show two-fold elevated risks of the same disease in first-degree relatives of patients².

To date, no high penetrance susceptibility loci have been identified for prostate cancer and most of the inherited risk is thought to be a consequence of the co-inheritance of multiple low risk variants. While high penetrance susceptibility to breast cancer in the form of *BRCA1* and *BRCA2* mutations is well-recognised, classical mutations in these genes only account for ~20% of familial disease².

Recent genome-wide association (GWA) studies of breast and prostate cancers have identified several common single nucleotide polymorphisms (SNPs) at different loci that influence the risk of these tumors³⁻⁵. These risk variants are common in the general population (minor allele frequency [MAF] >5%), and are associated with odds ratios (ORs) typically <1.3. Despite the relatively small predisposing effects they individually confer on tumor risk, they can have more profound effects by acting in concert. Moreover, their identification has provided important and novel insights into the biology of both breast and prostate cancer.

The majority of cancer predisposition genes that have to date been identified through GWA studies act in a co-dominant fashion and studies have found no good evidence for recessively acting disease loci. Although this may be reflective of the biology, it may also be a consequence of GWA studies having suboptimal ability to detect recessively acting disease-alleles. Clues that tumor susceptibility may have a recessive basis come from some, but not all, studies showing an increased incidence associated with

consanguinity and in populations characterised by a high degree of inbreeding⁶⁻¹¹. Further evidence for the role of homozygosity in cancer predisposition is provided by experimental animal inbreeding (e.g. backcrossing mice) being associated with increasing tumor incidence¹², although such animal model systems are impoverished of variation compared to natural populations, their alleles being selected for survival in homozygous state. Specific situations of homozygosity have also been directly associated with cancer such as uniparental disomy through altered imprinting¹³. Common regions of homozygosity (ROH), the result of autozygosity, have recently been shown to occur at a high frequency in outbred populations as a result of selection¹⁴. Such selected regions are not likely to have, however, been selected in relation to cancer, which is generally late-onset relative to past human life expectancy. Searching for ROH on a genome-wide basis therefore provides a means of potentially exposing recessively acting disease genes. Recently Assié *et al.* studied breast, prostate and head/neck cancer patients of Northern/Western European ancestry by whole-genome loss of heterozygosity analysis using a series of microsatellite markers¹⁵. A significant increase in the frequency of homozygosity in cases compared with controls was reported. In a separate study of colorectal cancer using Affymetrix *Xba*I 50K SNP arrays, Bacolod *et al.* showed that cases harbored significantly more homozygous regions than healthy individuals¹⁶. Collectively, these data provide support to the hypothesis that there exist multiple, recessive, cancer-predisposing loci, which are not readily detected using a conventional GWA approach based on analysis of individual SNPs. A possible explanation for this is that relative risks per locus are too low and/or that the disease-associated variants are not in strong linkage disequilibrium (LD) with tagSNPs, perhaps because of low allele frequencies.

While GWA studies have limited ability to identify recessive disease causing alleles through single SNP analyses, these datasets can potentially be exploited to search for this class of susceptibility allele through whole genome homozygosity analysis (WGHA). Hence, to examine whether homozygosity is associated with an increased risk of developing breast or prostate cancer and to search for novel recessively-acting disease loci we conducted a WGHA of breast and prostate cancer GWA study data generated by the Cancer Genetic Markers of Susceptibility (CGEMS) initiative.

MATERIALS AND METHODS

Dataset

This study was based on GWA study data on breast and prostate cancer generated by Cancer Genetics Markers of Susceptibility (CGEMS), which are publicly accessible. Full details of the studies are provided in previously published material. Briefly, using Illumina Infinium HD Human550 Duo BeadChips (Illumina, San Diego, USA) the GWA study of breast cancer involved genotyping 1,183 breast cancer cases and 1,185 controls¹⁷. In the prostate GWA study 1,177 prostate cancer cases and 1,149 controls were genotyped using Illumina Infinium Human217 and Human 317 BeadChips (Illumina, San Diego, USA)¹⁸. 689 of the 1,177 prostate cancer cases analysed had aggressive disease, as defined by a Gleason score ≥ 7 and disease stage $\geq III$, and 488 prostate cancer cases had non-aggressive disease.

