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RESEARCH ARTICLE

Noninvasive Colour-Based Card for Risk Assessment of Anaemia in Children: PAN-India User Experience

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ABSTRACT

Background: Easy-to-use inexpensive tools are needed for the detection and timely intervention for anemia in children. As an extension of an earlier user-experience in 9 paediatricians, we present real-world experience of 84 paediatricians for the assessment of anemia with the use of the Non-invasive Screening of Anemia (NISA) colour-based shade card in 1459 children aged 6- 60 months.

Methods: The NISA shade card was used to determine the presence and severity (mild, moderate, severe) of anemia by comparing the natural colour tint or pallor of palpebral conjunctiva with 13 ordinal shades. A feedback questionnaire was used to assess the experience of using the NISA card. Results were summarized using descriptive statistics.

Results: The sensitivity and specificity of the NISA shade card for detection of anemia were 80.83% (95% CI: 72.64% to 87.44%) and 66.04% (95% CI: 51.73% to 78.48%) respectively. The NISA card had an accuracy of 81.28%; this ranged from 77.74% to 85.23% in different regions across India. Most clinicians rated the NISA card as good to excellent for its utility in screening for anemia especially in situations where drawing of blood could be avoided.

Conclusion: With an accuracy and sensitivity of >80%, the NISA shade card is a simple tool that can easily be deployed in community settings for the assessment of anemia. The tool can find its extended use in planning and implementing nutritional practices in children.

keywords: Anemia, Non-invasive, Children, Screening tool, Sensitivity, Specificity, Colour-based card

Introduction

Anemia is the most common haematological condition in children. According to the World Health Organization (WHO), anemia is diagnosed in children aged 6 months to 5 years when haemoglobin (Hb) levels are <11 g/dl.[1] Despite high prevalence in developing countries, anemia is a global burden affecting around 40% of children worldwide.[2] Anemia is widely prevalent in India and the National Family Health Surveys (NFHS) have reported an increase in the prevalence of anemia from 58% to 67%, in NFHS-4 & NFHS-5 respectively in children <5 years of age.[3,4,5]

Screening is the mainstay in the detection and management of anemia in children. Prevention of anemia is key to support the growth and development of children. It can have adverse short-term and long-term consequences of morbidity from infections, stunted growth, and cognitive impairment in children. It is important to develop sensitive, specific, and cost-effective screening tools for the easy and early risk assessment of anemia in children.[6] This can improve the growth and psychomotor development in children. Timely risk assessment and intervention of anemia can reduce its debilitating effects. The conventional laboratory assessment of haemoglobin (Hb) level is used to diagnose and determine the severity of anemia. Other tests include haematocrit and serum ferritin. These laboratory tests require a blood sample to be drawn and are invasive and require expertise. The cost of these tests is a limitation to the wider application of the tests in a larger population.[7] Periodic screening for anemia in children should be adopted at the population level in a developing country like India which could help in the timely intervention and its correction. This becomes challenging in a resource-limiting setting. Acceptance of the screening procedure is also a challenge as mothers are hesitant for the child to be subjected to a prick for retrieving a blood sample.

In clinical settings, the first step to the diagnosis of anemia is the assessment of pallor in the conjunctiva, tongue, palm, and nailbed. This is an easy clinical sign that can be evaluated without the use of any specific tools. Conjunctival pallor is a common cue to the presence and severity of anemia. More the pallor of the conjunctiva, greater the probability of the patient being anemic.[8,9] However, clinical signs and symptoms of anemia are not that evident in its early stages and may manifest only in moderate to severe cases.

Palpebral conjunctiva is a mucous membrane with superficial and rich capillaries not readily influenced by skin colour and temperature. The colour of palpebral conjunctiva tends to be gradually lighter with the reduction in Hb concentration and erythrocytes. Given that the deep or light colour of palpebral conjunctiva reflects the degree of anemia, a visual examination is commonly used to rapidly assess anemia during clinical examination.

