Received: 5 May 2017 | Accepted: 24 June 2017
DOI: 10.1002/ajmg.a.38362

ORIGINAL ARTICLE

WILEY AMERICAN JOURNAL OF medical genetics PART A

Noonan syndrome in diverse populations

Paul Kruszka[1] | Antonio R. Porras[2] | Yonit A. Addissie[1] | Angélica Moresco[3] | Sofia Medrano[3] | Gary T. K. Mok[4] | Gordon K. C. Leung[4] | Cedrik Tekendo-Ngongang[5] | Annette Uwineza[6] | Meow-Keong Thong[7] | Premala Muthukumarasamy[7] | Engela Honey[8] | Ekanem N. Ekure[9] | Ogochukwu J. Sokunbi[9] | Nnenna Kalu[9] | Kelly L. Jones[10] | Julie D. Kaplan[10] | Omar A. Abdul-Rahman[10] | Lisa M. Vincent[11] | Amber Love[11] | Khadija Belhassan[1,12] | Karim Ouldim[12] | Ihssane El Bouchikhi[12,13] | Anju Shukla[14] | Katta M. Girisha[14] | Siddaramappa J. Patil[15] | Nirmala D. Sirisena[16] | Vajira H. W. Dissanayake[16] | C. Sampath Paththinige[16] | Rupesh Mishra[16] | Eva Klein-Zighelboim[17] | Bertha E. Gallardo Jugo[17] | Miguel Chávez Pastor[17] | Hugo H. Abarca-Barriga[17] | Steven A. Skinner[18] | Eloise J. Prijoles[18] | Eben Badoe[19] | Ashleigh D. Gill[1] | Vorasuk Shotelersuk[20] | Patroula Smpokou[21] | Monisha S. Kisling[21] | Carlos R. Ferreira[21] | Leon Mutesa[6] | Andre Megarbane[22] | Antonie D. Kline[23] | Amy Kimball[23] | Emmy Okello[24] | Peter Lwabi[24] | Twalib Aliku[24] | Emmanuel Tenywa[24,25] | Nonglak Boonchooduang[26] | Pranoot Tanpaiboon[21] | Antonio Richieri-Costa[27] | Ambroise Wonkam[5] | Brian H. Y. Chung[4] | Roger E. Stevenson[18] | Marshall Summar[21] | Kausik Mandal[28] | Shubha R. Phadke[28] | María G. Obregon[3] | Marius G. Linguraru[2] | Maximilian Muenke[1]

[1] Medical Genetics Branch, National Human Genome Research Institute, The National Institutes of Health, Bethesda, Maryland
[2] Children's National Health System, Sheikh Zayed Institute for Pediatric Surgical Innovation, Washington, District of Columbia
[3] Servicio de Genética, Hospital de Pediatría Garrahan, Buenos Aires, Argentina
[4] LKS Faculty of Medicine, Department of Paediatrics and Adolescent Medicine, The University of Hong Kong, Hong Kong Special Administrative Region, Hong Kong, China
[5] Division of Human Genetics, University of Cape Town, Cape Town, South Africa
[6] Center of Human Genetics, School of Medicine and Pharmacy, College of Medicine and Pharmacy, University of Rwanda, Kigali, Rwanda
[7] Faculty of Medicine,Department of Paediatrics, University of Malaya, Kuala Lumpur, Malaysia
[8] Department of Genetics, University of Pretoria, Pretoria, South Africa
[9] Department of Paediatrics College of Medicine, University of Lagos, Lagos University Teaching Hospital, Lagos, Nigeria
[10] Division of Medical Genetics, Department of Pediatrics, University of Mississippi Medical Center, Jackson, Mississippi
[11] GeneDx, Gaithersburg, Maryland
[12] Medical Genetics and Oncogenetics Unit, Hassan II University Hospital, Fez, Morocco
[13] Faculty of Sciences and Techniques,Laboratory of Microbial Biotechnology, University of Sidi Mohammed Ben Abdellah, Fez, Morocco
[14] Department of Medical Genetics, Kasturba Medical College, Manipal University, Manipal, India
[15] Mazumdar Shaw Medical Center, Narayana Health City, Bangalore, India

 © 2017 Wiley Periodicals, Inc.

[16] Faculty of Medicine, Human Genetics Unit, University of Colombo, Colombo, Sri Lanka
[17] Instituto Nacional de Salud del Niño, Lima, Peru
[18] Greenwood Genetic Center, Greenwood, South Carolina
[19] School of Medicine and Dentistry,Department of Child Health, College of Health Sciences, Accra, Ghana
[20] Faculty of Medicine,Center of Excellence for Medical Genetics, Department of Pediatrics, Chulalongkorn University, Bangkok, Thailand
[21] Division of Genetics and Metabolism, Children's National Health System, Washington, District of Columbia
[22] Institut Jérôme Lejeune, Paris, France
[23] Harvey Institute for Human Genetics, Greater Baltimore Medical Center, Baltimore, Maryland
[24] Uganda Heart Institute, Kampala, Uganda
[25] Jinja Regional Referral Hospital, Jinja, Uganda
[26] Division of Developmental and Behavioral Pediatrics, Department of Pediatrics, Chiangmai University, Chiang Mai, Thailand
[27] Hospital for the Rehabilitation of Craniofacial Anomalies, São Paulo University, Bauru, Brazil
[28] Department of Medical Genetics, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow, Uttar Pradesh, India

Correspondence
Paul Kruszka and Maximilian Muenke, Medical Genetics Branch, National Human Genome Research Institute, The National Institutes of Health, Bethesda, MD.
Email: paul.kruszka@nih.gov (PK); mamuenke@mail.nih.gov (MM)

Funding information
National Human Genome Research Institute, Grant number: Division of Intramural Research; Government of Abu Dhabi, Grant number: philanthropic gift

Noonan syndrome (NS) is a common genetic syndrome associated with gain of function variants in genes in the Ras/MAPK pathway. The phenotype of NS has been well characterized in populations of European descent with less attention given to other groups. In this study, individuals from diverse populations with NS were evaluated clinically and by facial analysis technology. Clinical data and images from 125 individuals with NS were obtained from 20 countries with an average age of 8 years and female composition of 46%. Individuals were grouped into categories of African descent (African), Asian, Latin American, and additional/other. Across these different population groups, NS was phenotypically similar with only 2 of 21 clinical elements showing a statistically significant difference. The most common clinical characteristics found in all population groups included widely spaced eyes and low-set ears in 80% or greater of participants, short stature in more than 70%, and pulmonary stenosis in roughly half of study individuals. Using facial analysis technology, we compared 161 Caucasian, African, Asian, and Latin American individuals with NS with 161 gender and age matched controls and found that sensitivity was equal to or greater than 94% for all groups, and specificity was equal to or greater than 90%. In summary, we present consistent clinical findings from global populations with NS and additionally demonstrate how facial analysis technology can support clinicians in making accurate NS diagnoses. This work will assist in earlier detection and in increasing recognition of NS throughout the world.

