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# Hermansky-Pudlak syndrome and oculocutaneous albinism in Chinese children with pigmentation defects and easy bruising

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## Abstract

**Background:** Determining the etiology of oculocutaneous albinism is important for proper clinical management and to determine prognosis. The purpose of this study was to genotype and phenotype eight adopted Chinese children who presented with oculocutaneous albinism and easy bruising.

**Results:** The patients were evaluated at a single center; their ages ranged from 3 to 8 years. Whole exome or direct sequencing showed that two of the children had Hermansky-Pudlak syndrome (HPS) type-1 (HPS-1), one had HPS-3, one had HPS-4, and four had non-syndromic oculocutaneous albinism associated with *TYR* variants (OCA1). Two frameshift variants in *HPS1* (c.9delC and c.1477delA), one nonsense in *HPS4* (c.416G > A), and one missense variant in *TYR* (c.1235C > T) were unreported. The child with HPS-4 is the first case with this subtype reported in the Chinese population. Hypopigmentation in patients with HPS was mild compared to that in OCA1 cases, who had severe pigment defects. Bruises, which may be more visible in patients with hypopigmentation, were found in all cases with either HPS or OCA1. Whole mount transmission electron microscopy demonstrated absent platelet dense granules in the HPS cases; up to 1.9 mean dense granules per platelet were found in those with OCA1. Platelet aggregation studies in OCA1 cases were inconclusive.

**Conclusions:** Clinical manifestations of oculocutaneous albinism and easy bruising may be observed in children with HPS or OCA1. Establishing definitive diagnoses in children presenting with these phenotypic features is facilitated by genetic testing. Non-syndromic oculocutaneous albinism and various HPS subtypes, including HPS-4, are found in children of Chinese ancestry.

**Keywords:** Bruising, Chinese, Dense granule, Hermansky-Pudlak syndrome, Hypopigmentation, Oculocutaneous albinism, Platelet

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## Background

Hypopigmentation of the skin, hair and eyes is a manifestation of various inherited disorders, including non-syndromic oculocutaneous albinism (OCA), Hermansky-Pudlak syndrome (HPS), Chediak-Higashi syndrome, and Griscelli syndrome [1–3]. The underlying cellular mechanisms causing pigmentation defects differ in each of these disorders. Non-syndromic OCA (MIM #203100), comprised of 7 genetically distinct disorders, is characterized by hypopigmentation, due to absent or reduced melanin biosynthesis within melanocytes [4, 5]. There are 10 known genetic subtypes of HPS (MIM #203300, #614072, #614073), characterized by hypopigmentation and a bleeding diathesis, associated with abnormal biogenesis of lysosome-related organelles, which include melanosomes that are melanin-containing organelles within melanocytes [3]. Chediak-Higashi syndrome (MIM#214500) is a rare lysosomal disorder associated with biallelic variants in *LYST* and enlarged, membrane-rich, lysosome-related organelles, resulting in pigmentary, hematologic, and neurologic involvement [6, 7]. Griscelli syndrome (MIM#214450), characterized by hypopigmentation, is a melanosome transport disorder associated with variants in *MYO5A*, *RAB27A*, or *MLPH* [2]. Pigmentation is generally reduced in these disorders, but the severity of the pigment defect is variable.

Non-syndromic OCA and HPS are autosomal recessive disorders that share some, but not all, clinical features. Manifestations of both non-syndromic OCA and HPS include decreased skin and hair pigmentation, nystagmus, iris transillumination, reduced visual acuity, increased optic nerve decussation, and foveal hypoplasia [1–3, 8]. HPS, but not non-syndromic OCA, is further characterized by a bleeding diathesis due to a platelet storage pool defect [9]. In addition, some patients with HPS develop granulomatous colitis, neutropenia, or pulmonary fibrosis [2, 3, 8]. Specifically, children and young adults with HPS type-2 (HPS-2) can develop neutropenia responsive to granulocyte-stimulating factor, immunodeficiency, interstitial lung disease, and/or pulmonary fibrosis [10–13]. Fibrotic lung disease is highly prevalent in middle-aged adults with HPS-1 and HPS-4, and this condition is associated with substantial morbidity and mortality [9, 14, 15]. Given the high risk of fibrotic lung disease in patients with some subtypes of HPS, but not in non-syndromic OCA, establishing an accurate diagnosis of HPS in patients with oculocutaneous albinism and identifying their HPS subtype are essential for anticipating symptoms, providing proper clinical management, and for estimating prognosis.