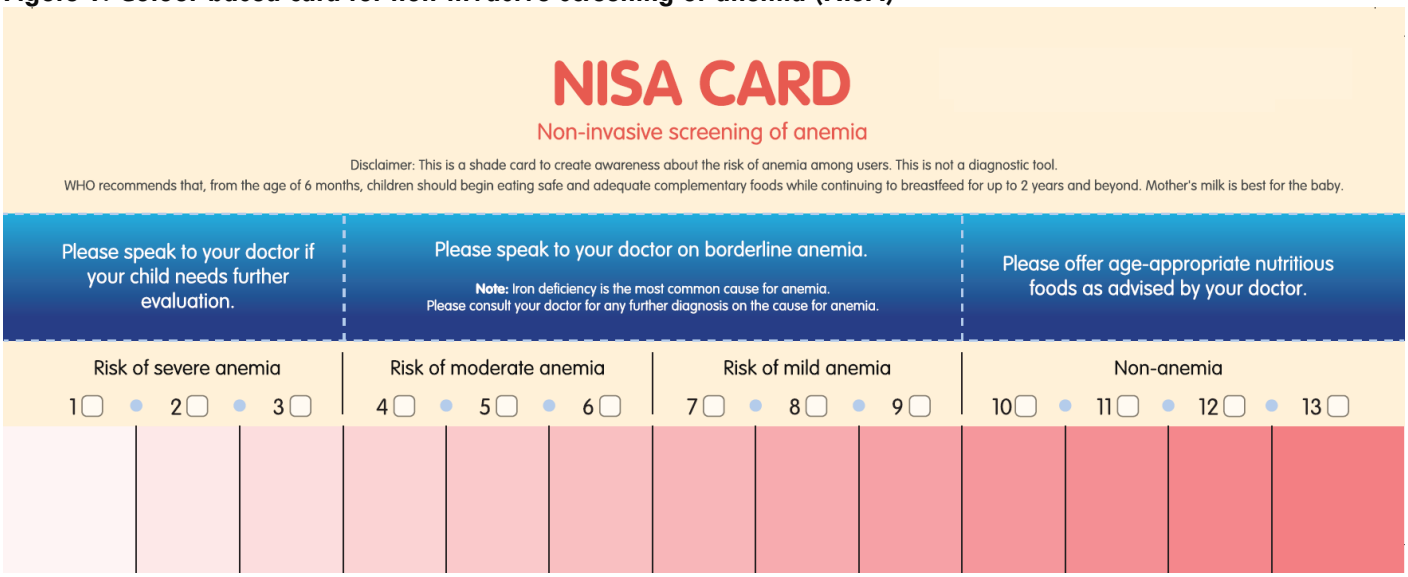
The WHO has developed a haemoglobin colour scale (HCS) for early risk assessment of anemia. A study in 194 pregnant women yielded a sensitivity and specificity of 70.9% and 49.1%, respectively for the HCS. This easy-to-use point-of-care tool has a high accuracy for the diagnosis of severe anemia. When used with clinical assessments, the HCS can significantly reduce the misdiagnosis of anemia in primary healthcare settings.[10,11] India is a geographically diverse country and varied skin tones across regions may impact the clinical impressions of anemia since it is assessed clinically, by visual inspection for the presence of significant pallor in the skin, nail beds, and palms. The conjunctiva provides a dependable estimation for presence of anemia independent of skin pigmentation. Validation of colour-based shade cards based on conjunctival pallor examination is key to screening of anemia.

We have deployed a non-invasive method to screen children for anemia by using a colour-based card (Figure 1). The Non-Invasive Screening of Anemia (NISA) shade card was developed using the 13 ordinal shade guide reported by Chowdhury et al. The shade spectrum represents the conjunctival colour ranging from no anemia (Colour code: 13; deep pink) to severe anemia (Colour code 1: paper white).[12] The shades were developed by mixing specific proportions of colour on Microsoft PowerPoint 97 and printed at a resolution of 1440 × 720 dpi. The reference for the NISA shade card was Hb level estimated by using an automated blood analyser (Gold standard test). The NISA card was used to match the colour shades in the card to the colour of the conjunctiva. With 13 shades, the card allows screening for mild, moderate, and severe categories of anemia (Figure 1).