KEYWORDS
Africa, Asia, diverse populations, facial analysis technology, Latin America, Middle East, Noonan syndrome

1 | INTRODUCTION

Noonan syndrome (NS) is characterized by congenital heart disease, short stature, distinctive facial features, chest deformities, variable developmental delay, and other anomalies (Bhambhani & Muenke, 2014; Noonan, 1968). Diagnostic criteria have been established (van der Burgt et al., 1994) as well as management guidelines (Roberts, Allanson, Tartaglia, & Gelb, 2013; Romano et al., 2010). The typical facial features of NS include widely spaced eyes, down slanted palpebral fissures, ptosis, and low-set ears. The prevalence of NS is roughly 1:1,000 to 1:2,500 and is inherited in an autosomal dominant manner (Romano et al., 2010). Although NS is a common genetic syndrome, there are few phenotype and genotype studies in non-European cohorts.

The genetic etiologies of NS occur in genes associated with the Ras-mitogen-activated protein kinase (Ras/MAPK) pathway. Genes in this pathway are involved in cell differentiation, growth, and death. Other syndromes associated with Ras/MAPK genes include Costello syndrome, Cranio-facio-cutaneous (CFC) syndrome, NS with multiple

lentigines (formerly called LEOPARD syndrome), and neurofibromatosis. Significant phenotypic overlap exists with other RASopathies including CFC syndrome and Costello syndrome. The most common genetic cause of NS is gain-of function mutations in the *PTPN11* gene encoding the Src homology protein-tyrosine phosphatase-2 (SHP-2) with a variant occurring in 50% of individuals with NS (Tartaglia et al., 2001, 2002). *PTPN11* was the first gene to be associated with NS and now there are more than eight known genes (*PTPN11*, *SOS1*, *RAF1*, *RIT1*, *KRAS*, *NRAS*, *BRAF*, *MAP2K1*, *RRAS*, *RASA2*, *A2ML1*, *SOS2*, *LZTR1*) that cause NS (Allanson & Roberts, 1993). *SOS1* is the second most common causative gene with variants occurring in 16–20% of individuals without *PTPN11* variants (Roberts et al., 2007; Tartaglia et al., 2007).

A number of investigators have evaluated *PTPN11* variants in diverse populations. In a Brazilian cohort of 50 individuals with NS, 42% were found to have *PTPN11* variants, but the most common variant, p.N308D, which was found in 31% of a North American cohort (Tartaglia et al., 2002) was not present (Bertola et al., 2006), demonstrating that different variants are found in different populations. In contrast, Lee et al. (2007) found half of a Korean cohort of individuals with NS to have previously reported *PTPN11* variants including 29% of the cohort with the p.N308D variant (Lee, Ki, & Lee, 2007). In another Korean cohort of 59 patients, Ko et al. (2008) found that in 13 patients with *PTPN11* variants, 12 had been previously reported. Although genotypes have been compared, the phenotype in NS has not been contrasted between different countries and populations. However, investigators in some countries have defined regional height phenotypes in NS and have developed growth charts specific to their country including Japan (Isojima et al., 2016) and Brazil (Malaquias et al., 2012).

NS can be a difficult diagnosis to make as the phenotype is variable and changes with age (Allanson, Hall, Hughes, Preus, & Witt, 1985; van der Burgt et al., 1999). Comprehensive characterization of NS in diverse populations has not yet been done in the medical literature. In this report, we use images, subjective examination data, and facial analysis technology to describe NS in diverse populations.

2 | METHODS

2.1 | Review of medical literature

Studies that characterize NS in diverse populations were obtained from a Medline search. The search terms used included: NS, Africa, Asia, Latin America, Middle East, diverse populations, and facial analysis technology. Further studies were found using reference lists of journal studies. After obtaining journal permissions, photos of individuals with NS were used to supplement study participants described below (Addissie et al., 2015; Aoki et al., 2013; Edwards et al., 2014; Lee & Sakhalkar, 2014; Ndiaye et al., 2014; Yaoita et al., 2016).

2.2 | Patients

Individuals with NS were evaluated from 20 countries. All participants (Supplementary Table S1) had NS diagnosed by both clinical evaluation and/or molecular diagnosis. The patients were grouped by geographic area of origin or ethnicity (African and African American, Asian, Latin American, and Additional). Local clinical geneticists examined patients for established clinical features found in NS (van der Burgt et al., 1994).

Consent was obtained by local institutional review boards and the Personalized Genomics protocol at the National Institutes of Health (11-HG-0093). Exam findings from the current study and those from the medical literature are recorded in Table 1.

2.3 | Facial analysis technology

As previously described (Kruszka, Addissie, et al., 2017; Kruszka, Porras, et al., 2017), digital facial analysis technology (Cerrolaza et al., 2016; Zhao et al., 2013; Zhao, Okada, et al., 2014; Zhao, Werghi et al., 2014) was used to evaluate 161 images of individuals with NS, and 161 ethnic, gender, and age matched controls from our previously described database (Zhao, Okada, et al., 2014; Zhao et al., 2013; Zhao, Werghi, et al., 2014). The 161 individuals with NS used for facial analysis technology included individuals from Supplementary Table S1 and additional archival images of individuals with NS. A Caucasian ethnic group was identified in addition to African, Asian, and Latin American for the purpose of facial analysis. The distribution of the dataset is presented in Table 2. Only frontal images were analyzed by this technology.

Our algorithms analyzed the images of our study participants with output variables consisting of feature extraction, feature selection, and classification. As in our previous studies (Kruszka, Addissie, et al., 2017; Kruszka, Porras, et al., 2017), from a set of 44 landmarks placed on the frontal face images, a total of 126 facial features, including both geometric and texture biomarkers, were isolated. The geometric biomarkers consisted of a set of distances and angles calculated between the different inner facial landmarks. Figure 1 represents both the landmark locations and the geometric features extracted. Texture patterns (Cerrolaza et al., 2016) were calculated at each of the acial landmarks to quantify texture information (Figure 1). From the collection of geometric and texture features, the most significant ones were selected using the method proposed previously (Cai, Zhang, & He, 2010). For each feature set, a support vector machine classifier (Cortes & Vapnik, 1995) was trained using a leave-one-out cross-validation strategy (Elisseeff & Pontil, 2003). The optimal number of features was selected as the minimum number for which the classification accuracy converged to its maximum; Supplementary Figures S1–S5 graphically demonstrate how the addition of features improves the measures of sensitivity, specificity, and accuracy. As an estimator of the individual discriminant power of each feature selected, the *p*-value of each feature was also estimated using the Student's *t*-test. Significance between methods used to detect NS was assessed using Fisher's exact test.