The prevalence of non-syndromic OCA and HPS differs considerably. The worldwide prevalence of all known forms of OCA is estimated to be 1 per 17,000 [16]. OCA1, which is associated with variants in *TYR*,

affects approximately 1 per 40,000 in the general population [17, 18]. OCA is reported to affect approximately 1 per 18,000 in the Chinese Han population in the eastern Chinese province of Shandong [5]. In contrast, HPS is estimated to occur in only 1–9 persons per 1,000,000 worldwide and, similar to OCA, also varies based on genetic subtype and region. For example, Puerto-Ricans are predisposed to HPS-1 and HPS-3 due to the presence of founder variants in the *HPS1* and *HPS3* genes in the Puerto-Rican population [19, 20]. HPS-1 affects 1 per 1800 persons in northwest Puerto Rico, and HPS-3 occurs in 1 per 16,000 persons throughout Puerto Rico [3, 20, 21]. Thus far, few patients of Chinese ancestry are reported with HPS [22–25].

In general, ocular findings and hypopigmentation are prominent features of patients affected with either HPS or non-syndromic OCA, and patients can be misdiagnosed [21]. HPS is rare, so children with oculocutaneous albinism are initially suspected of having non-syndromic OCA. Conversely, although excessive bruising is a feature of HPS, bruises in patients with non-syndromic OCA can be readily apparent due to reduced skin pigmentation, and ecchymoses in a person with albinism may be interpreted as easy bruisability. Hence, some children with non-syndromic OCA and a normal amount of bruising may be initially misdiagnosed with HPS.

We report eight adopted children of Chinese ancestry with oculocutaneous albinism, a tendency to bleed, and platelet dysfunction who were suspected to have HPS. Whole exome or direct sequencing analyses revealed variants in genes associated with HPS or the tyrosinase gene (*TYR*). Two children were diagnosed with HPS-1, one with HPS-3, one with HPS-4, and four with OCA1. Children with HPS had absent dense granules; those with OCA1 had low numbers of dense granules per platelet and inconclusive platelet functional test results. Some of their genetic variants are novel, and the patient with HPS-4 is the first reported HPS-4 case of Chinese ethnicity. This study highlights the importance of genetic testing in establishing an accurate diagnosis in patients of Chinese ancestry with albinism and suspected bleeding diathesis.

## Results

### Patient genotyping

Whole exome or Sanger sequencing analysis identified homozygous or compound heterozygous variants in *HPS1* (NM\_000195.4), *HPS3* (NM\_032383.4), *HPS4* (NM\_022081.5), or *TYR* (NM\_000372.4) in 8 children of Chinese descent (Table 1). Single-nucleotide polymorphism analysis excluded hemizygoty in the child (435) with a homozygous variant in *HPS4*, but hemizygoty is possible in the two children (215 and 351) whose

**Table 1** Gene Variants in our Cohort and Frequencies in the East Asian Population

Patient	Gene	Variant 1	East Asian MAF <sup>a</sup>	Variant 2	East Asian MAF <sup>a</sup>
215	<i>HPS1</i>	c.9delC p.Cys3Trpfs*26	N/R	c.9delC p.Cys3Trpfs*26	N/R
351	<i>HPS3</i>	c.1555_1595dup p.Leu533Phefs*10	0.0001160 (gnomAD)	c.1555_1595dup p.Leu533Phefs*10	0.0001160 (gnomAD)
435	<i>HPS4</i>	c.416G > A p.Trp139*	N/R	c.416G > A p.Trp139*	N/R
471	<i>HPS1</i>	c.972dupC p.Met325Hisfs*128	0.001458 (ExAc) <sup>b</sup>	c.1477delA p.Arg493Glyfs*22	N/R
468	<i>TYR</i>	c.929dup p.Arg311Lysfs*7	0.000694 (ExAc) <sup>c</sup>	c.896G > A p.Arg299His	0.000347 (ExAc) <sup>d</sup>
469	<i>TYR</i>	c.230_232dupGGG p.Arg77_Glu78insGly	0.0001168 (ExAc)	c.896G > A p.Arg299His	0.000347 (ExAc) <sup>d</sup>
470	<i>TYR</i>	c.230_232dupGGG p.Arg77_Glu78insGly	0.0001168 (ExAc)	c.1235C > T; p.Pro412Leu	N/R
474	<i>TYR</i>	c.655G > A p.Glu219Lys	N/R <sup>e</sup>	c.896G > A p.Arg299His	0.000347 (ExAc) <sup>d</sup>

