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High prevalence of luminal B breast cancer intrinsic subtype in Colombian women

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Abstract

Breast cancer is the most frequent malignancy in women worldwide. Distinct intrinsic subtypes of breast cancer have different prognoses, and their relative prevalence varies significantly among ethnic groups. Little is known about the prevalence of breast cancer intrinsic subtypes and their association with clinicopathological data and genetic ancestry in Latin Americans. Immunohistochemistry surrogates from the 2013 St. Gallen International Expert Consensus were used to classify breast cancers in 301 patients from Colombia into intrinsic subtypes. We analyzed the distribution of subtypes by clinicopathological variables. Genetic ancestry was estimated from a panel of 80 ancestry informative markers. Luminal B breast cancer subtype was the most prevalent in our population (37.2%) followed by luminal A (26.3%), non-basal triple negative (NBTN) (11.6%), basal like (9%), human epidermal growth factor receptor 2 (HER2) enriched (8.6%) and unknown (7.3%). We found statistical significant differences in distribution between Colombian region ($P = 0.007$), age at diagnosis ($P = 0.0139$), grade ($P < 0.001$) and recurrence ($P < 0.001$) according to intrinsic subtype. Patients diagnosed with HER2-enriched, basal-like and NBTN breast cancer had the highest African ancestry. Future studies analyzing the molecular profiles of breast cancer in Colombian women will help us understand the molecular basis of this subtype distribution and compare the molecular characteristics of the different intrinsic subtypes in Colombian patients.

Introduction

Breast cancer is the most frequent cancer type in women worldwide (1) representing a highly heterogeneous group of tumors with particular molecular features, prognosis and response to therapy (2–4). The first gene expression-based classification of breast cancer into intrinsic subtypes was published in 2000 (5), which included estrogen receptor positive (ER+) subtypes luminal A and B and ER subtypes basal-like and human epidermal growth factor receptor 2 enriched (HER2 enriched) (6). Subsequent studies showed differences in the outcomes according to intrinsic subtypes (7,8).

Molecular tests have become routine in breast cancer treatment planning. For example, the *PAM50* gene expression profile accurately classifies breast cancers into luminal A, luminal B, normal like, basal like and HER2 enriched (9) and can predict risk of recurrence in node-negative breast cancer patients (9,10). However, because of the logistical and financial obstacles to widespread implementation of nanostring assays, an immunohistochemistry (IHC)-based surrogate panel has been proposed (11) as a reasonable substitute, especially in underresourced health care systems.

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Abbreviations

EGFR	epidermal growth factor receptor
ER	estrogen receptor
FFPE	formalin-fixed paraffin embedded
HER2	human epidermal growth factor receptor 2
IHC	immunohistochemistry
NBTN	non-basal triple negative
PgR	progesterone receptor

There are ethnic differences in the relative prevalence of breast cancer intrinsic subtypes in the USA. Sweeney *et al.* used *PAM50* to classify breast cancer into intrinsic subtypes and found different distributions according to ethnicity (12). African-Americans have higher prevalence of basal-like cancers compared with non-Hispanic Whites and Asian-Pacific Islanders (30.4% versus 8.2% and 5%, respectively); US Hispanic/Latinas, compared with non-Hispanic Whites, have lower prevalence of luminal A (44.2% versus 55.2%, respectively) but relative increases in luminal B (24% versus 20.9%, respectively), HER2-enriched (15.6% versus 12.5%, respectively) and basal-like (11.6% versus 8.2%, respectively) cancers. It is unclear, however, whether this information is specific to US patients or can be replicated in other populations with different environmental exposures (lifestyle, diet etc.).

Hispanic/Latinas is an ethnic group with variable ancestry including different proportions of European, Indigenous American and African background (13). Colombia has one of the most diverse populations in Latin America. According to the National Administrative Department of Statistics (DANE, acronym for its name in Spanish), from a population of approximately 50 million, 86% of Colombians self-report as mestizos, which is defined as a person with mixed ancestry, 10.5% as Colombian-African and 3.4% as Indigenous Americans (14). Global statistics regarding age-adjusted incidence and age-adjusted mortality of breast cancer in Colombian women have been published (15). However, there is a lack of information about the distribution of the intrinsic subtypes, especially according to the genetic ancestry of the patient.