Quality control

We restricted our analysis to the autosomal SNPs. We considered that a DNA sample had failed if it did not generate a genotype for $>95\%$ of loci. Similarly, a SNP was

considered a failure if <90% of DNA samples generated a genotype at the locus. To identify samples showing relatedness, identity by state values (IBS) were calculated for pairs of individuals and for any pair with >80% identical SNP genotypes, we removed the sample with the lower call rate from the analysis. We excluded SNPs on the basis of deviation from Hardy-Weinberg Equilibrium (HWE) using a threshold of $P < 1 \times 10^{-3}$ in either the cases or controls. We also removed SNPs with minor allele frequency (MAF) <0.01. To identify and exclude individuals with non-Western European ancestry, case and control data was merged with individuals of different ethnicities from the International HapMap Project, genome-wide IBS distances for markers shared between HapMap and our SNP panel determined, and dissimilarity measures used to perform principal component analysis.

After imposing these stringent quality control measures for the breast cancer GWA study 512,159 SNP genotypes were available on 1,144 cases and 1,141 controls. For the prostate cancer GWA study 509,008 SNP genotypes were available on 1,168 cases (685 with aggressive disease) and 1,093 controls. These two datasets formed the basis of our WGH analysis.

Statistical and bioinformatics analysis

We detected ROH using PLINK¹⁹ v1.06 software. The ROH tool moves a sliding window of SNPs across the entire genome. To allow for genotyping error or other sources of artificial heterozygosity, such as paralogous sequences, within a stretch of truly homozygous SNPs and, hence, to prevent underestimating the number and size of ROHs, 2% heterozygous SNPs were permitted in each window. We left the remaining options set to the default values (including allowing 5 missing calls per window,

thereby ensuring >90% positive predictive value of each ROH), except that we varied the parameters for homozyg-snp according to our heuristic preferences for defining the ROH as detailed below. Subsequent statistical analyses were performed using packages available in R (version 2.7.0) and specifically written Perl code. Comparison of the distribution of categorical variables was performed using the χ^2 test. To compare the difference in average number of ROHs between cases and controls we used the Student t-test. Naïve adjustment for multiple testing was based on the Bonferroni correction.

We used three metrics to investigate the selection pressure on each ROH. Integrated Haplotype Score (iHS) is based on LD surrounding a positively selected allele compared to background, providing evidence of recent positive selection at a locus²⁰. An iHS score >2.0 reflects that haplotypes on the ancestral background are longer compared to the derived allelic background. Episodes of selection tend to skew SNP frequencies in different directions and Tajima's D is based on the frequencies of SNPs segregating in the region of interest²¹. Fixation index (F_{st}) measures the degree of population differentiation at a locus, taking values from 0 to 1.0²². iHS, D and F_{st} metrics were obtained from Haplotter Software²⁰.

Identification of runs of homozygosity

To examine commonly occurring ROH and to detect statistically significant associations we only considered ROH which occurred in 10 or more individuals, thereby ensuring our study had 80% or better power to identify statistically significant differences in ROH between cases and controls. The initial search for ROH was performed using PLINK¹⁹ with a specified length of 80 consecutive SNPs. This ROH

length was chosen to be more than an order of magnitude larger than the mean haploblock size in the human genome without being too large as to be very rare. The likelihood of observing 80 consecutive chance events can be calculated as follows¹⁴. In the breast cancer series, mean heterozygosity in the controls was calculated to be 34%. Thus, given 512,159 SNPs and 2,285 individuals, a minimum length of 58 would be required to produce <5% randomly generated ROH across all subjects ($(1-0.34)^{58} \times 512,159 \times 2,285 = 0.040$). A consequence of linkage disequilibrium (LD) is that the SNP genotypes are not always independent thereby inflating the probability of chance occurrences of biologically meaningless ROH. Analysis based on PLINK's pairwise LD SNP pruning function, with a default value of $r^2 > 0.8$ being necessary to declare that one SNP tags another and restricting the search of tags to within 250kb, showed 370,611 separable tag groups, representing a 27.6% reduction of information compared to the original number of SNPs. Thus ROH of length 80 were used to approximate the degrees of freedom of 58 independent SNP calls. Corresponding calculation in the prostate cancer series resulted in the same criterion being adopted.