MAF Minor Allele Frequency, N/R None reported

<sup>a</sup>The ExAc (Beta; Exome Aggregation Consortium; <http://exac.broadinstitute.org/>) and GnomAD browsers (Beta; Genome Aggregation Database; <http://gnomad.broadinstitute.org/>)

were searched in September 2018

<sup>b</sup>ClinVar Accession RCV000005596.4

<sup>c</sup>ClinVar Accession RCV000003969.5

<sup>d</sup>ClinVar Accession RCV000004000.4

<sup>e</sup>Reported in Reference [29]; not reported in ExAc or GnomAD

variants were identified by direct sequencing. No pathogenic variants were identified in genes associated with Chediak-Higashi syndrome (*LYST*) or Griscelli syndrome (*MYO5A*, *RAB27A*, *MLPH*) in the 6 patients who underwent whole exome sequencing analysis. None of the children were known to be related, but three children with OCA1 were from the same orphanage. Four patients originated from the Nanjing area, one from Beijing, one from northwest China, one from southern China, and one from an unknown region of China (Table 2). Limited to no information was available about the children's biological parents, and thus, history of consanguinity is unknown. Two of four children with HPS had HPS-1. One patient (215) had a frameshift variant in *HPS1*, and the other patient (471) had compound heterozygous frameshift variants, leading to premature

termination codons. Two of the three *HPS1* variants (c.9delC; p.Cys3Trpfs\*26 and c.1477delA; p.Arg493Glyfs\*22) were not previously reported. The third variant (c.972dupC; p.Met325Hisfs\*128) is a frequently reported *HPS1* variant in cases of different ethnicities, including Caucasian and Chinese [23, 26, 27]. One child (351) had a variant in *HPS3* (c.1555\_1595dup41; p.Leu533Phefs\*10). A query of the ExAC and GnomAD databases showed that the allele frequency for this mutation is very low in East Asian populations, suggesting that this variant is rare. One patient (435) with HPS-4 was homozygous for a novel nonsense variant in *HPS4* (c.416G > A; p.Trp139\*); this variant is not reported in ExAc nor GnomAD databases.

Four patients (468, 469, 470, 474) with compound heterozygous variants in *TYR* were diagnosed with OCA1.

**Table 2** Patient Characteristics

Patient	Diagnosis	Region	Age (yrs)	Gender	Bruising	Epistaxis	Infections
215	HPS-1	Northwest	6	Male	yes	yes	no
351	HPS-3	South	6	Female	yes	yes	no
435	HPS-4	Nanjing	3	Male	yes	yes	no
471	HPS-1	N/A	3	Female	yes	no	no
468	OCA1	Nanjing	8	Female	yes	no	yes
469	OCA1	Nanjing	5	Male	yes	no	yes
470	OCA1	Nanjing	3	Female	yes	no	yes
474	OCA1	Beijing	8	Female	yes	yes	no

HPS, Hermansky-Pudlak syndrome

N/A, not available

OCA, oculocutaneous albinism

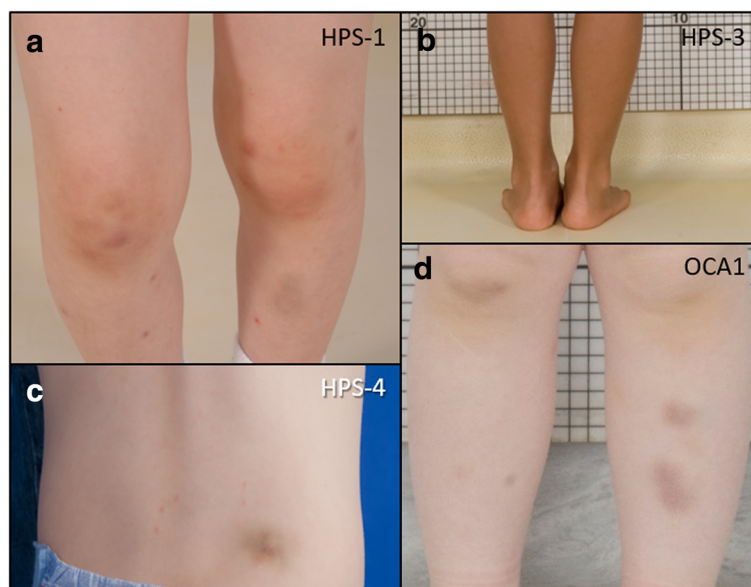
Three children (468, 469, 474) had one previously reported missense variant in common (c.896G>A; p.Arg299His), but their second variant was different. Overall, five variants in *TYR* were found in these four individuals with OCA1. Two of these variants were previously reported (c.896G>A; p.Arg299His and c.230\_232dupGGG; p.Arg77\_Glu78insGly), including frequently in subjects of Chinese and Taiwanese ethnicity [22, 28]. Pathogenicity of the missense variant c.896G>A was ascribed to its location in the copper-binding site of *TYR*, while pathogenicity of c.230\_232dupGGG was ascribed to truncation of the *TYR* peptide [22, 25]. One missense *TYR* variant (c.655G>A; p.Glu219Lys) was not reported in the ExAC and GnomAD variant databases, but was published [29]. The c.655G>A variant is located in a copper-binding site of *TYR*; in silico pathogenicity analysis predicted severe pathogenicity to protein function, including ‘damaging’ (score 0) by SIFT (<http://sift.jcvi.org/>), ‘probably damaging’ (score 1) by PolyPhen-2 (<http://genetics.bwh.harvard.edu/pph2/>), and ‘disease’ (score 0.92 [94%]) by PMut (<http://mmb.pcb.ub.es/PMut/>). One novel missense *TYR* variant (c.1235C>T; p.Pro412Leu) was not reported in the ExAC and GnomAD variant databases nor in the albinism database (<http://www.ifpcs.org/albinism/>). The c.1235C>T variant also yielded severe in silico pathogenicity prediction scores, including ‘damaging’ (score 0) by SIFT, ‘probably damaging’ (score 1) by PolyPhen-2, and ‘disease’ (score 0.81 [89%]) by pMut.