Our goal was to analyze the distribution of breast cancer intrinsic subtypes in Colombian women and its association with clinicopathological variables and genetic ancestry. We classified breast cancer by IHC from formalin-fixed paraffin embedded (FFPE) tumor blocks and determined the proportions of European, African and Indigenous American ancestry. The most important finding of our study was the high prevalence of luminal B intrinsic subtype in our sample of Colombian women.

Methods

Sample collection

The Colombian National Cancer Institute (INC) is a governmental body that has a double role in cancer control in the Country: (i) it advises the Ministry of Health on all national cancer-related issues (policies, strategies and surveillance for cancer control and prevention) and (ii) it is the national comprehensive reference center for cancer treatment. For the present study, we selected samples from an INC database containing 857 breast cancer patients diagnosed between 2008 and 2012. Our inclusion criteria comprised histologically confirmed diagnosis of primary invasive breast cancer and the availability of archival material (FFPE) that contained at least 10% of the tumor left in the paraffin block from mastectomies or breast-conserving surgeries. Patients with *in situ* disease or patients whose archival paraffin blocks only included biopsy materials were excluded. Slides from each patient were reevaluated by a group of pathologists to confirm the histological diagnosis.

A total of 301 breast cancer patients, 283 from the INC and 18 from the Hospital Universitario del Caribe (HUC), a public hospital from Cartagena, were included in this study. This study was approved by the INC and HUC ethics committee. Since we worked with deidentified FFPE tissues collected more than 3 years before the analysis done for this work, the Colombian NCI, according to the Colombian laws, considered that no informed consent was required.

Clinical and pathological characteristics

Pathology reports were reviewed to obtain information regarding histopathological diagnosis, nodal status, surgical margins and histological grade. Demographic information including place of birth, age at diagnosis as well as information on tumor size, clinical stage, treatments (neoadjuvant and/or adjuvant), recurrence and death was extracted from clinical records.

Assessment of IHC

Five intrinsic subtypes were defined by IHC according to the expression of ER, progesterone receptor (PgR), HER2, Ki67, Cytokeratin (CK5/6) and EGFR (epidermal growth factor receptor) in whole sections of the paraffin block selected following the scheme proposed by the St. Gallen panel 2013 (Table 1) (11).

Information on hormone receptor status and HER2 was obtained from previous pathology reports, if available. Expression of hormone receptors (ER and PgR) was considered positive when they exceeded 1% of nuclear staining in tumor cells. HER2 measurement was semiquantitative according to the recommendations of the American Society of Clinical Oncology (ASCO)/College of American Pathologists (CAP) guideline (16).

Based on previous reports (17), proliferation index was graded as 'low' if Ki67 staining was positive in less than 20% of tumor cells or 'high' when at least 20% of tumor cells stained positive. CK5/6 and EGFR were scored as positive if cytoplasmic staining was present in ≥ 10% of tumor cells.

Ancestry estimation

DNA was extracted from normal FFPE tissues using the RecoverAll™ Total Nucleic Acid Isolation Kit (Life Technologies, Carlsbad, CA) following the manufacturer's recommendations. A panel of 106 Single Nucleotide Polymorphisms previously validated as Ancestry Informative Markers were used to estimate individual genetic ancestry of the study samples (18). Genotyping was performed at the University of Minnesota Genomics Center using Sequenom. Single nucleotide polymorphisms with call rate <90% or deviated from Hardy–Weinberg equilibrium were removed from the analysis, leaving 80 single nucleotide polymorphisms for ancestry estimation. A total of 283 cases were genotyped, but only 251 cases remained after excluding samples with genotype call rates <85%. We genotyped 63 duplicate pairs and the overall discordance rate was 0.04. The software STRUCTURE version 2.222 (19) was used under an admixture model fixing the number of ancestral components to $k = 3$ to estimate Indigenous American, European and African proportions for each of the samples. We used a burn-in period of 10000 iterations followed by 50000 iterations. Parental populations that include 42 Europeans (Coriell's North American Caucasian panel), 37 West Africans (non-admixed Africans living in London, UK and South Carolina) and 30 Indigenous Americans (15 Mayans and 15 Nahuas) (18) were included to perform a supervised analysis of our samples.

Statistical analysis

All the statistical analyses were performed using R project (www.r-project.org) and SPSS Inc. (Released 2007; SPSS for Windows, Version 16.0. Chicago, IL, USA). Clinicopathological variables according to breast cancer intrinsic subtypes were studied using frequency and percentage tables. Differences in clinicopathological characteristics among breast cancer subtypes were analyzed using X^2 test and a P value < 0.05 was considered statistically significant. We compared mean ancestry proportions between intrinsic subtypes of breast cancer and between geographic regions using analysis of variance tests.