3 | RESULTS

Clinical information and images were collected on 125 individuals (13 individuals were obtained from the medical literature) from 20

TABLE 1 Summary of exam findings of individuals with Noonan syndrome from diverse backgrounds including 99 unpublished individuals from present study and 370 individuals from the medical literature (Bertola et al., 2006; Essawi et al., 2013; Hung et al., 2007; Jongmans et al., 2005; Ko et al., 2008; Lee et al., 2011; Lee et al., 2007; Ndiaye et al., 2014; Simsek-Kiper et al., 2013; Yoshida et al., 2004)

	Present study				Hung et al. (2007)	Ndiaye et al. (2014)	Bertola et al. (2006)	Yoshida et al. (2004)	Lee et al. (2007)	Ko et al. (2008)	Simsek-Kiper et al. (2013)	Essawi et al. (2013)	Jongmans et al. (2005)	Lee et al. (2011)
	Africa, *n* = 30	Asia, *n* = 36	Latin America, *n* = 33	*p*-values	Taiwan, *n* = 34	Senegal, *n* = 6	Brazil, *n* = 50	Japan, *n* = 45	Korea, n = 14	Korea, *n* = 59	Turkey, *n* = 26	Egypt, *n* = 21	The Netherlands, *n* = 56	Korea, *n* = 59
Average age (years)	9.3	8.6	9.9		9.06	12	10	14.8	8.8	3.7				
Age range (years)	0.33–30	0.17–31	0.17–31			1–31	0.3–24.1	0.1–34.5	2–29	0.1–17.2	0.25–29	2–20		
PTPN11 (%)	6/9 (67)	22/26 (85)	17/21 (81)		13 (38)	2 (33)	21 (42)	18 (40)	7 (50)	16 (27)	7 (27)		56 (100)	23 (39)
SOS1 (%)		2/26 (8)	2/21 (10)							10 (17)	5 (19)			12 (20)
SHOC2 (%)			0								3 (12)			
Widely spaced eyes	80%	96%	94%	0.10		6 (100%)	22 (44%)	45 (100%)			22 (85%)	21 (100%)		
Ptosis	63%	72%	94%	0.011	12 (35%)						25 (96%)	6 (29%)		
Downslanted palpebral fissures	87%	86%	73%	0.25	20 (59%)		33 (66%)				19 (73%)	21 (100%)		
Epicanthal folds	70%	64%	55%	0.44	19 (56%)									
Low-set ears	82%	94%	88%	0.30		1 (17%)					15 (58%)	12 (57%)		
Lowset posterior hairline	64%	76%	69%	0.57	25 (74%)									
Webbed neck	57%	36%	69%	0.023	21 (62%)	3 (50%)	46 (92%)		10 (71%)		16 (62%)	21 (100%)	10 (18%)	48.30%
Pulmonary stenosis	50%	53%	48%	0.94	12 (35%)	2 (33%)	28 (56%)	16 (36%)	8 (57%)	24 (41%)	16 (62%)	5 (24%)	38(68%)	22/43 (51%)
Hypertrophic cardiomyopathy	7%	11%	10%	0.82		3 (50%)	6 (12%)	5 (11%)		13 (22%)	2 (8%)	4 (19%)	4 (7%)	11/43 (26%)
ASD	27%	14%	24%	0.39				14 (31%)	6 (43%)	19 (32%)	11 (42%)		17 (30%)	12/43 (28%)
VSD	7%	17%	6%	0.26					2 (14%)	13 (22%)	3 (12%)			7/43 (16%)

(Continues)

TABLE 1 (Continued)

	Present study				Hung et al. (2007)	Ndiaye et al. (2014)	Bertola et al. (2006)	Yoshida et al. (2004)	Lee et al. (2007)	Ko et al. (2008)	Simsek-Kiper et al. (2013)	Essawi et al. (2013)	Jongmans et al. (2005)	Lee et al. (2011)
	Africa, *n* = 30	Asia, *n* = 36	Latin America, *n* = 33	*p*-values	Taiwan, *n* = 34	Senegal, *n* = 6	Brazil, *n* = 50	Japan, *n* = 45	Korea, n = 14	Korea, *n* = 59	Turkey, *n* = 26	Egypt, *n* = 21	The Netherlands, *n* = 56	Korea, *n* = 59
Septal defects	37%	36%	27%	0.66	16 (47%)				11 (79%)		14/26 (54%)			
PDA (%)	0	8	0							7 (12)				7/43 (16)
Short stature (<3rd centile%)	71%	80%	83%	0.53	32 (94%)	6 (100%)	48 (96%)		9 (64%)	30 (51%)	14/26 (54%)	15 (71%)	41 (73%)	27/41 (66%)
Chest deformity	59%	49%	70%	0.21	13 (38%)	4 (67%)	28 (56%)	9 (20%)	9 (64%)	14 (24%)		17 (81%)	24 (43%)	16/43 (37%)
Scoliosis	37%	17%	13%	0.08			9/47 (19%)							
Undescended testes	47%	26%	47%	0.17		1/2 (50%)	14/27 (52%)	9/25 (36%)	4/9 (44%)	19/41 (46%)		4/12 (33%)	27/32 (84%)	11/29 (38%)
Coagulopathy	26%	14%	6%	0.12			20/48 (42%)	5 (11%)		3 (5.1%)			32 (57%)	4/43 (9%)
Learning disability/ intellectual disability	64%	54%	63%	0.69	19 (56%)			11/43 (26%)			12 (46%)	11 (52%)	28 (50%)	11/40 (28%)
Renal anomalies	11%	17%	10%	0.75			7/50 (14%)							
Skin pigmentation lesions	12%	38%	27%	0.12										

TABLE 2 Population data used in facial analysis technology which includes 161 individuals with Noonan syndrome from Supplementary Table S1 and additional archival images of individuals with Noonan syndrome

	Noonan syndrome (*N* = 161)		Controls (*N* = 161)	
Age	Number	%	Number	%
Newborn	0	0	0	0
Infant	45	28	45	28
Toddler	29	18	29	18
Child	47	29	47	29
Adolescence	18	11	18	11
Adult	22	14	22	14
Total	161		161	
Ethnicity				
African Descent	35	22	35	22
Asian	40	25	40	25
Caucasian	40	25	40	25
Latino	46	29	46	29
Total	161		161	
Gender				
Male	93	58	93	58
Female	68	42	68	42
Total	161		161	

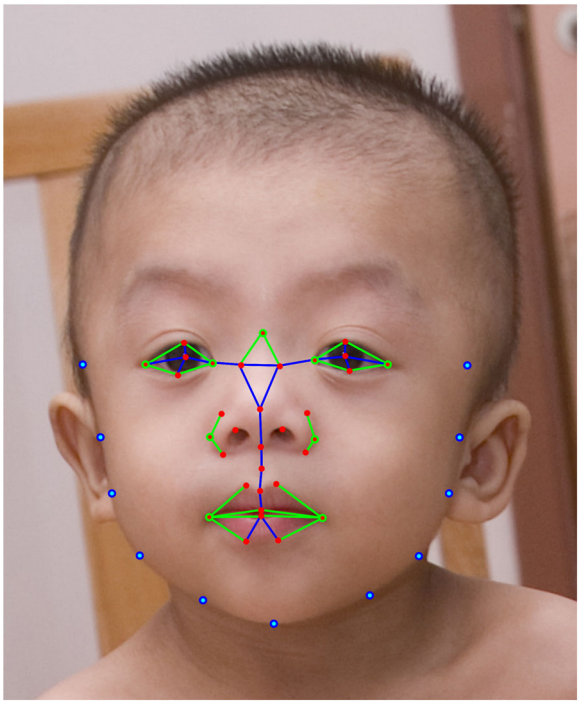

FIGURE 1 Facial landmarks on a Noonan syndrome patient. Inner facial landmarks are represented in red, while external landmarks are represented in blue. Blue lines indicate the calculated distances. Green circles represent the corners of the calculated angles. Texture features are extracted only from the inner facial landmarks