### Clinical characteristics

The mean age of the patients with HPS was 4 years, 8 months and that of patients with OCA1 was 6 years, 2 months (Table 2). Two patients with HPS and one with OCA1 were male. The subjects initially presented with a mild albinotic phenotype and a tendency to bleed with multiple bruises at varying stages of resolution or in unusual sites (Fig. 1a-d). Patients with HPS-1 or HPS-4 had pale skin and light brown hair, which is atypical for individuals of East Asian ancestry who generally have black hair color (Fig. 2a-d). One child with HPS-3 had dark brown hair, and her skin was able to darken with exposure to sunlight. The patients with OCA1 generally had a more severe albinotic phenotype. These patients had white skin and hair indicating a severe pigment defect (Fig. 2e-h). All eight patients reported easy bruising (Table 2). Three children with HPS and one with OCA1 had epistaxis. Three children with OCA1 reported frequent upper respiratory tract infections with several episodes per year. No patients in either group reported colitis or lung disease, and chest computed tomography scans did not show interstitial lung disease or pulmonary fibrosis (Fig. 3a-d).

### Eye findings

All subjects underwent ophthalmologic examination to assess visual acuity, nystagmus and motility, foveal hypoplasia, and iris transillumination (Table 3, Fig. 4a-h). Snellen visual acuity was abnormal in all patients. Visual



**Fig. 1** Bruising in Chinese Children with Hermansky-Pudlak Syndrome or Non-syndromic Oculocutaneous Albinism Type 1. Clinical images depict large or multiple bruises in different stages of resolution in four Chinese children with Hermansky-Pudlak syndrome (HPS) or non-syndromic oculocutaneous albinism type 1 (OCA1). Lower extremities of a patient with HPS-1 (a), HPS-3 (b), or OCA1 (d) are shown; posterior lumbar region of the patient with HPS-4 (c) is also shown. Variable degrees of skin pigmentation defects are evident in these four Chinese children. Hypopigmentation is milder in three children with HPS, especially the one with HPS-3, compared to one with OCA1, who has white skin



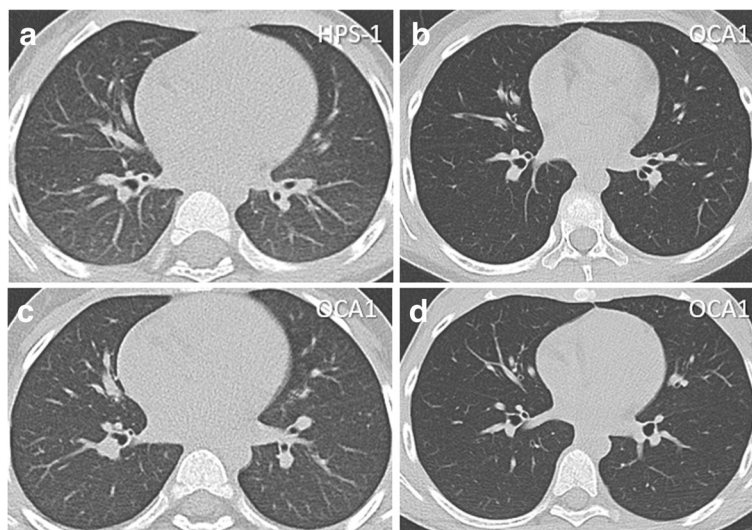
**Fig. 2** Hypopigmentation in Chinese Pediatric Patients with Hermansky-Pudlak Syndrome or Non-syndromic Oculocutaneous Albinism Type 1. An albinotic phenotype is observed in four Chinese children with Hermansky-Pudlak syndrome (HPS) (a-d) and four Chinese children with non-syndromic oculocutaneous albinism type 1 (OCA1) (e-h). The patients with HPS-1 or HPS-4 had light brown or blond hair (a, c, or d), which is atypical for Chinese individuals who generally have black hair. The child with HPS-3 had dark brown hair. A severe pigment defect is observed in four patients with OCA1, who had white hair (e-h)