To assess the association between breast cancer intrinsic subtype risk and genetic ancestry, we used unadjusted and adjusted multinomial logistic regression models. African ancestry was modeled as a continuous variable. The adjusted model included as a covariate the recruitment institution (INC and HUC).

Results

The characteristics of 301 patients included in this study are described in Table 2. The tumors were invasive ductal carcinomas in

Table 1. Breast cancer intrinsic subtypes defined by the expression of ER, PR, HER2, Ki67, CK5/6 and EGFR according to the recommendations of 2013 St. Gallen consensus

Intrinsic subtype	Clinicopathologic surrogate definition
Luminal A	*All of*
	ER positive
	PgR positive (≥20%)
	Ki67 low expression (<20%)
Luminal B	'Luminal B–like (HER2 negative)'
	ER positive
	HER2 negative
	And *at least one of*
	PgR positive (<20%)
	Ki67 high expression (≥20%)
	'Luminal B–like (HER2 positive)'
	ER positive
	HER2 overexpressed or amplified
	Any Ki67
	Any PgR
HER2 enriched	HER2 overexpressed or amplified
	ER and PgR negative
Basal like	ER and PgR negative
	HER2 negative
	CK5/6 and/or EGFR positive
NBTN	ER and PgR negative
	HER2 negative
	CK5/6 and/or EGFR negative

92.7% of the cases, lobular in 2.3% and other histological tumor subtypes in 5% of the patients. Most of the patients had a mastectomy (51.5%), did not present metastases at diagnosis (86.4%) and received radiotherapy (76.7%).

Luminal B is the most common subtype in Colombia when St. Gallen 2013 surrogates were applied

We compared the frequency of breast cancer intrinsic subtypes in a sample of Colombian women using different available surrogates for molecular classification (Figure 1). Based on the simplest classification (expression of hormone receptors and HER2), luminal A subtype was the most frequent (52.5%) followed by luminal B (16%). Using the surrogates proposed by the 2011 St. Gallen panel consensus (20), we observed that although luminal A was still the most frequent subtype (36.2%), there was an enrichment of the luminal B subtype (30.2%). Finally, using the most current surrogates proposed by the 2013 St. Gallen panel (11,21), which in addition to Ki67 includes the evaluation of the expression of PgR, the luminal B subtype was found to be the most prevalent in our population (37.2%), followed by luminal A (26.2%), non-basal triple negative (NBTN, 11.6%), basal like (9%) and HER2 enriched (8.6%). Nearly 7% of our cases were excluded from the study because they were not classifiable due to missing data.

Breast cancer intrinsic subtypes and clinicopathological variables

The geographic origins of patients and the relative prevalence of intrinsic breast cancer subtypes are summarized in Table 3. Patients from Coastal regions presented more NBTN and HER2-enriched and basal-like tumors (18.9%, 16.2% and 13.5%, respectively) compared with those from Andean Region (9.8%, 5% and 7.8%, respectively). Patients from Andean region were more probably to present luminal B and A tumors (43.1% and 26.9%, respectively).

The clinicopathological characteristics of the tumors are summarized in Table 4. We found statistically significant differences in the distribution of intrinsic subtypes according to age at diagnosis ($P = 0.0139$), grade ($P < 0.0001$) and recurrence ($P < 0.001$).

The average age at diagnosis was 56.6 years (range 26–94). Women with basal-like and NBTN tumors were younger (51.6. and 51.5 years of age, respectively) than women with luminal and HER2-enriched tumors ($P = 0.0139$). Only luminal subtypes included histological grade 1 tumors. Histological grade 3 was found, in order, in NBTN, basal-like, HER2-enriched, luminal B and A tumors (62.9%, 59.3%, 50%, 25% and 6.3%, respectively).