countries, average age was 8 years, the median age was 5 years, and 46% were females (Supplementary Table S1). Individuals of African descent are shown in Figure 2 (facial), Figure 3 (chronological sequence images), and Figure 4 (facial and torso profiles); Asian in Figure 5 (facial), Figure 3 (chronological sequence images), and Figure 6 (facial and torso profiles); Latin American in Figure 7 (facial), Figure 3 (chronological sequence images), and Figure 8 (facial and torso profiles); and additional patients in Supplementary Figure S6. Supplementary Figures S7 and S8 display hand and feet images, respectively. From the medical literature in Table 1, we found 10 non-European descent studies of NS that evaluated at least five participants and at least one facial feature (Bertola et al., 2006; Essawi et al., 2013; Hung et al., 2007; Jongmans et al., 2005; Ko, Kim, Kim, & Yoo, 2008; Lee et al., 2011; Lee et al., 2007; Ndiaye et al., 2014; Simsek-Kiper et al., 2013; Yoshida et al., 2004). We compared unpublished patients from the present study with the above-mentioned studies from the medical literature (Table 1). The most common phenotype element in both the present study and the medical literature is widely spaced eyes. In our study, all population groups had widely spaced eyes in 80% or greater of individuals, and in the medical literature, four of five studies report 85% or more of their cohorts as having widely spaced eyes (Table 1). Low-set ears, also common in our cohort, was found in over 80% of the present study but not consistently reported in the literature. And lastly, short stature as defined by <3rd centile (when centiles were provided) was found in greater than 70% of the present study and in seven of the nine studies in the medical literature. The remainder of clinical exam findings in the present study were consistent between the different population groups; the only exam elements that differed statistically among groups in the present study were ptosis and webbed neck ($p = 0.01$ and $p = 0.02$, respectively; χ^2 test). Consistent with the medical literature (Allanson & Roberts, 1993), this study's most common congenital heart disease was pulmonary stenosis, found in roughly 50% of all three population groups (Table 1).

As a more objective measure of phenotype, but limited to facial features, facial analysis technology was applied to 161 individuals (Caucasian, African, Asian, and Latin American) with results shown in Table 3. The sensitivity and specificity to discriminate between NS and controls was 0.88 and 0.89, respectively, when the entire cohort was evaluated concurrently. The test accuracy of the facial recognition technology increased significantly when the cohort was analyzed by specific ethnic population (p-value < 0.001 for all comparisons), with sensitivities and specificities for Caucasian, African, Asian, and Latin American of 0.95 and 0.93, 0.94 and 0.91, 0.95 and 0.90, and 0.96 and 0.98, respectively (Table 3).

4 | DISCUSSION

We present the first study that evaluates the clinical presentation of NS and uses facial analysis technology in diverse populations. Both

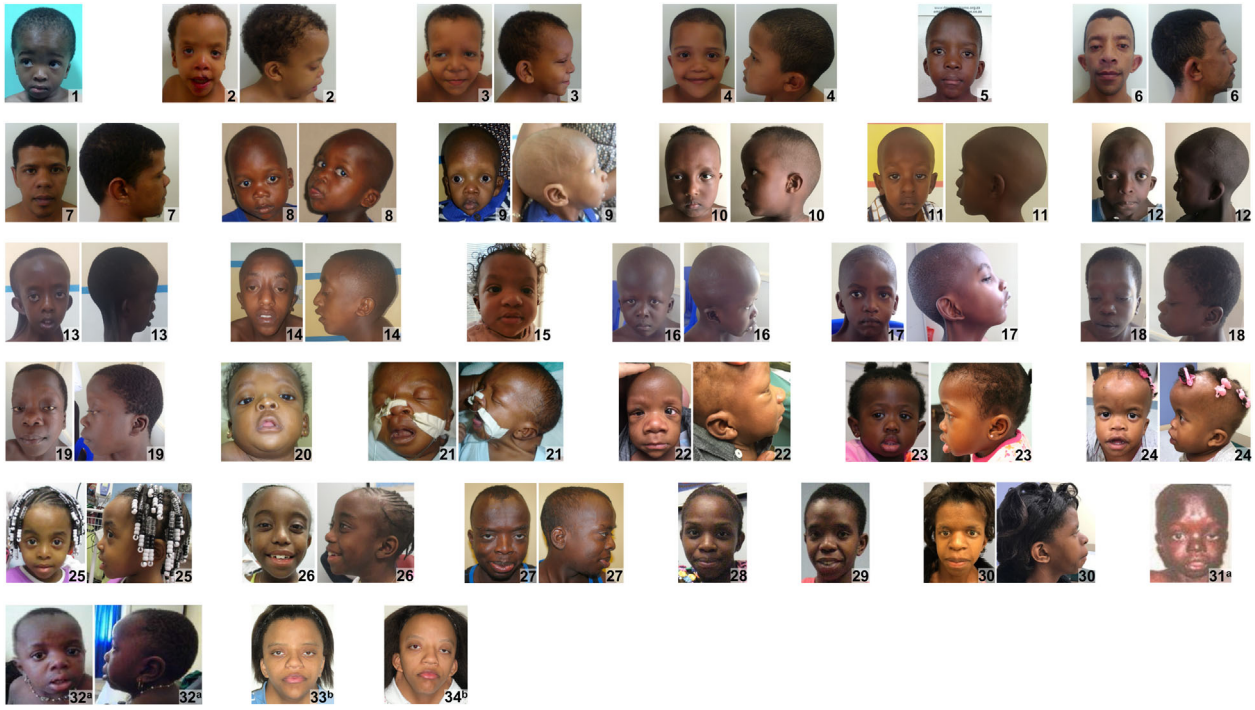

FIGURE 2 Frontal and lateral facial profiles of individuals of African descent with Noonan syndrome. Gender, age, and country of origin found in Supplementary Table S1 [a](Ndiaye et al., 2014), [b](Lee & Sakhalkar, 2014)

clinical diagnostic guidelines (van der Burgt et al., 1994) and facial analysis technology (Hammond et al., 2004) have been reported for the diagnosis of NS cohorts in the past, but not in multiple ethnic population groups. Hammond et al. (2004) used an elaborate combination of deep surface models from three-dimensional scans combined with pattern recognition algorithms to allow for a sensitivity of 88% and specificity of 94% to discriminate between NS and controls. However, that study examined only patients of European descent and is not applicable to facial photographs.