Once all ROH of at least 80 SNPs in length were identified, these were pruned to only those ROH, which occurred in more than 10 individuals. To ensure that a minimum length and minimum number of SNPs in each ROH was maintained, each individual's SNP data was recoded as one if the SNP was in an ROH for that individual and zero otherwise. Then, for each SNP, those SNPs with less than 10 individuals coded as one were recoded to zero before removing any ROH that due to this recoding were now less than 80 SNPs in length. This process therefore resulted in a list of "common" ROH having a minimum of 80 consecutive ROH calls across 10 or more samples and with

each ROH having the identical start and end locations across all individuals where that ROH is observed.

RESULTS

Prior to conducting a WGHA of the two GWA datasets we subjected cases and controls to rigorous quality control in terms of excluding samples and SNPs with poor call rates. Furthermore, we excluded SNPs showing significant departure from HWE. We then critically evaluated datasets for ancestral differences by principal component analysis. Figure 1 shows that all sample series were ancestrally comparable and were representative of European ancestry after excluding individuals of different ethnicity.

A total of 415 and 426 ROH were identified in breast cancer series and prostate cancer series respectively (Supplementary Tables 1 and 2), encompassing approximately 35% of the genome as measured by both the total chromosomal length and the number of included SNPs in both series. Figure 2 shows the similarity between genome-wide plots of the location of each ROH among the genomes of both cancer cases and controls from the two studies.

In both datasets six ROH exceeded 12Mb in length and included ROH encompassing the centromeric regions of chromosomes 3, 5, 6, 8, 11 and 16 (Supplementary Tables 1 and 2). Five other ROH exceeded 12Mb in one of the series and were all greater than 8.7Mb in the other series. While the length of these large ROH across centromeres is partly a consequence of extended regions which are not annotated by SNPs this is however unlikely to be the sole explanation, as these centromeric regions are flanked by large homozygous regions. One of these centromeric regions (8p11.21-8q11.23) has been previously highlighted in several genome-wide studies of selective sweeps, thus

providing validation of our methodology^{20,23-25}. Five non-centromeric regions harboring ROH greater than 12Mb in length in one of the series and greater than 8.5Mb in the other series were also identified: 3q25.31-26.1, 4q13.1-13.3, 6q13.5-15, 7q31.1-32.1 and 14q13.3-22.1 (Supplementary Tables 1 and 2).

In both series the ROH covering the largest genomic region (28Mb) spanned the centromere of chromosome 3 (3p12.3-3q13.11); a region previously shown to be characterised by a high frequency of ROH in the European population²⁴. This ROH was also defined by the largest number of SNPs in a single ROH in both studies.

There were fifteen ROHs that had a frequency greater than 25% in the breast cancer control series and sixteen in the prostate cancer control series (Table 1). Thirteen of the ROHs were common to both datasets. Of those common to both datasets five have previously been reported to have high frequency and harbor several gene categories that have been identified to be influenced by a high degree of selective pressure^{20,23-25}. Publicly available data from HapMap do not indicate that these regions have excessive copy number variation or segmental duplication, nor do they have very low recombination rates²³. However, the high *iHS*, *D* and *F_{st}* metrics for each region are compatible with positive selection in the Caucasian samples from which the two case-series are based (Table 1).

The total number of common ROH observed in each individual was calculated to permit genome-wide comparison between the case and control groups in each of the two datasets. Each individual therefore was assigned a value between 0 and 415 in the breast cancer series and a value between 0 and 426 in the prostate cancer series. Overall, patients with breast cancer (mean = 21.10, SD = 4.75) and controls (mean = 20.97, SD = 4.75) showed no significant difference in the average number of ROH ($t_{2277} = 0.6135$,

$P = 0.54$). Similarly for prostate cancer overall statistics showed no significant difference ($t_{2255} = 0.5434$, $P = 0.59$) in the average number of ROH between cases (mean = 21.07, SD = 4.79) and controls (mean = 20.96, SD = 4.71).

Additionally, to examine if there were differences in the distributions of ROH in the genomes of cases and controls we computed the cumulative distributions for both series (Figure 3). This analysis also provided no support for a difference in autozygosity profiles between cases and controls in either dataset on a genome-wide basis. Furthermore, cases and controls from both series were similarly directly comparable.