Women with luminal subtypes were less probably to have recurrences compared with the other intrinsic subtypes. Systemic recurrences were more frequent in women with HER2-enriched tumors followed by basal-like tumors (19.2% and 14.8%, respectively). Patients with luminal B tumors developed

Table 2. Characteristics of the patients

Variables	*n* (%)
Age groups	
<55	126 (41.8)
≥55	154 (51.1)
Unknown	21 (7)
Histopathology	
Invasive ductal carcinoma	279 (92.7)
Invasive lobular carcinoma	7 (2.3)
Other	15 (5)
Metastasis at diagnosis	
Positive	5 (1.6)
Negative	260 (86.4)
Unknown	36 (12)
Surgery	
Mastectomy	155 (51.5)
Quadrantectomy	123 (40.9)
Unknown	23 (7.6)
Bilaterality	
Positive	3 (1)
Negative	274 (91)
Unknown	24 (8)
Estrogen receptor expression	
Positive	208 (69.1)
Negative	93 (30.9)
PgR expression	
Positive	184 (61.1)
Negative	117 (38.9)
HER2 expression	
Positive	77 (25.6)
Negative	220 (73.1)
Unknown	4 (1.3)
Ki67 expression	
<20	186 (61.8)
≥20	106 (35.2)
Unknown	9 (3)
CK5/6 expression	
Positive	40 (13.3)
Negative	260 (86.4)
Unknown	1 (0.3)
EGFR expression	
Positive	40 (13.3)
Negative	259 (86)
Unknown	2 (0.6)
Radiotherapy	
Positive	231 (76.7)
Negative	34 (11.3)
Unknown	36 (12)

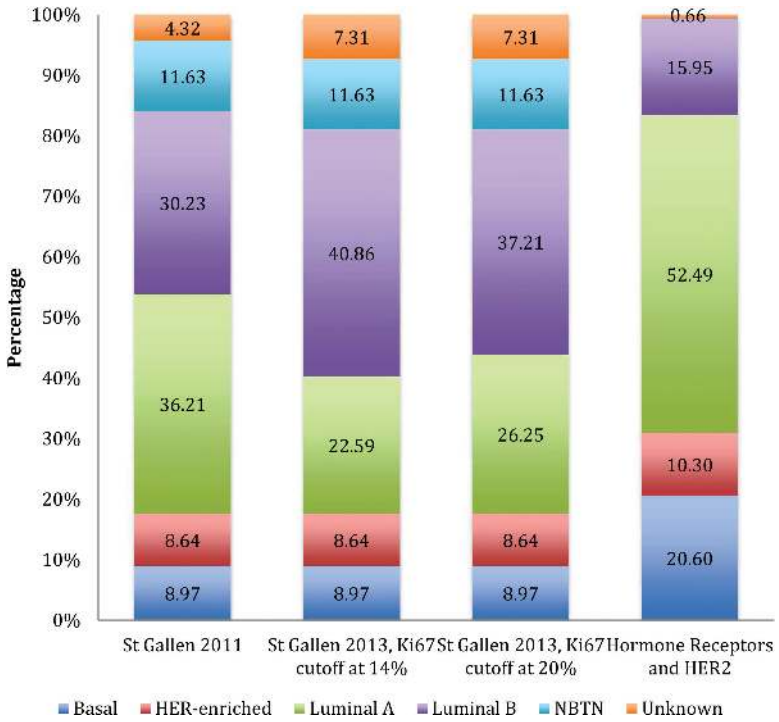

Figure 1. Frequency of breast cancer molecular subtypes in 301 patients according to different proposed classification. Different surrogates were used to compare the frequency of breast cancer intrinsic subtypes. Each bar represents the distribution of intrinsic subtypes according to the classification proposed.

Table 3. Distribution of breast cancer intrinsic subtypes among geographic regions from Colombia

	Luminal A	Luminal B	HER enriched	Basal	NBTN	Unknown	*P* value
	n (%)	*n* (%)	*n* (%)	*n* (%)	*n* (%)	*n* (%)	
Colombia region							0.007
Andean (*n* = 204)	55 (26.9)	88 (43.1)	10 (5)	16 (7.8)	20 (9.8)	15 (7.3)	
Coastal (*n* = 37)	7 (18.9)	7 (18.9)	6 (16.2)	5 (13.5)	7 (18.9)	5 (13.5)	
Plain (*n* = 18)	4 (22.2)	4 (22.2)	5 (27.8)	1 (5.5)	4 (22.2)	0 (0)	
Unknown (*n* = 42)	13 (30.9)	13 (30.9)	5 (11.9)	5 (11.9)	4 (9.5)	2 (4.8)	

more systemic recurrences than those with luminal A tumors (8.9% versus 7.6% respectively).