acuity ranged from 20/80 to 20/400 in patients with HPS and 20/125 to 20/640 in those with OCA1. Although pigmentation defect was relatively mild in the child with HPS-3, visual acuity of this patient was similar to that of other cases with HPS or OCA1. All patients had nystagmus and foveal hypoplasia. Iris transillumination was assessed using a 5 point grading scale [30]. Consistent with her mild pigmentation defect, the patient with HPS-3 had an iris transillumination score of 0. However, although the children with OCA1

generally had more severe hypopigmentation of their skin and hair compared to those with HPS, iris transillumination scores in children with OCA1 (mean score = 2.8) and HPS (mean score = 2) did not differ significantly ( $p = 0.46$ ).

**Platelet Electron microscopy and function**

Prothrombin time, activated partial thromboplastin time, and platelet counts were within normal range for patients with HPS or OCA1, except for one patient with



**Fig. 3** Chest Imaging in Chinese Children with Hermansky-Pudlak Syndrome or Non-syndromic Oculocutaneous Albinism Type 1. Representative images of computer tomography scans of the chest demonstrate normal lung parenchymal tissue in a Chinese pediatric patient with HPS-1 (a) or three with OCA1 (b-d). There is no radiographic evidence of interstitial lung disease or pulmonary fibrosis

**Table 3** Eye Manifestations of Chinese Children with Hermansky-Pudlak Syndrome or Non-syndromic Oculocutaneous Albinism Type 1

Patient	Diagnosis	Visual Acuity OD/OS	Nystagmus	Foveal Hypoplasia	Iris Transillumination Score
215	HPS-1	20/400 20/400	yes	yes	4
351	HPS-3	20/160+ 20/125	yes	yes	0
435	HPS-4	20/150 20/80	yes	yes	3
471	HPS-1	20/200 20/200	yes	yes	1
468	OCA1	20/400 20/640	yes	yes	3
469	OCA1	20/400 20/320	yes	yes	2
470	OCA1	20/200 20/250	yes	yes	3
474	OCA1	20/125 20/125	yes	yes	3

HPS with a mildly reduced platelet count (Table 4). Platelet analysis by whole mount transmission electron microscopy showed absent dense granules in the patients with HPS regardless of their HPS subtype (Fig. 5a and b; Table 4). Chinese pediatric patients with OCA1 had 1.35 to 1.9 mean dense granules per platelet (Fig. 5c and d; Table 4). The lower limit of normal for children is 2 dense granules per platelet [31], however, the normal range for children of Chinese heritage is not reported.

Functional studies of platelets from patients with OCA1 were performed. Platelet aggregation using epinephrine was assessed in whole blood and platelet-rich plasma specimens. Whole blood aggregation studies were normal. However, the secondary wave of platelet aggregation was absent or delayed in response to high or low epinephrine in two patients (468 and 470) with OCA1, and adenosine triphosphate release was absent or delayed in all four patients with OCA1 (Table 5).