At an individual level six ROH differed significantly ($P < 0.01$) between breast cancer cases and controls (Table 2). Five of these six ROH were more common in the cases than in the controls. ROH286 was identified in 1.4% of cases ($n=16$) compared with 0.2% of controls ($n=2$) ($P=0.0009$). ROH202 mapping to 6q22.31-22.3 was identified in 4.3% ($n=49$) of cases compared to 2.1% of controls ($n=24$). Intriguingly this region of association to which *ECHDC1* (enoyl coenzyme a hydratase domain-containing protein 1; MIM 612136) and *RNF146* (ring finger protein 146; MIM 612137) map has previously been linked to breast cancer susceptibility in a GWA study of Ashkenazi Jews²⁶. After adjusting for multiple testing, however, these associations and the others were not statistically significant.

Only one of the four ROH which differed significantly ($P < 0.01$) between prostate cancer cases and controls had a higher prevalence in cases (ROH86) occurring in 19.2% ($n=224$) of cases and 14.7% ($n=161$) of controls ($P=0.0049$; Table 2). Interestingly, *PCGEM1* (prostate-specific gene 1; MIM 605443) maps to the centromeric region of ROH86. *PCGEM1* is a prostate tissue-specific, and prostate cancer-associated noncoding RNA (ncRNA) gene, which plays a role in regulation of apoptosis^{27,28}.

PCGEM1 displays androgen-dependent expression and is overexpressed in ~80% of prostate cancer suggesting specific functions of *PCGEM1* in the biology and tumorigenesis of the prostate gland^{27,28}. ROH149 which maps to 4q28.2-28.3 was identified in 7.0% (n=82) cases compared with 10.1% (n=110) of controls ($P=0.0095$). An association between 4q28.2 defined by microsatellite D4S2394 and prostate cancer was identified by Assié *et al*¹⁵ ($P<0.001$). The clinical behaviour of prostate cancer is heterogeneous and is likely to reflect differences in tumor biology and possibly molecular etiology. In view of this we conducted a subgroup analysis of prostate cancer cases stratified by aggressive phenotype. This analysis did not however provide any further support for a relationship between ROH and prostate cancer risk (data not shown).

DISCUSSION

Two recent studies of cancer have reported that signatures of autozygosity correlate to tumor incidence and it has been proposed that these regions showing IBD may be the locations of genes contributing to tumor heritability^{15,16}. Moreover these data have been interpreted as providing an explanation for increased risk of a variety of cancers in inbred groups that has often been reported.

Here we have used a high-density genomic scan data to compare the structure of genetic variation in patients with breast and prostate cancer with healthy controls. By imposing stringent quality control we have ensured individuals in our study were from an apparently panmictic population with no evidence of stratification. Our data provides further evidence that ROH, ranging in size from 1 to 28Mb, are common in individuals from an outbred population²⁹⁻³². As documented in Table 1, the common ROH we have

identified in controls are representative of autozygosity due to distant consanguinity rather than chromosomal abnormalities or common copy number variants. Moreover, these homozygous regions are too common and small to be caused by recent consanguinity but rather are consistent with the regions under selective pressure³³. Based on our analysis there was however no evidence for an association between homozygosity and risk of breast or prostate cancer – either on the basis of total ROH size per individual or specific ROH.

The assertion that increased autozygosity correlates to cancer incidence provides an attractive explanation for reported increased cancer risk in inbred populations. However, as recently articulated, several criticisms can be levelled at this assertion. The observation of an increased cancer risk associated with consanguinity has often been based on studies of a small number of individuals in an isolated community or a single large family with a high level of inbreeding³⁴. Thus, the relevance of inbreeding to the population risk of cancer is unclear as inbreeding and founder effects may be confounded. Sample sizes in the molecular studies^{15,16}, which have sought to establish a relationship between ROH and cancer risk, have generally been small and, crucially, cases and controls groups ethnically heterogeneous or unmatched. Furthermore, the study of breast, prostate, and head/neck cancer reported by Assié *et al.* made use of relatively sparse microsatellite data. Here we have addressed these shortcomings in our study of breast and prostate cancer by analysing a large set of cases and controls that have been genotyped for several hundred thousand SNPs and imposed a high level of quality control both in terms of genotyping and sample ancestry.

Not advocating the use of uncorrected *P*-values, it is intriguing that our analysis identified two regions previously implicated in susceptibility to these cancers. While

this suggests that further interrogation of other regions may prove profitable, there is limited support from our analysis that WGHA of outbred populations provides a robust methodology for identifying novel cancer susceptibility loci.