This screening is the extended phase of the previously published report of the experience of 9 paediatricians who used the card in 173 children.[13] Further, we used a questionnaire-based survey to gather real-world experience for the use of the NISA card and assess if skin tone had a possible impact on the risk assessment of anemia.

The goal of gathering this experience was to determine the accuracy, specificity and sensitivity of the colour-based tool for NISA in infants and children and to assess opinions of paediatricians for the use of the tool.

Figure 1: Colour-based card for non-invasive screening of anemia (NISA)



Materials and Methods

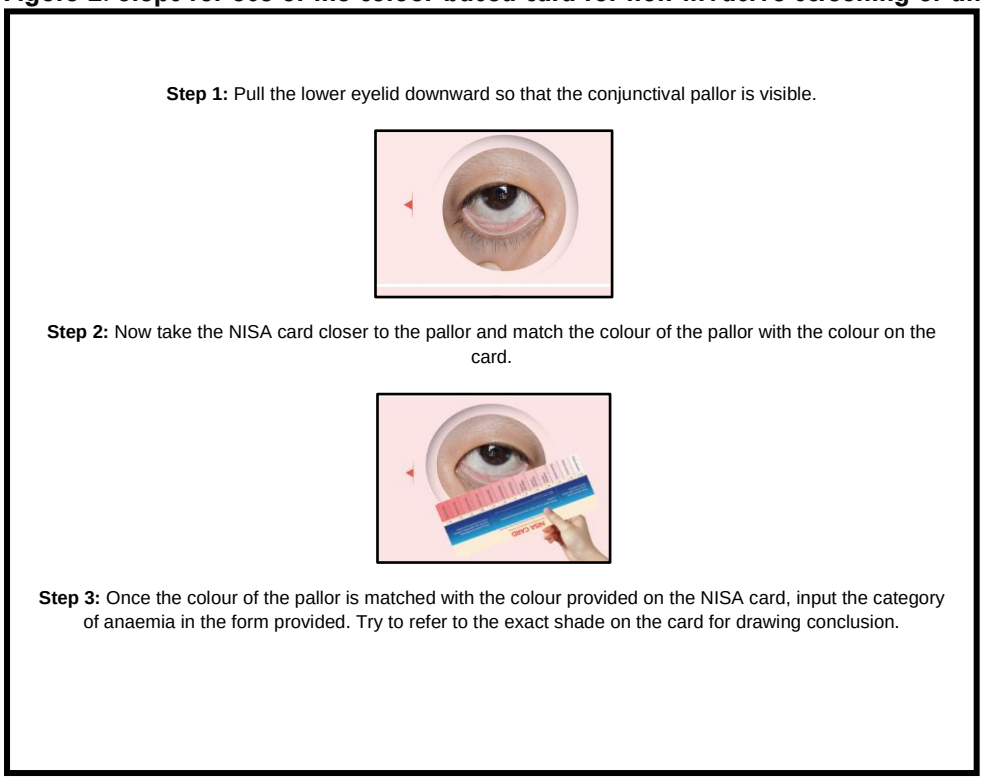
This real-world experience for the use of the NISA card included infants and children (aged 6-60 months) diagnosed with anemia in 86 outpatient clinics across India: North (n=10), South (n=37), East (n=20), and West (n=19). From Mar 2023 to May 2023, 84 physicians identified children who had undergone a recent (<15 days) laboratory hemoglobin test and categorized them for the severity of anemia : mild (10-10.9 g/dl), moderate (7-9.9 g/dl), and severe (<7 g/dl).[1]

Clinicians were educated on the use of NISA cards to match conjunctival pallor with color shades on the cards. They were asked to use NISA card to detect anemia and, if present, to grade its severity before conducting clinical history and taking physical examination. Anonymized data from children who had undergone laboratory hemoglobin tests within the last 15 days were considered and categorized by severity of anemia : mild (10-10.9 g/dl), moderate (7-9.9 g/dl), and severe (<7 g/dl).
As a next step, the physicians completed a 6-item questionnaire assessing their experience of using the NISA card as a screening tool.