Other clinicopathological variables such as node status, surgical margins, clinical tumor stage and tumor size did not differ by intrinsic subtype. HER2-enriched subtype showed the highest fraction of positive margins (30.8%) whereas luminal A showed the lowest percentage (10.7%). Regarding clinical tumor stage at diagnosis, as defined by the American Joint Committee of Cancer (AJCC), women with luminal A and B tumors showed the highest percentage of stage I disease (8.9% and 9.8%, respectively). Higher percentage of stage III tumors were found in patients with NBTN, luminal B, basal-like and HER2-enriched subtypes (60%, 48.2%, 48.1% and 42.3%, respectively). Tumor size ranged from 1 to 20 centimeters (cm) with an average of 4.2 cm. Most of the cases (53.2%) presented tumor size between 2 and 5 cm independently of the intrinsic subtype.

We did not find any differences regarding administration or response to neoadjuvant and adjuvant chemotherapy between the different subtypes (Supplementary Table 1, available at *Carcinogenesis* Online). The administration of neoadjuvant chemotherapy did not change the distribution of breast cancer intrinsic subtypes significantly (Supplementary Table 2, available at *Carcinogenesis* Online).

Genetic ancestry in breast cancer patients from Colombia

In the present sample of Colombian women with breast cancer, average African ancestry was estimated to be 9% (range 0–87%), Indigenous American 38% (range 0–92%) and European 53% (range 0–92%) (Figure 2). We found statistically significant differences in the ancestry fractions between the geographic regions in Colombia ($P < 0.0001$) (Table 5). The Andean region showed the highest fraction of European ancestry (0.56) whereas the highest fraction of African ancestry was found in Coastal regions (0.32). Plains showed the highest fraction of Indigenous American ancestry (0.45).

To test differences in ancestry fractions among intrinsic subtypes, we combined luminal A and B tumors into a single 'luminal' category and NBTN and basal-like tumors into a 'triple negative' category. We excluded from the analysis cases with 'unknown' intrinsic subtype. There were differences in the proportion of African ancestry between the intrinsic subtypes ($P = 0.02$) when we analyzed the patients from both institutions together. However, when we analyzed the association between genetic ancestry and intrinsic subtype separately by institution, we did not find differences. Patients from HUC showed high fraction of African ancestry (0.34) comparable with patients

Table 4. Clinical and pathological characteristics of breast cancer patients from Colombian population according to breast cancer subtype based on 2013 St. Gallen consensus