In conclusion our findings do not provide evidence that levels of measured homozygosity, whatever their aetiology, from autozygosity, uniparental isodisomy or hemizyosity, confer an increased risk of developing either breast or prostate cancer in a predominantly outbred population. Moreover it is unlikely that there exist large numbers of recessive alleles that predispose to breast or prostate cancer and are unmasked by autozygosity in most European populations. This analysis does not exclude the possibility that recessively acting disease alleles exist for these cancers, whatever their penetrances, or that autozygosity may operate in populations characterised by a high level of inbreeding.

WEB ADDRESSES

PLINK: <http://pngu.mgh.harvard.edu/purcell/plink/>

R: <http://cran.r-project.org/>

Cancer Genetics Markers of Susceptibility (CGEMS): <http://cgems.cancer.gov/>

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FIGURE AND TABLE LEGENDS

FIG. 1 Comparison of ethnicity in each of the sample series. The first two principal components of the analysis were plotted. HapMap data is plotted in gray; CEU individuals as “●”, CHB+JPT individuals as “▲” and YRI individuals as “+”. Plotted in black, as “X”, are (a) Breast cancer cases (b) Prostate cancer cases, (c) Breast cancer controls and (d) Prostate cancer controls.

FIG. 2 Genome-wide plots showing the location of each run of homozygosity among the genomes in (a) Breast cancer cases and (b) controls; (c) Prostate cancer cases and (d) controls. The threshold limit was set to a minimum of at least 80 consecutive homozygous SNPs.

FIG. 3 Cumulative distributions of ROH in Breast cancer and Prostate cancer series. The graph is presented in such a way that each data point represents the cumulative fraction (y-axis) of the samples with the corresponding minimum cumulative run of homozygosity (x-axis).

TABLE 1 List of ROHs with frequency of >25% in either of the two control series. Chromosomal coordinates derived from the National Center for Biotechnology Information (NCBI) build 36.

*represent maximal values for alternate metrics of positive selection, derived from Haplotter (<http://hg-wen.uchicago.edu/selection/haplotter.htm>). The number of deletions, duplications, and recombination hotspots are derived from HapMap release 27 (<http://hapmap.org>).

TABLE 2 List of ROHs significantly associated with breast and prostate cancer risk ($P < 0.01$).

ROH	Chromosome	Type of cancer	Start (bp)	End (bp)	Length (bp)	Number of SNPs	Number of controls (%)	*iHSmax	*Tajima Dmax	*Fst max	Number of deletions/duplications/hotspots
ROH239	8p11.21 - 8q11.23	Breast	41,825,641	55,432,493	13,606,852	1140	660 (57.8)	3.80	2.25	0.83	10/5/34
ROH243		Prostate	41,830,189	53,468,390	11,638,201	802	655 (59.9)				
ROH104	3p12.3 - 3q13.11	Breast	77,216,563	105,137,759	27,921,196	2853	576 (50.5)	2.22	2.90	0.79	14/6/96
ROH108		Prostate	77,847,362	105,681,807	27,834,445	2759	564 (51.6)				
ROH306	11p11.2 - 11q12.2	Breast	45,354,793	60,278,000	14,923,207	1266	557 (48.8)	2.33	2.53	0.68	8/6/24
ROH311		Prostate	45,435,794	59,758,149	14,322,355	1137	459 (42.0)				
ROH129	4p15.1 - 4p14	Breast	32,049,891	36,697,547	4,647,656	590	455 (39.9)	3.70	2.64	0.84	2/6/32
ROH132		Prostate	31,396,728	36,697,547	5,300,819	666	424 (38.8)				
ROH74	2q21.2 - 2q22.1	Breast	134,402,624	141,119,426	6,716,802	1163	376 (33.0)	6.35	2.34	0.68	3/1/52
ROH75		Prostate	134,412,810	141,911,735	7,498,925	1366	389 (35.6)				
ROH189	6p22.2 - 6p21.31	Breast	25,260,653	33,998,429	8,737,776	2492	371 (32.5)	2.86	2.14	0.71	16/8/28
ROH188	6p22.2 - 6p21.32	Prostate	25,143,899	33,432,505	8,288,606	2310	346 (31.7)				
ROH165	5p13.1 - 5q11.2	Breast	39,470,981	51,995,314	12,524,333	1185	357 (31.3)	1.74	2.30	0.54	6/2/29
ROH164		Prostate	39,437,377	51,876,385	12,439,008	1159	333 (30.5)				
ROH179	5q23.2 - 5q31.1	Breast	127,011,681	132,523,084	5,511,403	727	348 (30.5)	2.00	1.69	0.64	3/1/24
ROH179		Prostate	125,929,340	132,561,958	6,632,618	876	355 (32.5)				
ROH230	7q31.1 - 7q32.1	Breast	111,717,516	127,567,084	15,849,568	2157	342 (30.0)	2.21	3.29	0.97	12/2/54
ROH233		Prostate	116,522,322	127,506,517	10,984,195	1513	340 (31.1)				