SCREENING TOOLS

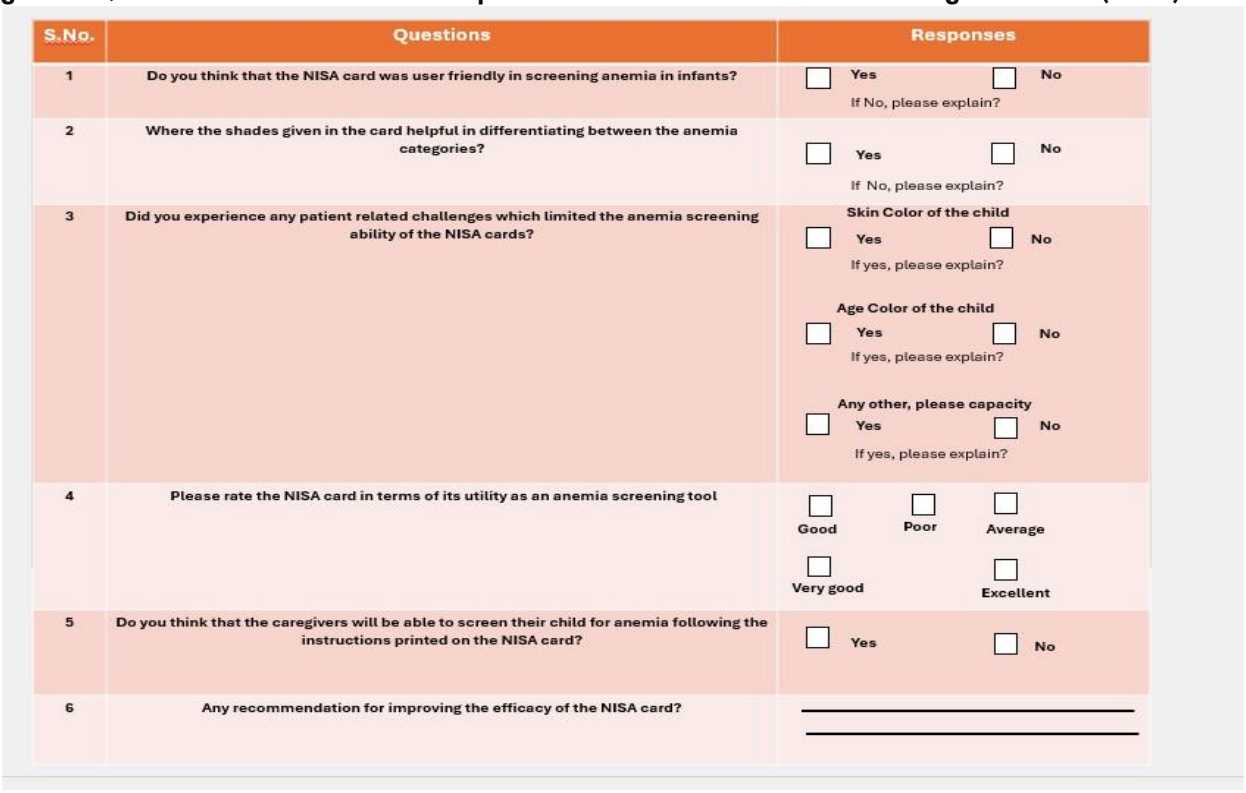
The three key steps for the use of the NISA card, a simple point-of-care tool, are shown in Figure 2. The details of methodology for gathering insights through NISA, was mentioned in the previous phase of this publication.[13]

Figure 2: Steps for use of the colour-based card for non-invasive screening of anemia (NISA)



Clinicians who used the NISA card were invited to fill another questionnaire to obtain user experiences for the ease of use, utility, severity categorization, challenges encountered in the use of the card, and any recommendations (Figure 3).