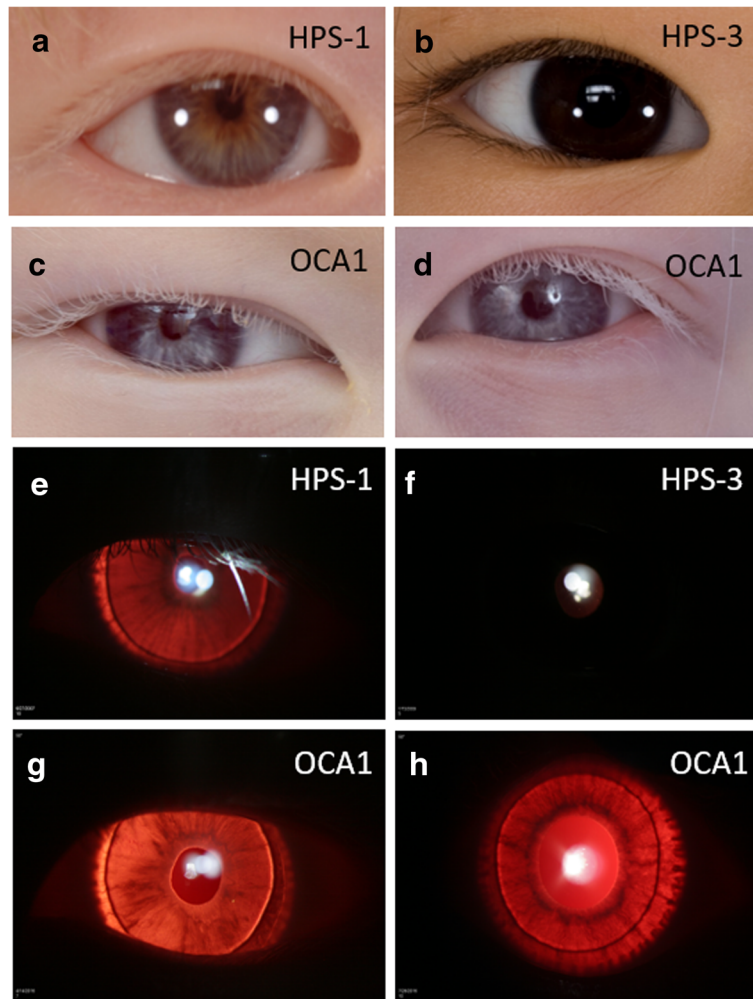
## Discussion

Hermansky-Pudlak syndrome and non-syndromic oculocutaneous albinism are inherited disorders that present with pigmentation defects affecting the hair, skin, and eyes. A bleeding diathesis secondary to a platelet storage pool defect, due to absent dense granules, is found in patients with HPS, but has not been reported in OCA1. Notably, bruises in patients with pigment defects are generally easily detected and may be misinterpreted as a bleeding diathesis.

Given some of their shared manifestations, establishing a diagnosis of HPS or OCA1 solely based on history and physical examination is unreliable, but certain clinical findings may raise the suspicion of one diagnosis over the other. For example, striking pigment defects

with seemingly no hair color and blue irides were found in our patients with OCA1 compared to less severe hypopigmentation in our patients with HPS. However, pigment defects in HPS vary, and some patients with HPS can have profound hypopigmentation [21, 32]. In addition, colitis resembling Crohn's disease, neutropenia, immunodeficiency, and pulmonary fibrosis can affect some patients with HPS, and the diagnosis of any of these co-morbidities in a patient with oculocutaneous albinism should raise the index of suspicion for HPS. Pulmonary fibrosis affecting patients with HPS-1, HPS-2 and HPS-4 is an important diagnosis, because it is a major cause of morbidity and mortality in patients with HPS [9, 15]. Medical therapy is not currently approved as treatment for this complication. Anecdotal cases of patients with HPS pulmonary fibrosis receiving long-term open-label pirfenidone suggest that some may benefit from pirfenidone [33]. However, results of clinical trials investigating pirfenidone as treatment for HPS pulmonary fibrosis were inconclusive [34, 35]. Some patients with severe HPS pulmonary fibrosis received lung transplants [36, 37]. Unfortunately, not all lung transplant candidates with HPS-1 receive donor organs, and alloimmunization is a barrier to lung transplantation for some HPS-1 patients [36]; this can be avoided by judicious use of platelet transfusions throughout patients' lives. Thus, establishing a reliable diagnosis of HPS, identifying the HPS genetic subtype, and long-term clinical planning to maintain candidacy for potential lung transplantation are essential for these patients.

Diagnostic testing of patients presenting with albinism and easy bruising can include platelet analyses and genetic studies. Functional platelet testing can provide results suggestive of storage pool disorders, such as HPS.