ROH100	3p21.31 - 3p21.1	Breast	45,268,383	53,571,572	8,303,189	746	341 (29.9)	1.37	1.51	0.68	15/4/16
ROH104		Prostate	45,797,441	53,710,806	7,913,365	663	256 (23.4)				
ROH324	12p11.21 - 12q12	Breast	32,513,364	41,213,854	8,700,490	1106	331 (29.0)	2.16	2.22	0.82	11/4/22
ROH331		Prostate	32,345,552	41,341,610	8,996,058	1183	270 (24.7)				
ROH289	10q22.1 - 10q22.2	Breast	73,174,430	76,830,688	3,656,258	359	329 (28.8)	1.99	2.85	0.76	1/0/10
ROH291		Prostate	73,048,417	77,330,771	4,282,354	467	319 (29.2)				
ROH336	12q21.31 - 12q21.33	Breast	82,014,452	90,780,916	8,766,464	1084	316 (27.7)	1.87	3.03	0.77	6/1/31
ROH341		Prostate	82,035,008	88,201,390	6,166,382	718	301 (27.5)				
ROH366	15q15.1 - 15q21.1	Breast	38,368,637	44,046,400	5,677,763	622	297 (26.0)	2.03	3.23	0.95	5/1/23
ROH378		Prostate	38,385,586	44,379,085	5,993,499	658	298 (27.3)				
ROH247	8q21.13 - 8q22.1	Breast	82,426,387	93,568,993	11,142,606	1501	287 (25.2)	1.75	2.59	0.73	8/9/59
ROH249	8q21.13 - 8q21.3	Prostate	83,012,725	93,265,947	10,253,222	1333	289 (26.4)				
ROH384	15q23 - 15q25.1	Prostate	69,643,146	76,421,786	6,778,640	889	355 (32.5)	2.26	3.14	0.72	7/1/31

TABLE 1 List of ROHs with frequency of >25% in either of the two control series. Chromosomal coordinates derived from the National Center for Biotechnology Information (NCBI) build 36

*represent maximal values for alternate metrics of positive selection, derived from Haplotter (<http://hg-wen.uchicago.edu/selection/haplotter.htm>). The number of deletions, duplications, and recombination hotspots are derived from HapMap release 27 (<http://hapmap.org>)