Figure 3: Questionnaire to record user experiences of the non-invasive screening of anemia (NISA) card



S.No.	Questions	Responses
1	Do you think that the NISA card was user friendly in screening anemia in infants?	☐ Yes ☐ No If No, please explain?
2	Where the shades given in the card helpful in differentiating between the anemia categories?	☐ Yes ☐ No If No, please explain?
3	Did you experience any patient related challenges which limited the anemia screening ability of the NISA cards?	Skin Color of the child ☐ Yes ☐ No If yes, please explain? Age Color of the child ☐ Yes ☐ No If yes, please explain? Any other, please capacity ☐ Yes ☐ No If yes, please explain?
4	Please rate the NISA card in terms of its utility as an anemia screening tool	☐ Good ☐ Poor ☐ Average ☐ Very good ☐ Excellent
5	Do you think that the caregivers will be able to screen their child for anemia following the instructions printed on the NISA card?	☐ Yes ☐ No
6	Any recommendation for improving the efficacy of the NISA card?	_____ _____

The Hb level estimated using an automated blood analyser was considered the gold standard for the comparison of the results obtained from using NISA card. For the NISA card, the true positive (TP), true negative (TN), false positive (FP), and false negative (FN) results were recorded. To determine and compare the appropriateness of the diagnostic tools, specificity, sensitivity, positive predictive values (PPV), negative predictive values (NPV) positive likelihood ratios, negative likelihood ratios were calculated using standard formulae.[14] Accuracy of the diagnostic tests was calculated using: TP+TN/TP+TN+FP+FN. Components of the confusion matrix were defined as the following:

- True Positive: Assessed as anemic using the gold standard test and assessed as anemic with the tool used
- True Negative: Assessed as non-anemic using the gold standard test and assessed as non-anemic with the tool used
- False Positive: Assessed as non-anemic using the gold standard test and assessed as anemic with the tool used
- False Negative: Assessed as anemic using the gold standard test and assessed as non-anemic with the tool used

STATISTICAL METHODS

Summary data were presented as mean ± standard deviation (SD) for continuous variables and numbers and percentages for categorical variables. Region-wise diagnostic were reported. Bar diagrams are presented for graphical representation. Odds ratio (95% confidence intervals) was determined for the possibility of diagnosis of anemia with the use of NISA when compared to that without the use of NISA. All analyses were performed using SPSS Version 25.

Results

A total of 1459 children from different regions of India were screened for anemia using the NISA cards. Table 1 shows the demographic characteristics of children from various regions in India. The mean Hb concentration (laboratory testing) was 9.8 ± 2.0 g/dl. Across regions, the mean Hb concentration was 9.75 ± 1.8 g/dl (East), 9.89 ± 1.5 g/dl (West), 8.80 ± 2.0 g/dl (North), and 10.25 ± 1.9 g/dl (South).

Severe anemia was detected in 6.6% (n=97) children while 42.2% (n=616) and 21.5% (n=314) children had moderate and mild anemia, respectively. Across regions, the percentages of mild, moderate, severe, and non-anemia are mentioned in Table 1. Anemia was detected in Laboratory test revealed that 70.4% of the children were anemic. While using NISA card, 62% children were found to be anemic.

Table 1: Demographic characteristics

	East India (n=270)	West India (n=298)	North India (n=289)	South India (n=602)	Total (N=1459)
Age*, months	25.3 ± 16.3	24.9 ± 15.7	22.6 ± 13.8	24.9 ± 15.8	24.5 ± 15.5
Female, n	113	122	128	282	645
Male, n	157	175	161	320	813
Hemoglobin (g/dl)*	9.75 ± 1.8	9.89 ± 1.5	8.80 ± 2.0	10.25 ± 1.9	9.8 ± 2.0
Anemic **, n(%)	204 (75.5%)	217 (72.8%)	247 (85.4%)	359 (59.6%)	1027 (70.4%)