In this study, we demonstrate that the clinical presentation of NS is similar across different population groups. When looking at 21 clinical characteristics (Table 1), only two elements where

FIGURE 3 Sequential photos of individuals with Noonan syndrome at different ages. Gender, age, and country of origin found in Supplementary Table S1

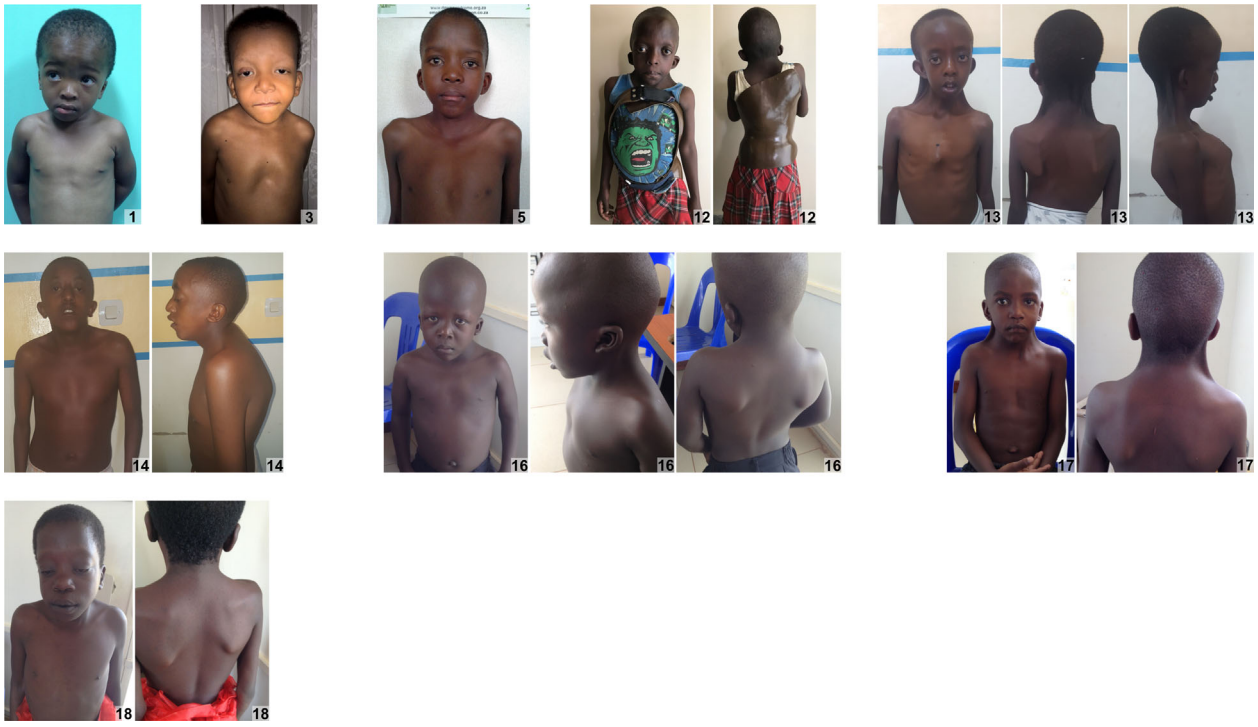

FIGURE 4 Facial and torso profiles of individuals of African descent with Noonan syndrome. Gender, age, and country of origin found in Supplementary Table S1

statistically different between the African, Asian, and Latin American groups: ptosis and webbed neck. Three clinical characteristics in our study were present in over 70% of participants including widely spaced eyes (≥80%), low-set ears (>80%), and short stature (>70%).

Experienced clinicians are often able to make a diagnosis of NS by recognizing characteristic facial features of NS. Allanson et al. (2010) concluded after subjective clinical exam by two well-trained and experienced clinical geneticists that facial features alone are not sufficient to predict a patient's genotype due to the presence

FIGURE 5 Frontal and lateral facial profiles of Asian individuals with Noonan syndrome. Gender, age, and country of origin found in Supplementary Table S1. [c](Aoki et al., 2013), [d](Edwards et al., 2014), [e](Addissie et al., 2015), [f](Yaoita et al., 2016)

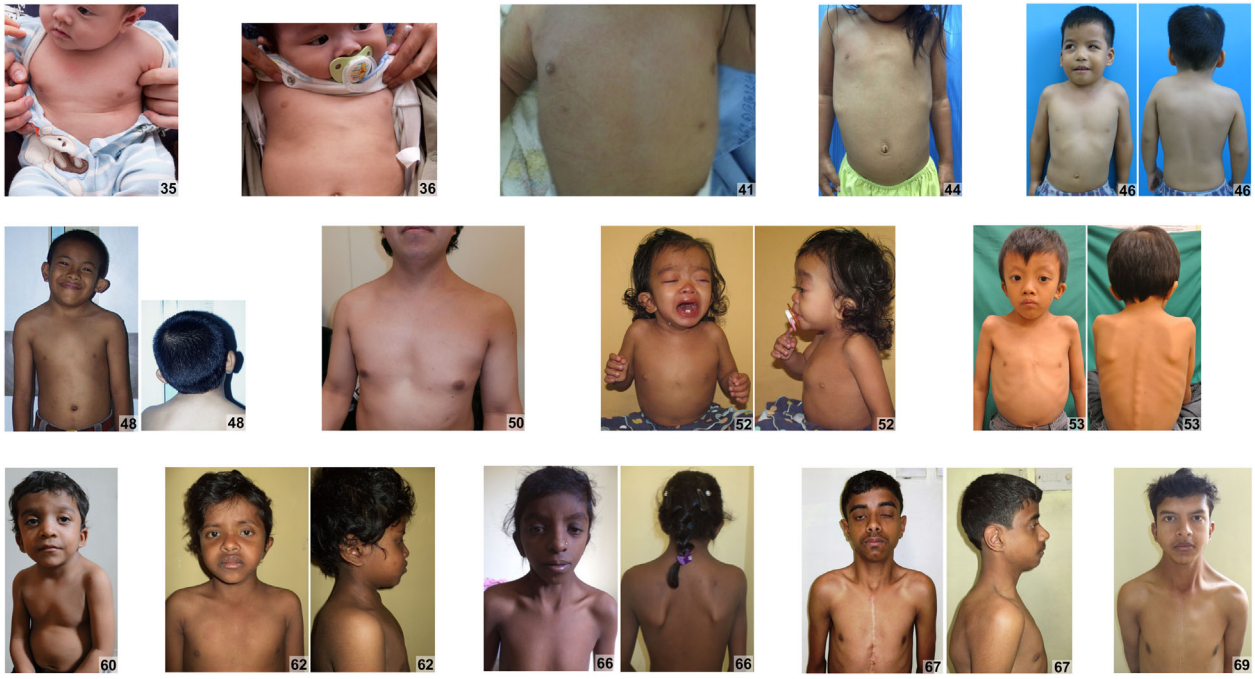

FIGURE 6 Facial and torso profiles of individuals of Asian individuals with Noonan syndrome. Gender, age, and country of origin found in Supplementary Table S1

of atypical features in some of the patients. Given the potential difficulties in clinically recognizing NS, especially when the presentation is atypical, facial analysis technology can be a useful complement to the physician's dysmorphology examination. The facial analysis technology used in our study was able to diagnose patients from all population groups with a sensitivity and specificity of 88% and 89%, respectively (Table 3). There was a significant improvement when separately evaluating population groups by the facial analysis algorithm, which led to sensitivity equal to or greater than 94%, and specificity equal or greater than 90% for all groups (Table 3). The technology identified quantitative facial biometrics specific to NS for each ethnic group. As expected,