	Luminal A (*n* = 79)	Luminal B (*n* = 112)	HER enriched (*n* = 26)	Basal (*n* = 27)	NBTN (*n* = 35)	Unknown (*n* = 22)	Total (*N* = 301)	*P* value
	N (%)	*N* (%)	*N* (%)	*N* (%)	*N* (%)	*N* (%)	*N* (%)	
Mean age at diagnosis	58.7 ± 11.9	57.9 ± 11.2	57.2 ± 12.8	51.6 ± 14	51.5 ± 9.8	54.4 ± 13.1	56.6 ± 11.9	0.0139
Node status								
Positive	42 (53.2)	58 (51.8)	15 (57.7)	18 (66.7)	23 (65.7)	6 (27.3)	162 (53.8)	
Negative	35 (44.3)	46 (41.1)	7 (26.9)	6 (22.2)	8 (22.9)	11 (50.0)	113 (37.5)	0.014
Unknown	2 (2.5)	8 (7.1)	4 (15.4)	3 (11.1)	4 (11.4)	5 (22.7)	26 (8.6)	
Surgical margins								0.439
Positive	11 (13.9)	12 (10.7)	8 (30.8)	4 (14.8)	6 (17.1)	5 (22.7)	46 (15.3)	
Negative	61 (77.2)	87 (77.7)	17 (65.4)	19 (70.4)	27 (77.1)	15 (68.2)	226 (75.1)	
Unknown	7 (8.9)	13 (11.6)	1 (3.8)	4 (14.8)	2 (5.7)	2 (9.1)	29 (9.6)	
AJCC stage								0.079
Stage I	7 (8.9)	11 (9.8)	1 (3.8)	1 (3.7)	0 (0)	2 (9.1)	22 (7.3)	
Stage IIA/IIB	41 (51.9)	39 (34.8)	9 (34.6)	9 (33.3)	10 (28.6)	9 (40.9)	117 (38.9)	
Stage IIIA/IIIB/IIIC	27 (34.2)	54 (48.2)	11 (42.3)	13 (48.1)	21 (60)	7 (31.8)	133 (44.2)	
StageIV	2 (2.5)	3 (2.7)	1 (3.8)	0 (0)	0 (0)	0 (0)	6 (2)	
Unknown	2 (2.5)	5 (4.5)	4 (15.4)	4 (14.8)	4 (11.4)	4 (18.2)	23 (7.6)	
Tumor size (cm)							4.2 ± 2.8	0.089
<2	11 (13.9)	15 (13.4)	0 (0)	3 (11.1)	2 (5.7)	4 (18.2)	35 (11.6)	
2–5	49 (62)	60 (53.6)	14 (53.8)	10 (37)	18 (51.4)	9 (40.9)	160 (53.2)	
>5	13 (16.5)	23 (20.5)	6 (23.1)	8 (29.6)	10 (28.6)	2 (9.1)	62 (20.6)	
Unknown	6 (7.6)	14 (12.5)	6 (23.1)	6 (22.2)	5 (14.3)	7 (31.8)	44 (16.6)	
Grade								<0.001
1	7 (8.9)	9 (8.0)	0 (0)	0 (0)	0 (0)	1 (4.5)	17 (5.6)	
2	58 (73.4)	66 (58.9)	5 (19.2)	6 (22.2)	8 (22.9)	13 (59.1)	156 (51.8)	
3	5 (6.3)	28 (25)	13 (50)	16 (59.3)	22 (62.9)	5 (22.7)	89 (29.6)	
NA	5 (6.3)	3 (2.7)	0 (0)	1 (3.7)	1 (2.9)	0 (0)	10 (3.3)	
Unknown	4 (5.1)	6 (5.4)	8 (30.8)	4 (14.8)	4 (11.4)	3 (13.6)	29 (9.6)	
Recurrence								< 0.001
Local	0 (0)	0 (0)	0 (0)	1 (3.7)	0 (0)	0 (0)	1 (0.3)	
Local and systemic	3 (3.8)	1 (0.9)	0 (0)	4 (14.8)	1 (2.9)	0 (0)	9 (3.0)	
No recurrences	66 (83.6)	87 (77.7)	11 (42.3)	15 (55.6)	26 (74.3)	15 (68.2)	220 (73.1)	
Regional	0 (0)	1 (0.9)	2 (7.7)	0 (0)	1 (2.9)	0 (0)	4 (1.3)	
Regional and systemic	0 (0)	0 (0)	0 (0)	0 (0)	1 (2.9)	0 (0)	1 (0.3)	
Systemic	6 (7.6)	10 (8.9)	5 (19.2)	4 (14.8)	2 (5.7)	2 (9.1)	29 (9.6)	
Unknown	4 (5.1)	13 (11.6)	8 (30.8)	3 (11.1)	4 (11.4)	5 (22.7)	37 (12.3)	

NA, not applicable.

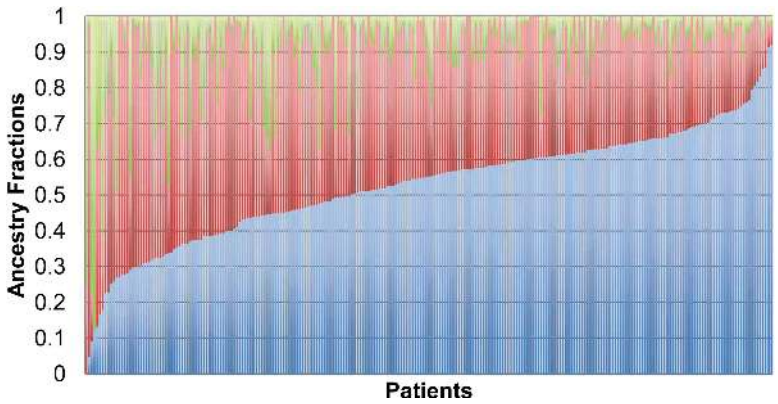

Figure 2. Distribution of genetic ancestry among 251 breast cancer patients from Colombia. Each patient is represented by a vertical bar in the *x*-axis. Bars represent percentage of European (blue), Indigenous American (red) and African genetic ancestry (green).