Mild anemia**, n (%)	75 (27.8%)	68 (22.8%)	43 (14.8%)	128 (21.2%)	314 (21.5%)
Moderate Anemia**, n (%)	108 (40%)	137 (45.9%)	165 (57.0%)	206 (34.2%)	616 (42.2%)
Severe anemia**, n (%)	21 (7.8%)	12 (4%)	39 (13.5%)	25 (4.1%)	97 (6.6%)
Non-anemic*, n (%)	66 (24.4%)	81 (27.1%)	42 (14.5%)	243 (40.3%)	432 (29.6%)

*Mean ± standard deviation; **As per the laboratory reports

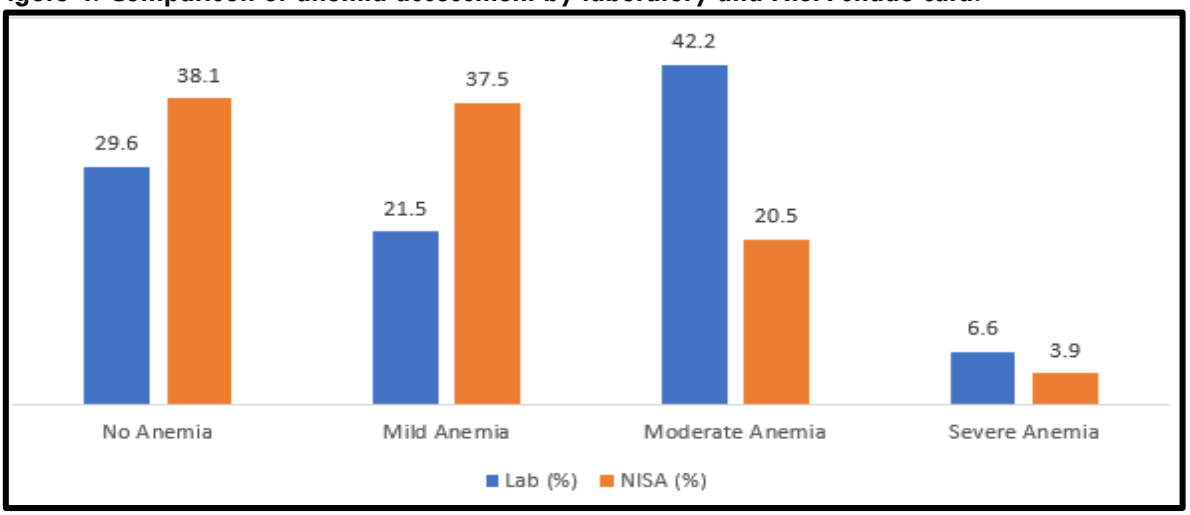
When screened for severity of anemia with the NISA card, mild anemia (37.6%) was the most common, followed by moderate (20.5%) and severe (3.9%) anemia (Table 2). When screened by laboratory assessment, mild, moderate, and severe anemia was detected in 21.5%, 42.2%, and 6.6% children, respectively.

Table 2: Severity of anemia among children screened with the non-invasive screening of anemia (NISA) card

Closest colour shades matching the conjunctiva		Severity of anemia	
No. on the NISA card	n (%)	Category	n (%)
1	3 (0.2%)	Severe	57 (3.9%)
2	20 (1.4%)		
3	34 (2.3%)		
4	44 (3.0%)	Moderate	299 (20.5%)
5	94 (6.4%)		
6	161 (11.0%)		
7	144 (9.9%)	Mild	548 (37.6%)
8	176 (12.1%)		
9	228 (15.6%)		
10	227 (15.6%)	Non-anemic	555 (38.0%)
11	168 (11.5%)		
12	133 (9.1%)		
13	27 (1.9%)		