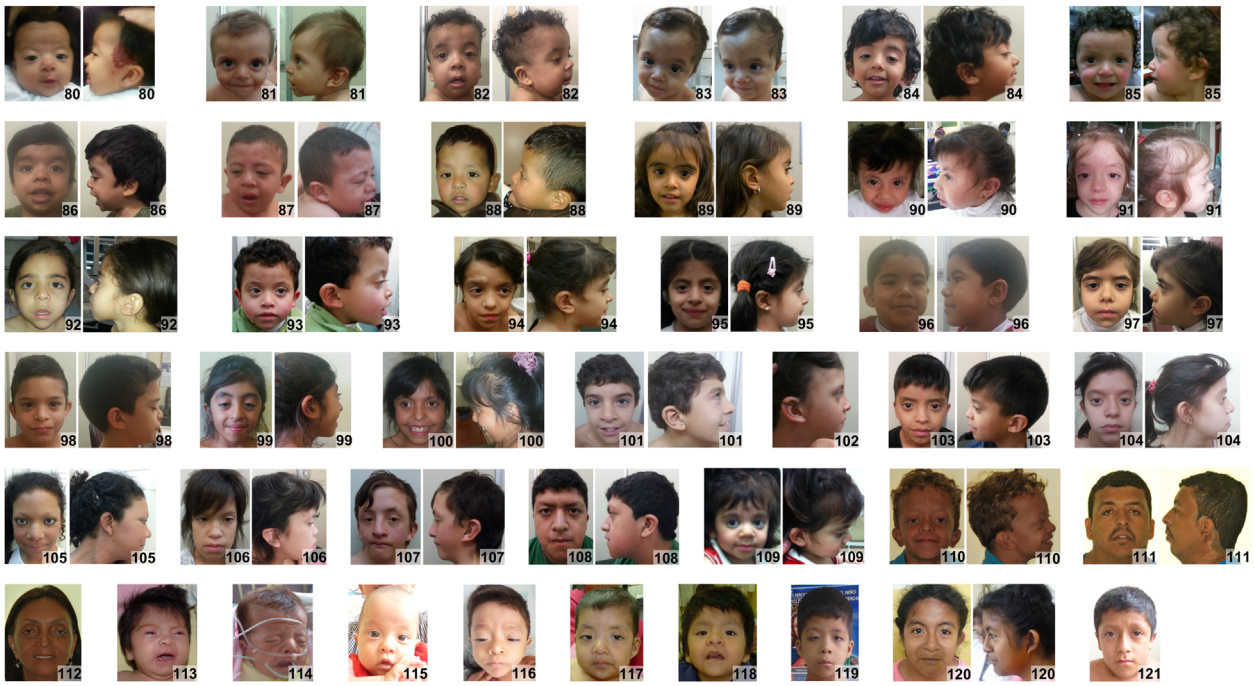

FIGURE 7 Frontal and lateral facial profiles of Latin Americans with Noonan syndrome. Gender, age, and country of origin found in Supplementary Table S1

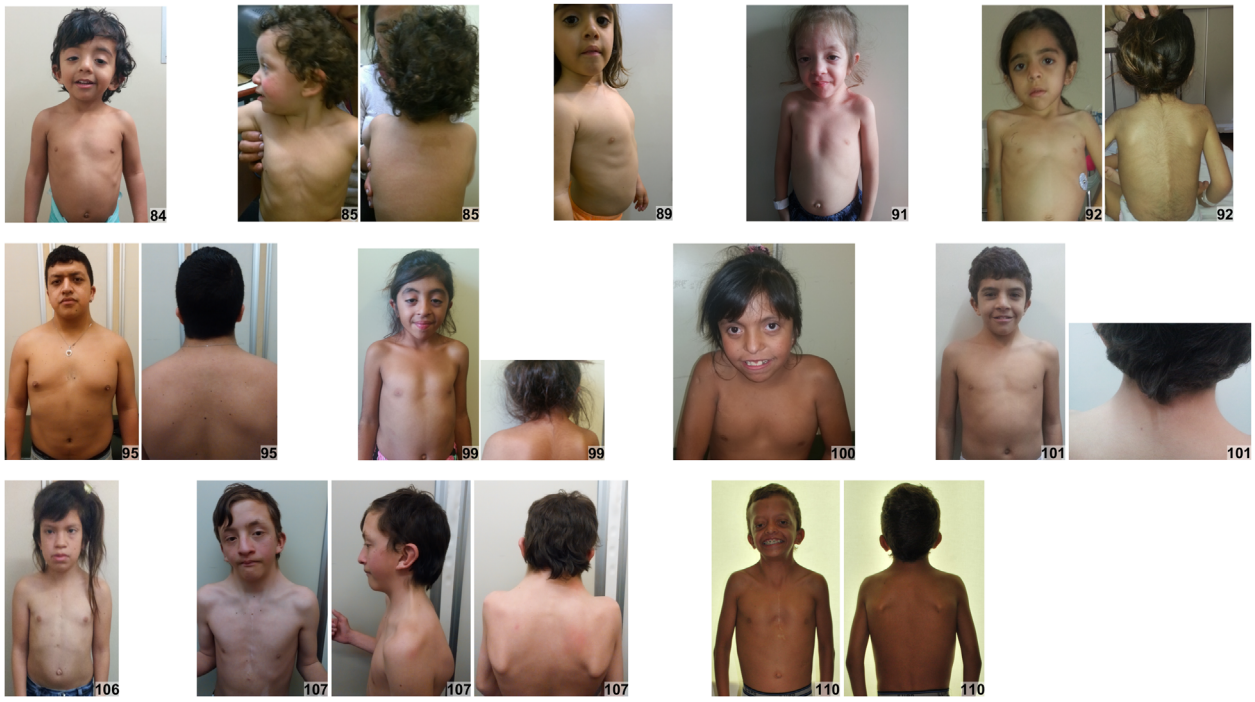

FIGURE 8 Facial and torso profiles of individuals of Latin American individuals with Noonan syndrome. Gender, age, and country of origin found in Supplementary Table S1

our algorithm for facial analysis found widely spaced eyes as a significant facial feature in all ethnic groups (Supplementary Tables S3–S6) as well as for the global population (Supplementary Table S2).