Table 5. Average of genetic ancestry according to geographic origin of patient and molecular subtype of breast cancer in patients from both institutions included in the study (INC and HUC), analyzed together and separately

	African	*P* value	European	*P* value	Indigenous American	*P* value
Natural region from Colombia (*n* = 215)		<2E-16		3.59E-07		1.98E-06
Andean (*n* = 167)	0.05 ± 0.05		0.56 ± 0.13		0.39 ± 0.13	
Coastal (*n* = 31)	0.32 ± 0.17		0.41 ± 0.17		0.27 ± 0.09	
Plains (*n* = 17)	0.07 ± 0.08		0.48 ± 0.18		0.45 ± 0.19	
Molecular subtypes of breast cancer in all patients (*n* = 232)	0.09 ± 0.13	0.0211	0.53 ± 0.15	0.584	0.38 ± 0.14	0.158
Luminal (*n* = 160)	0.07 ± 0.11		0.53 ± 0.15		0.39 ± 0.14	
HER2 enriched (*n* = 22)	0.14 ± 0.15		0.51 ± 0.16		0.35 ± 0.11	
Triple negative (*n* = 50)	0.12 ± 0.16		0.51 ± 0.16		0.37 ± 0.13	
HUC patients only (*n* = 12)	0.34 ± 0.11	0.327	0.36 ± 0.14	0.556	0.29 ± 0.11	0.571
Luminal (*n* = 3)	0.24 ± 0.07		0.44 ± 0.15		0.32 ± 0.08	
HER2 enriched (*n* = 3)	0.40 ± 0.18		0.29 ± 0.13		0.31 ± 0.14	
Triple negative (*n* = 6)	0.34 ± 0.09		0.34 ± 0.17		0.32 ± 0.14	
INC patients only (*n* = 220)	0.08 ± 0.11	0.551	0.54 ± 0.15	0.686	0.38 ± 0.14	0.475
Luminal (*n* = 157)	0.07 ± 0.10		0.53 ± 0.15		0.39 ± 0.14	
HER2 enriched (*n* = 19)	0.10 ± 0.11		0.54 ± 0.14		0.36 ± 0.10	
Triple negative (*n* = 44)	0.09 ± 0.15		0.53 ± 0.14		0.38 ± 0.13	

from Coastal regions recruited in the INC. Results from multinomial logistic regression unadjusted and adjusted by institution are concordant with the results from the analysis of variance (Supplementary Table 3, available at *Carcinogenesis* Online).

Discussion

Ours is the first study performed in a Colombian population aimed at determining the prevalence of breast cancer intrinsic subtypes and its association with clinicopathological data. We found that the mean age at diagnosis in our study population was 56.6 years, which is earlier than what is reported in non-Hispanic White women and consistent with what has been reported for Hispanic/Latinas (22,23). In addition, patients with luminal subtypes were older compared with those with basal-like and NBTN subtypes. The latter tumors are usually diagnosed at younger ages and more probably to be poorly differentiated with higher histological grade (24–27), which is consistent with our findings.

Using the surrogate criteria recommended by the 2013 St. Gallen panel of experts (11,17,20), we found that luminal B was the most prevalent intrinsic subtype of breast cancer in our population. This suggests that among Colombian patients with luminal tumors, high-risk luminal B tumors are more common than low-risk luminal A tumors. These findings do not support the suggestion by Prat *et al.* (21) that this new classification would enrich for luminal A subtype. In contrast to previous reports (28–30), we did not see an effect of neoadjuvant chemotherapy on IHC markers used in our cohort.

Previous studies in Hispanic/Latinas from the USA that analyzed the expression of markers such as hormone receptors and HER2 have shown that the most prevalent subtype is the luminal A (hormone receptor+/HER2–) followed by triple negative (31–34). Our results are concordant with these studies as we found luminal A as the most frequent subtype (52.5%) followed by triple negative (20.6%), luminal B (16%) and HER2 (10.3) and unknown (0.6) when we group tumors by the expression of these three markers (ER, PgR and HER2).

There are few studies that have used the most recent surrogates to classify breast cancer into intrinsic subtypes; nevertheless, two recent studies found increased prevalence of luminal B subtypes in European (57.1%) and Chinese (68.5%) women, respectively, when breast cancer classification was performed using the recommendations of the 2013 St. Gallen surrogates (35,36). Gomez *et al.* (37) recently analyzed the distribution of breast cancer intrinsic subtypes in 328 clinic-based patients recruited from Medellin, Colombia. They found that luminal B tumor represented more than 50% of the intrinsic subtypes