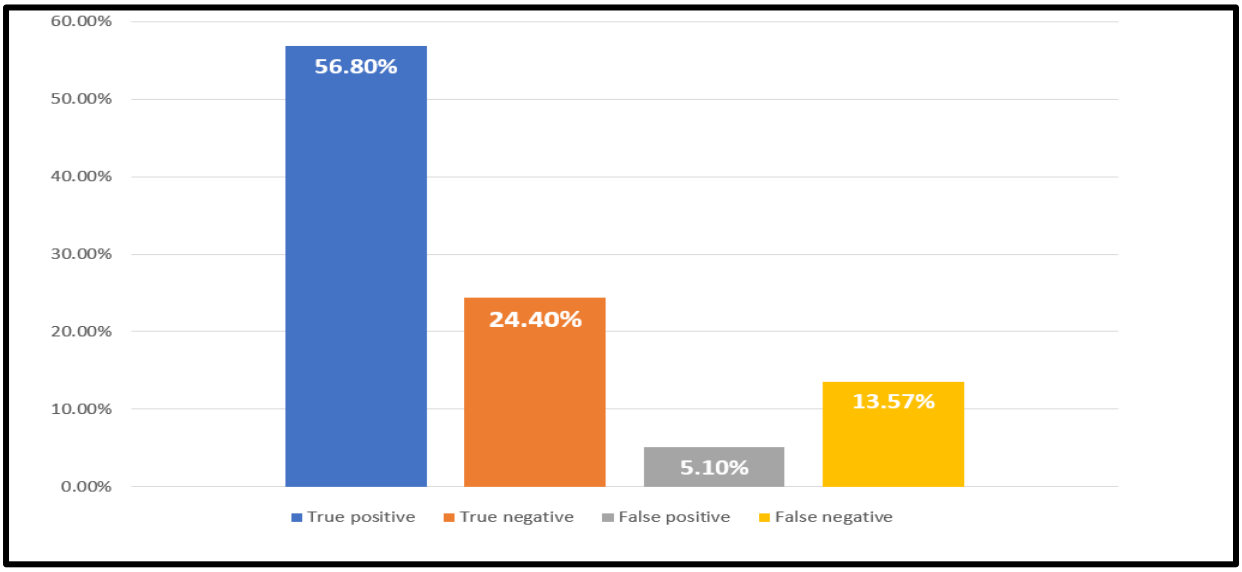
Figure 4 illustrates the comparisons of presence and severity of anemia states by laboratory and NISA card assessments.

Figure 4: Comparison of anemia assessment by laboratory and NISA shade card.



Using the laboratory assessments as reference, the NISA card assessments for anemia yielded a TN rate of 24.40%. Rates of TP, FP, and FN results were 56.80%, 5.10%, and 13.57%, respectively (Figure 5).

Figure 5: Accuracy of anemia assessment by laboratory and non-invasive screening of anemia (NISA) card



The NISA card had an accuracy of 81.28% and this ranged from 77.74% to 85.23% in different regions across India.The sensitivity and specificity of the NISA card for the diagnosis of anemia were 80.83% (95% CI:72.64% to 87.44%) and 66.04% (95% CI:51.73% to 78.48%), respectively (Table 3).

Table 3: Level of confidence for the non-invasive screening of anemia (NISA) card assessments of anemia across India

	East India	West India	North India	South India
Sensitivity	79.41% (73.21% to 84.74%)	84.79% (79.31% to 89.29%)	87.45% (82.66% to 91.31%)	74.37% (69.53% to 78.81%)
Specificity	89.39% (79.36% to 95.63%)	86.42% (77.00% to 93.02%)	64.29% (48.03% to 78.45%)	82.72% (77.36% to 87.25%)
Positive likelihood ratio	7.49 (3.70 to 15.14)	6.24 (3.59 to 10.85)	2.45 (1.63 to 3.68)	4.30 (3.25 to 5.70)
Negative likelihood ratio	0.23 (0.17 to 0.31)	0.18 (0.13 to 0.24)	0.20 (0.13 to 0.29)	0.31 (0.26 to 0.37)
Positive predictive value	95.86% (91.65% to 98.32%)	94.36% (90.13% to 97.15%)	93.51% (89.52% to 96.32%)	86.41% (82.07% to 90.03%)
Negative predictive value	58.42% (48.18% to 68.14%)	67.96% (58.04% to 76.82%)	46.55% (33.34% to 60.13%)	68.60% (62.95% to 73.87%)
Accuracy	81.85% (76.73% to 86.26%)	85.23% (80.69% to 89.06%)	84.08% (79.35% to 88.11%)	77.74% (74.20% to 81.00%)

Data are values (95% CI)

User Experience

Eighty paediatricians responded for the feedback survey. Most (91.7%) of the paediatricians agreed that the NISA card was user-friendly for screening of anemia in children. A rating of 'Good' to 'Excellent' was given by 87.5% of the paediatricians for the NISA card. The respondents considered colour shades in the card helpful for categorizing the intensity of anemia and 91.7% considered the printed instructions on the NISA card to be beneficial to caregivers for screening children for anemia.