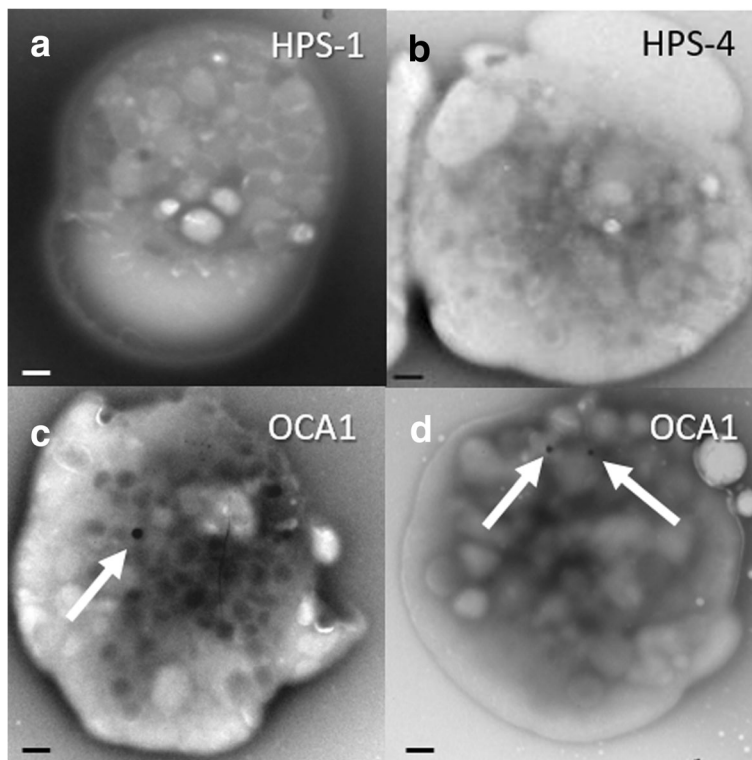


**Fig. 4** Ophthalmologic Findings in Chinese Pediatric Patients with Hermansky-Pudlak Syndrome or Non-syndromic Oculocutaneous Albinism Type 1. Representative images show light-colored eyes and eyelashes as well as iris transillumination in Chinese children with HPS-1 (a, e), HPS-3 (b, f), or OCA1 (c, d, g, h). Pigmentation defects in pediatric patients with HPS, particularly one with HPS-3 and dark brown irides, are not as severe as those in two patients with OCA1 and light blue irides. Eyelashes are blond in a child with HPS-1, dark brown in one with HPS-3, and white in two with OCA1

**Table 4** Hematological Profile of Chinese Children with Hermansky-Pudlak Syndrome or Non-syndromic Oculocutaneous Albinism Type 1

Patient	PT [11.6–15.2 s]	aPTT [25.3–37.3 s]	Platelet count [189–394 K/uL]	Mean dense bodies/platelet
215	N/A	N/A	324	0
351	13.6	34.5	160	0
435	13.7	32.7	264	0
471	13.8	34.9	289	0
468	13.9	38.4	268	1.4
469	13.5	32.4	216	1.9
470	14.5	35.3	292	1.9
474	14	37.1	321	1.35

N/A, not available



**Fig. 5** Platelet Imaging in Chinese Children with Hermansky-Pudlak Syndrome or Non-syndromic Oculocutaneous Albinism Type 1. Representative whole-mount transmission electron microscopy images of platelets demonstrate absent dense granules in platelets from Chinese pediatric patients with HPS-1 (a) or HPS-4 (b). A few dense granules (white arrows) are shown in platelets from two different children with OCA1 (c, d) (scale bar = 500 nm)

However, these results are not diagnostic and can be inconclusive, as shown in our patients with OCA1 who had abnormal platelet aggregation test results [38–41]. Indeed, the two patients with OCA1 and a mean of 1.9 dense granules per platelet had differing aggregation studies using platelet rich plasma. We acknowledge that normal ranges of platelet function assays and concentrations of dense granules, which can affect platelet aggregation results, have not been elucidated in Chinese children. The paucity of information in this specific population of patients further complicates the interpretation of their test results. Studies are indicated to define normal ranges in this population and to determine whether or not an unrecognized bleeding diathesis is

associated with OCA1. Whole mount transmission electron microscopy of platelets showing absent dense granules is another diagnostic test for patients with HPS [41]. However, proper collection and processing of blood samples are needed to avoid platelet activation and degranulation [42]. In addition, absent dense granules are typically found in all patients with HPS, so this test does not provide insights into HPS genetic subtypes. Indeed, this study showed absent platelet dense granules in patients with HPS-1, HPS-3, and HPS-4, who have differing prognoses. Given the prognostic implications associated with certain HPS genetic subtypes, clinicians should consider evaluating children with oculocutaneous albinism and an increased tendency to bleed using

**Table 5** Platelet Aggregation Studies of Chinese Children with Non-syndromic Oculocutaneous Albinism Type 1

Patient	PRP epinephrine low	PRP epinephrine high	PRP epinephrine low ATP release	PRP epinephrine high ATP release
468	absent secondary wave	absent secondary wave	absent	absent
469	normal	normal	delayed	delayed
470	absent secondary wave	delayed secondary wave	absent	absent
474	normal	normal	delayed	delayed

aPTT, activated partial thromboplastin time  
 ATP, adenosine triphosphate  
 PRP, platelet rich plasma  
 PT, prothrombin time



genetic testing for genes associated with HPS, non-syndromic oculocutaneous albinism, Chediak-Higashi syndrome, or Griscelli syndrome. Such genetic testing could be used on a case-by-case basis as a substitute for classical evaluations involving platelet functional analyses or whole mount transmission electron microscopy.

In this cohort of eight Chinese children with clinical features consistent with HPS, genetic sequencing showed that two had HPS-1, one had HPS-3, one had HPS-4, and four had OCA1. Previous studies focusing on Chinese patients with hypopigmentation identified several individuals with OCA1, OCA2, and different HPS subtypes, including HPS-1, HPS-3, HPS-5, and HPS-6 [5, 23, 24, 43]. Notably, our study is the first to report a Chinese patient with HPS-4; this result is consistent with the absence of available data for this novel *HPS4* variant in the East Asian population in ExAC or GnomAD. In addition, some children in our study have unreported genetic variants in *HPS1* and *TYR*, which are predicted to be pathogenic.