There are several limitations inherent to studies of genetic syndromes in diverse populations. We acknowledge that ascertainment bias exists with only the most severe phenotypes or those with severe congenital heart disease seeking medical attention. Thus, the milder cases of NS are most likely missed, as seen in adults who are often diagnosed only after their more severely affected child is diagnosed; this is further reinforced by the fact that 30–75% of individuals with NS have an affected parent (Allanson & Roberts, 1993). Additionally, in countries with limited resources and access to medical care, molecular genetic testing is difficult compared to developed countries where molecular testing is more widely available. Due to this limitation, we only accepted patients into this study who were diagnosed clinically with NS by a trained clinical geneticist since molecular genetic testing was unavailable in a fraction of our cohort (Supplementary Table S1). Another challenge to these studies is arbitrarily grouping populations geographically, for example, Chinese, Indian, and Malaysian in the category of "Asian." Obviously, every population group is unique, and within countries a significant amount of ethnic diversity and admixture exists. As larger cohorts are assembled through public databases (Muenke, Adeyemo, & Kruszka, 2016), more precise population characterizations will be possible. Additionally, our study does not account for genotype-phenotype correlations which are known to exist, such as pulmonary valve stenosis being more common in individuals with *PTPN11* variants (Tartaglia et al., 2002), or hypertrophic cardiomyopathy being more common in those with *RIT1* variants (Aoki et al., 2013; Yaoita et al., 2016). However, it is important to note that Allanson et al. (2010) did not find a relationship between genotype and specific facial features in

TABLE 3 Measures of diagnostic accuracy for facial analysis technology that discriminate between Noonan syndrome and unaffected individuals, stratified by different populations

	Number of features	AUC	Accuracy	Sensitivity	Specificity
Global	10	0.94	0.89	0.88	0.89
Caucasian	11	0.98	0.94	0.95	0.93
African and African American	5	0.94	0.93	0.94	0.91
Asian	10	0.95	0.93	0.95	0.90
Latin American	6	0.97	0.97	0.96	0.98

AUC, area under the receiver operating characteristic curve.

individuals with NS (Allanson et al., 2010). Finally, it is known that the facial features of individuals with NS change over time making potential genotype-phenotype correlations of this disease aspect difficult to assess (Allanson et al., 1985). Even with the above study limitations, our clinical and facial analysis data appear to be consistent and accurate in the evaluation of NS based on the available data. We would like to emphasize that facial analysis technology is a tool and not a substitute for clinical evaluation as it does not consider other important features of NS such as webbed neck, chest deformities, and congenital heart disease.

Lastly, this study and similar reports (Kruszka, Addissie, et al., 2017; Kruszka, Porras, et al., 2017) and our recently created website, www.genome.gov/atlas will have widespread clinical significance for the diagnosis of individuals with NS, especially in countries without access to genetic services or genetic testing where the simplicity of facial analysis technology may be a useful asset.

ACKNOWLEDGMENTS

We are grateful to the individuals and their families who participated in our study. P.K., Y.A.A, and M.M. are supported by the Division of Intramural Research at the National Human Genome Research Institute, NIH. We thank the Chulalongkorn Academic Advancement into its 2nd Century Project. Partial funding of this project was from a philanthropic gift from the Government of Abu Dhabi to the Children's National Health System. We would like to acknowledge GeneDx and Dr. Benjamin Solomon for providing molecular testing for NS free of charge.

REFERENCES

Addissie, Y. A., Kotecha, U., Hart, R. A., Martinez, A. F., Kruszka, P., & Muenke, M. (2015). Craniosynostosis and Noonan syndrome with KRAS mutations: Expanding the phenotype with a case report and review of the literature. *American Journal of Medical Genetics Part A*, *167A*(11), 2657–2663.

Allanson, J. E., Bohring, A., Dorr, H. G., Dufke, A., Gillessen-Kaesbach, G., Horn, D., … Zenker, M. (2010). The face of Noonan syndrome: Does phenotype predict genotype. *American Journal of Medical Genetics Part A*, *152A*(8), 1960–1966.

Allanson, J. E., Hall, J. G., Hughes, H. E., Preus, M., & Witt, R. D. (1985). Noonan syndrome: The changing phenotype. *American Journal of Medical Genetics*, *21*(3), 507–514.

Allanson, J. E., Roberts, A. E., (1993). Noonan syndrome. In R. A. Pagon, M. P. Adam, H. H. Ardinger, S. E. Wallace, A. Amemiya, L. J. H. Bean, T. D. Bird, N. Ledbetter, H. C. Mefford, R. J. H. Smith, & K. Stephens (Eds.), *GeneReviews(R)*. Seattle (WA): University of Washington.

Aoki, Y., Niihori, T., Banjo, T., Okamoto, N., Mizuno, S., Kurosawa, K., … Matsubara, Y. (2013). Gain-of-function mutations in RIT1 cause Noonan syndrome, a RAS/MAPK pathway syndrome. *American Journal of Human Genetics*, *93*(1), 173–180.

Bertola, D. R., Pereira, A. C., Albano, L. M., De Oliveira, P. S., Kim, C. A., & Krieger, J. E. (2006). PTPN11 gene analysis in 74 Brazilian patients with Noonan syndrome or Noonan-like phenotype. *Genetic Testing*, *10*(3), 186–191.

Bhambhani, V., Muenke, M. (2014). Noonan syndrome. *American Family Physician*, *89*(1), 37–43.

Cai, D., Zhang, C., & He, X. (2010). Unsupervised feature selection for multi-cluster data. Proceedings of the 16th ACM SIGKDD international conference on Knowledge discovery and data mining: ACM. p333–342.

Cerrolaza, J. J., Porras, A. R., Mansoor, A., Zhao, Q., Summar, M., & Linguraru, M. G. (2016). Identification of dysmorphic syndromes using landmark-specific local texture descriptors. Biomedical Imaging (ISBI), 2016 IEEE 13th International Symposium on: IEEE. p1080–1083.

Cortes, C., Vapnik, V. (1995). Support-vector networks. *Machine Learning*, *20*(3), 273–297.

Edwards, J. J., Martinelli, S., Pannone, L., Lo, I. F., Shi, L., Edelmann, L., … Gelb, B. D. (2014). A PTPN11 allele encoding a catalytically impaired SHP2 protein in a patient with a Noonan syndrome phenotype. *American Journal of Medical Genetics Part A*, *164A*(9), 2351–2355.

Elisseeff, A., Pontil, M. (2003). Leave-one-out error and stability of learning algorithms with applications. *NATO Science Series Sub Series III Computer and Systems Sciences*, *190*, 111–130.

Essawi, M. L., Ismail, M. F., Afifi, H. H., Kobesiy, M. M., El Kotoury, A., & Barakat, M. M. (2013). Mutational analysis of the PTPN11 gene in Egyptian patients with Noonan syndrome. *Journal of the Formosan Medical Association*, *112*(11), 707–712.

Hammond, P., Hutton, T. J., Allanson, J. E., Campbell, L. E., Hennekam, R. C., Holden, S., … Winter, R. M. (2004). 3D analysis of facial morphology. *American Journal of Medical Genetics Part A*, *126A*(4), 339–348.

Hung, C. S., Lin, J. L., Lee, Y. J., Lin, S. P., Chao, M. C., & Lo, F. S. (2007). Mutational analysis of PTPN11 gene in Taiwanese children with Noonan syndrome. *Journal of the Formosan Medical Association*, *106*(2), 169–172.

Isojima, T., Sakazume, S., Hasegawa, T., Ogata, T., Nakanishi, T., Nagai, T., & Yokoya, S. (2016). Growth references for Japanese individuals with Noonan syndrome. *Pediatric Research*, *79*(4), 543–548.

Jongmans, M., Sistermans, E. A., Rikken, A., Nillesen, W. M., Tamminga, R., Patton, M., … van der Burgt, I. (2005). Genotypic and phenotypic characterization of Noonan syndrome: New data and review of the literature. *American Journal of Medical Genetics Part A*, *134A*(2), 165–170.