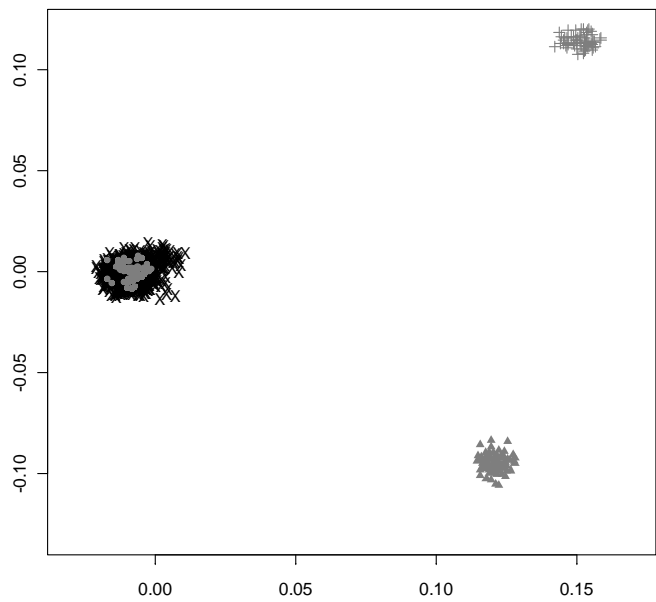
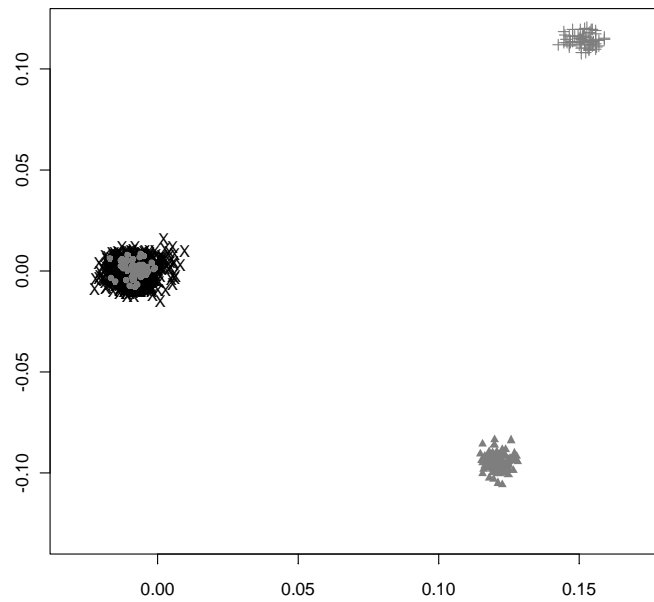
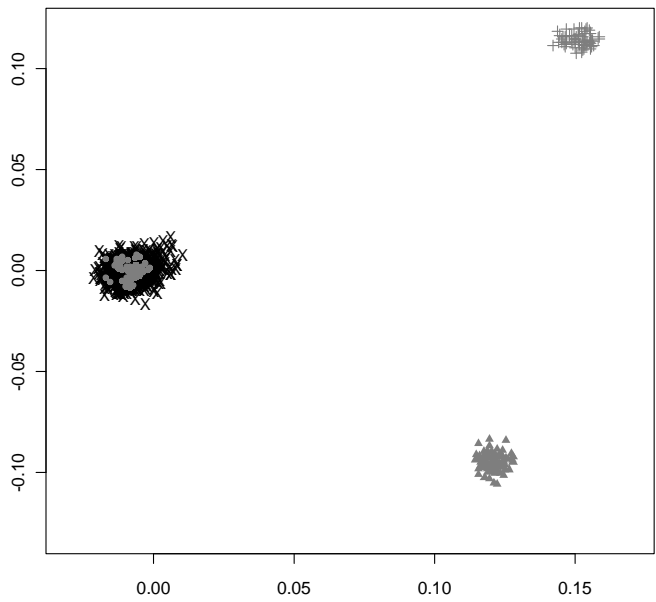
a.

ROH	Chromosome	Start (bp)	End (bp)	Length (bp)	Number of SNPs	Number of cases (%)	Number of controls (%)	Chi Square	P value
ROH286	10q21.2	62,297,871	63,439,845	1,141,974	187	16 (1.4)	2 (0.2)	10.9383	0.000942
ROH172	5q15 - 5q21.2	96,549,677	102,946,704	6,397,027	770	129 (11.3)	177 (15.5)	8.83979	0.002947
ROH202	6q22.31 - 6q22.33	125,818,032	129,746,282	3,928,250	497	49 (4.3)	24 (2.1)	8.77653	0.003051
ROH97	3p22.2	36,787,514	38,001,656	1,214,142	185	15 (1.3)	3 (0.3)	8.03181	0.004596
ROH110	3q21.2	126,407,221	127,368,259	961,038	115	10 (0.9)	1 (0.1)	7.37554	0.006612
ROH12	1p31.1	74,987,770	76,330,014	1,342,244	202	23 (2.0)	8 (0.7)	7.31802	0.006827

b.

ROH	Chromosome	Start (bp)	End (bp)	Length (bp)	Number of SNPs	Number of cases (%)	Number of controls (%)	Chi Square	P value
ROH86	2q32.3 - 2q33.1	192,910,770	201,169,666	8,258,896	898	224 (19.2)	161 (14.7)	7.90671	0.004925
ROH297	10q23.33	95,589,679	97,274,071	1,684,392	257	41 (3.5)	64 (5.9)	7.01252	0.008094
ROH307	11p14.1 - 11p13	29,779,181	32,102,771	2,323,590	272	56 (4.8)	81 (7.4)	6.78978	0.009168
ROH149	4q28.2 - 4q28.3	130,066,714	137,815,993	7,749,279	995	82 (7.0)	110 (10.1)	6.7305	0.009478

TABLE 2 List of ROHs significantly associated ($P<0.01$) with (a) Breast cancer and (b) Prostate cancer

A**B****C****D**