## Conclusions

Genetic testing in cases of suspected HPS and OCA is recommended to definitively diagnose patients. Although platelet whole mount transmission electron microscopy and aggregation studies can assist in establishing a diagnosis, the results may be inconclusive and, unlike molecular testing, they do not determine HPS genetic subtypes. Furthermore, these cases illustrate that various HPS subtypes associated with differing prognoses can affect individuals of Chinese ancestry. Our cohort also highlights the genetic heterogeneity of Chinese patients with oculocutaneous albinism and easy bruisability.

## Methods

### Informed consent

Patients' parents provided written informed consent to participate in protocols 76-HG-0238 (Clinical Trials number NCT00369421; Diagnosis and Treatment of Patients with Inborn Errors of Metabolism or Other Genetic Disorders), and/or 95-HG-0193 (Clinical Trials number NCT00001456; Clinical and Basic Investigations into Hermansky-Pudlak Syndrome), which were approved by the institutional review board of the National Human Genome Research Institute. Testing was performed for diagnostic evaluation of each patient at the National Institutes of Health Clinical Center in Bethesda, Maryland.

### Clinical examinations

Patients were examined for bruising, petechiae, and lung disease. Ophthalmologists with expertise in oculocutaneous

albinism also evaluated patients for visual acuity, iris transillumination, nystagmus, foveal hypoplasia and optical coherence tomography (OCT) when possible. Iris transillumination was graded using a 5-point scale as described (grade 0 = no iris transillumination; grade 4 = complete iris transillumination) [30].

Chemistries, complete blood counts, and coagulation tests were performed. Computed tomography scans of the chest with high resolution images were obtained in some patients as described [44].

### Platelet testing

For functional assays, platelets isolated from platelet-rich plasma were subjected to light-transmission aggregometry and adenosine triphosphate release using high and low concentrations of epinephrine as described [45]. For platelet dense granule analysis, transmission electron microscopy of whole mount platelets was performed by an experienced hematologist [46]. For each case, 100–200 platelets were counted.

### Genetic analysis

For patients 215 and 351, genomic DNA isolated from peripheral blood was analyzed by di-deoxy Sanger sequencing for candidate genes. Sequencing of polymerase chain reaction amplification products was performed using a genetic analyzer (Applied Biosystems 3130x; Foster City, CA) as described [46]. For the 6 other patients, genomic DNA isolated from peripheral blood was analyzed by single-nucleotide polymorphism array using the HumanOmniExpress DNA Analysis BeadChip (Illumina, San Diego, CA) and the GenomeStudio software (Illumina) as described [46]. Whole exome sequencing was performed by the National Institutes of Health Intramural Sequencing Center using the HiSeq2000 (Illumina) and the Illumina Genome Analyzer Pipeline software (V.1.13.48.0) as described [46]. Variant filtering was based on allele frequency less than 0.01 with no reported healthy homozygotes in publicly available databases, dbSNP, ExAC, gnomAD and 1000G. Pathogenicity of the identified missense variants or in-frame indels was assessed using online prediction tools, Polyphen, CADD, SIFT, and Mutation Taster. Denovo variants were not identified, because all of the affected individuals were adopted. Compound heterozygosity of the variants were predicted if more than one possible pathogenic variants were observed for a gene. Sanger sequencing in a CLIA (Clinical Laboratory Improvement Amendments) approved laboratory confirmed the identified variants in each patient.

### Abbreviations

CLIA: Clinical laboratory improvement amendments; HPS: Hermansky-Pudlak syndrome; OCA: Oculocutaneous albinism; OCT: Optical coherence tomography

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### Availability of data and materials

The datasets used and/or analysed during the current study may be available from the corresponding author to approved investigators on reasonable request.

### Authors' contributions

Conceptualization: WAG, BRG; Investigation, Methodology, and Analysis: BP, CRF, DC, WMZ, KJO, WJI, JS, WAG, MH, MCV, DRA, BRG.; Writing – Original Draft Preparation: BP, WMZ, BRG; Writing – Critical Review & Editing: BP, CRF, DC, WMZ, KJO, WJI, JS, WAG, MH, MCV, DRA, BRG. All authors read and approved the final manuscript.

### Ethics approval and consent to participate

Patients were admitted to the National Institutes of Health under protocols 76-HG-0238 (Clinical Trials number NCT00369421; Diagnosis and Treatment of Patients with Inborn Errors of Metabolism or Other Genetic Disorders), and/or 95-HG-0193 (Clinical Trials number NCT00001456; Clinical and Basic Investigations into Hermansky-Pudlak Syndrome), which were approved by the institutional review board of the National Human Genome Research Institute. Patients' parents provided written informed consent.

### Consent for publication

Written consent for publication was obtained from the patients' parents at study enrollment.

### Competing interests

The authors declare that they have no competing interests.

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