(luminal B/HER2– 33.5%, luminal B/HER2+ 22.9%). The molecular classification used in this study was based on the St. Gallen 2011 surrogates. When we applied the St. Gallen 2011 classification to the tumors in our study, the frequency of luminal B tumors was 36.21%. These results would support the hypothesis that the Colombian population has a high prevalence of luminal B intrinsic subtype as two independent studies showed similar results. However, we cannot rule out the possibility that the high prevalence of luminal B tumors in Colombians might be due to differences in classification methods between these studies and those conducted in other populations. In addition, there could be interlaboratory variation in evaluation of ER, PR and Ki67 expression (35). Notwithstanding, and consistent with previous finding (38–40), clinicopathological data indicate that our classification, based on St. Gallen consensus, was informative about tumor biology, as we found that luminal B tumors had a higher percentage of systemic recurrence compared with tumors classified as luminal A. Moreover, the high prevalence of luminal B subtype in our Colombian sample when using the 2013 St. Gallen classification appears to be independent of genetic ancestry. However, these findings should be analyzed with caution and not generalized to all Latin American populations since ancestry proportions vary across regions (41–43).

The prevalence of the intrinsic breast cancer subtypes has been reported to differ according to race/ethnicity. The largest study conducted so far evaluated the expression of the 50 genes included in the PAM50 panel in 1319 women and showed that African-Americans are significantly more probably to have basal-like tumors, while Hispanic/Latina patients showed a relatively low prevalence of luminal A tumors comparable with that of African-Americans, and increased rates of luminal B and HER2 tumors (12). In line with these results, our data show that individuals with HER2-enriched and basal-like tumors have the highest fractions of African ancestry. However, these results were no longer statistically significant after adjustment by institution. Patients from HUC have a higher fraction of African ancestry compared with patients from the INC, which is expected given that most of the INC patients are from the more European Andean region. Analyses in a larger sample of patients from the Colombian Coastal region are necessary to confirm the association between African ancestry and tumor subtype in Colombia beyond the particular tumor subtype distribution of a specific institution.

We are aware that a possible limitation of the study is the fact that we assessed intrinsic subtypes by IHC and not by gene expression profile, which is the gold standard for molecular classification. On the other hand, our results support the notion that, in situations when molecular profiling by gene expression is not feasible, the 2013 St. Gallen surrogate classification model is clinically informative and can be used for therapeutic planning. Importantly, although the St. Gallen 2013 criteria produced the highest fraction of luminal B tumors, similar results were obtained using the St. Gallen 2011 criteria (20). Another limitation of the study is the fact that the vast majority of the samples were recruited in a single institution from Colombia. As mentioned before, the INC is a national referral center for cancer and, therefore, it is possible that more aggressive cancers are overrepresented. However, we did not observe disproportionately high prevalence of other clinically aggressive subtypes, such as basal like and HER2 enriched. Our results cannot rule out the possibility that clonal selection by neoadjuvant chemotherapy produced a high fraction of luminal B tumors, but no significant difference was observed between the group of patients who received neoadjuvant chemotherapy and those who did not (Supplementary Table 2, available at *Carcinogenesis* Online).

One advantage of our study is the use of Ancestry Informative Markers, which in a highly admixed population, such as that of Colombia, provide precise identification of the different ancestral components of an individual's genome. The next step will be to investigate the molecular profiles of breast cancer in Colombian women, in order to: (i) determine how these profiles change when compared with those patients classified as luminal B from other populations and (ii) identify candidate genetic factors correlated with luminal B tumors.

Our findings are important as advances in cancer research are enabling personalized treatment of patients according to their molecular profiles. The fact that we found that IHC assignment into intrinsic subtypes is clinically informative in our sample suggests that the incorporation of this panel in routine diagnosis could identify patients with luminal B breast cancer that may need a more aggressive treatment to reduce the likelihood of recurrences. If the high prevalence of luminal B tumors is an intrinsic characteristic of our population, then the development of specific chemotherapeutic regimens for these patients would improve survival.

In summary, our results in Colombian patients showed a high prevalence of high-risk luminal B tumors when St. Gallen 2013 surrogates were applied. We found that the clinicopathological characteristics of breast cancer intrinsic subtypes in Colombian patients are similar to what has been reported in tumors from other populations. Our results indicate that using a surrogate intrinsic subtype classification based on the St. Gallen 2013 criteria rather than just ER, PgR and HER2 markers in Colombian patients identifies more luminal B tumors that are associated with higher recurrence rates and may benefit from more aggressive treatment.

Supplementary material

Supplementary Tables 1–3 can be found at http://carcin.oxfordjournals.org/

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