Ko, J. M., Kim, J. M., Kim, G. H., & Yoo, H. W. (2008). PTPN11, SOS1, KRAS, and RAF1 gene analysis, and genotype-phenotype correlation in Korean patients with Noonan syndrome. *Journal of Human Genetics*, *53*(11–12), 999–1006.

Kruszka, P., Addissie, Y. A., McGinn, D. E., Porras, A. R., Biggs, E., Share, M., … Muenke, M. (2017). 22q11.2 deletion syndrome in diverse populations. *American Journal of Medical Genetics Part A*, *173*(4), 879–888.

Kruszka, P., Porras, A. R., Sobering, A. K., Ikolo, F. A., La Qua, S., Shotelersuk, V., … Muenke, M. (2017b). Down syndrome in diverse populations. *American Journal of Medical Genetics Part A*, *173*(1), 42–53.

Lee, A., Sakhalkar, M. V. (2014). Ocular manifestations of Noonan syndrome in twin siblings: A case report of keratoconus with acute corneal hydrops. *Indian Journal of Ophthalmology*, *62*(12), 1171–1173.

Lee, B. H., Kim, J. M., Jin, H. Y., Kim, G. H., Choi, J. H., & Yoo, H. W. (2011). Spectrum of mutations in Noonan syndrome and their correlation with phenotypes. *The Journal of Pediatrics*, *159*(6), 1029–1035.

Lee, S. T., Ki, C. S., & Lee, H. J. (2007). Mutation analysis of the genes involved in the Ras-mitogen-activated protein kinase (MAPK) pathway in Korean patients with Noonan syndrome. *Clinical Genetics*, *72*(2), 150–155.

Malaquias, A. C., Brasil, A. S., Pereira, A. C., Arnhold, I. J., Mendonca, B. B., Bertola, D. R., & Jorge, A. A. (2012). Growth standards of patients with Noonan and Noonan-like syndromes with mutations in the RAS/MAPK pathway. *American Journal of Medical Genetics Part A*, *158A*(11), 2700–2706.

Muenke, M., Adeyemo, A., & Kruszka, P. (2016). An electronic atlas of human malformation syndromes in diverse populations. *Genetics in Medicine*, *18*, 1085–1087.

Ndiaye, R., Ndiaye, C., Leye, M., Mbengue, B., Diallo, M. S., Diop, J. P. D., … Sy, H. S. (2014). Mutation N308T of protein tyrosine phosphatase SHP-

2 in two Senegalese patients with Noonan syndrome. *Journal of Medical Genetics and Genomics*, *6*(1), 6–10.

Noonan, J. A. (1968). Hypertelorism with Turner phenotype. A new syndrome with associated congenital heart disease. *American Journal of Diseases of Children*, *116*(4), 373–380.

Roberts, A. E., Allanson, J. E., Tartaglia, M., & Gelb, B. D. (2013). Noonan syndrome. *Lancet*, *381*(9863), 333–342.

Roberts, A. E., Araki, T., Swanson, K. D., Montgomery, K. T., Schiripo, T. A., Joshi, V. A., … Kucherlapati, R. S. (2007). Germline gain-of-function mutations in SOS1 cause Noonan syndrome. *Nature Genetics*, *39*(1), 70–74.

Romano, A. A., Allanson, J. E., Dahlgren, J., Gelb, B. D., Hall, B., Pierpont, M. E., … Noonan, J. A. (2010). Noonan syndrome: Clinical features, diagnosis, and management guidelines. *Pediatrics*, *126*(4), 746–759.

Simsek-Kiper, P. O., Alanay, Y., Gulhan, B., Lissewski, C., Turkyilmaz, D., Alehan, D., … Boduroglu, K. (2013). Clinical and molecular analysis of RASopathies in a group of Turkish patients. *Clinical Genetics*, *83*(2), 181–186.

Tartaglia, M., Kalidas, K., Shaw, A., Song, X., Musat, D. L., van der Burgt, I., … Gelb, B. D. (2002). PTPN11 mutations in Noonan syndrome: Molecular spectrum, genotype-phenotype correlation, and phenotypic heterogeneity. *American Journal of Human Genetics*, *70*(6), 1555–1563.

Tartaglia, M., Mehler, E. L., Goldberg, R., Zampino, G., Brunner, H. G., Kremer, H., … Gelb, B. D. (2001). Mutations in PTPN11, encoding the protein tyrosine phosphatase SHP-2, cause Noonan syndrome. *Nature Genetics*, *29*(4), 465–468.

Tartaglia, M., Pennacchio, L. A., Zhao, C., Yadav, K. K., Fodale, V., Sarkozy, A., … Gelb, B. D. (2007). Gain-of-function SOS1 mutations cause a distinctive form of Noonan syndrome. *Nature Genetics*, *39*(1), 75–79.

van der Burgt, I., Berends, E., Lommen, E., van Beersum, S., Hamel, B., & Mariman, E. (1994). Clinical and molecular studies in a large Dutch family with Noonan syndrome. *American Journal of Medical Genetics*, *53*(2), 187–191.

van der Burgt, I., Thoonen, G., Roosenboom, N., Assman-Hulsmans, C., Gabreels, F., Otten, B., & Brunner, H. G. (1999). Patterns of cognitive functioning in school-aged children with Noonan syndrome associated with variability in phenotypic expression. *The Journal of Pediatrics*, *135*(6), 707–713.

Yaoita, M., Niihori, T., Mizuno, S., Okamoto, N., Hayashi, S., Watanabe, A., … Aoki, Y. (2016). Spectrum of mutations and genotype-phenotype analysis in Noonan syndrome patients with RIT1 mutations. *Human Genetics*, *135*(2), 209–222.

Yoshida, R., Hasegawa, T., Hasegawa, Y., Nagai, T., Kinoshita, E., Tanaka, Y., … Ogata, T. (2004). Protein-tyrosine phosphatase, nonreceptor type 11 mutation analysis and clinical assessment in 45 patients with Noonan syndrome. *The Journal of Clinical Endocrinology and Metabolism*, *89*(7), 3359–3364.

Zhao, Q., Okada, K., Rosenbaum, K., Kehoe, L., Zand, D. J., Sze, R., … Linguraru, M. G. (2014). Digital facial dysmorphology for genetic screening: Hierarchical constrained local model using ICA. *Medical Image Analysis*, *18*(5), 699–710.

Zhao, Q., Okada, K., Rosenbaum, K., Zand, D. J., Sze, R., Summar, M., & Linguraru, M. G. (2013). Hierarchical constrained local model using ICA and its application to Down syndrome detection. *Medical Image Computing and Computer-Assisted Intervention*, *16*(Pt 2), 222–229.

Zhao, Q., Werghi, N., Okada, K., Rosenbaum, K., Summar, M., & Linguraru, M. G. (2014). Ensemble learning for the detection of facial dysmorphology. *Conference Proceedings IEEE Engineering in Medicine and Biology Society*, *2014*, 754–757.

SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

How to cite this article: Kruszka P, Porras AR, Addissie YA, et al. Noonan syndrome in diverse populations. *Am J Med Genet Part A*. 2017;173A:2323–2334. https://doi.org/10.1002/ajmg.a.38362