On asking for recommendations for improving the efficacy of the NISA card, the paediatricians suggested to add additional parameters including the colours of palms, tongue, and soles in the NISA card. About 79% physicians considered the NISA card to be useful in risk screening irrespective of the skin tone in children. Others considered dark skin tone to influence the assessment of severity in anemia and a discrepancy in NISA card and laboratory assessments in children. Some physicians (Around 18%) considered younger age of children as a challenge for the use of the NISA card as infants and toddlers were non-cooperative. Others (Around 6%) mentioned that the screening with NISA tool could be doubtful in cases of jaundice, conjunctival congestion, infections, and fever.

Discussion

We present real-world experience of paediatricians for the deployment of the colour-based NISA card for the detection of anemia in children. The use of the NISA card for risk assessment of anemia yielded overall, mild, moderate, and severe anemia in 62%, 37.6%, 20.5%, and 3.9% children, respectively.

In this extended phase-2 validation, the NISA card had an accuracy, sensitivity, and specificity of 81.28%, 80.83%, and 66.04%, respectively for the detection of anemia. In the earlier phase 1 validation report, for the experience of 9 paediatricians for the use of NISA cards in 173 children, we observed an an accuracy, sensitivity, and specificity of 76.30%, 80.83%, and 66.04%, respectively.[19] This difference in accuracy, sensitivity & specificity could be due to the difference in the sample size between both phases of the NISA card validation.

At the community level, the most commonly used method for the screening of anemia in children is a clinical assessment of pallor.[15,16,17] Ugashoro et al. found that Home-Base anemia -screen tool (HB-Anae) significantly improved parents' ability to detect pallor in anemic children, nearly matching the accuracy of healthcare workers.[18] Home based anemia screening tool can be beneficial where parents generally lack knowledge about anemia , leading to its detection primarily during hospital visit.

The sensitivity of another 13-shade color scale (with clinical examination and medical history) comparing conjunctival pallor to HemoCue, for determination of anemia was 82.9% and the specificity for absence of anemia was 90.9%.[12]

The NISA card, with a Positive Predictive Value of 86.41% to 95.86% and a Negative Predictive Value of 46.55% to 68.60%, is a good tool for risk assessment of anemia in children. For the NISA card, the positive likelihood ratio for the assessment of anemia ranged from 2.45 to 7.49. Similarly, a likelihood ratio of 4.49 is reported for Hb <9 g/dl in the presence of pallor in another prospective study in 302 patients.[8] Systematic review of 14 studies from Africa has also reported that colour shades card are a promising tool for diagnosing anemia where there is no laboratory access.[19]

Laboratory assessments of Hb levels are commonly used for screening and treatment of anemia. The available tools for the determination of anemia require the use of a physical equipment or laboratory settings and require blood samples for testing. Accuracy of these tools and tests have been reported in various studies in literature.[20] McLennan and Steele have reported the use of complete blood count for the screening for anemia and treating it with iron supplements in a low-resource community in the Dominican Republic.[21] These laboratory tests need infrastructure and expertise and are associated with a cost.

Several non-invasive tools are existing which are used to measure the haemoglobin in blood and other tissues. These include fluorescence spectroscopy, photoplethysmography, reflectance spectroscopy, and others. Non-invasive digital methods for detecting anemia involve analyzing the color of the conjunctiva in images to measure hemoglobin levels. These techniques use advanced image processing and machine learning algorithms to accurately determine hemoglobin concentration based on the conjunctiva's color, providing a quick and painless alternative to traditional blood tests.[22–24]

These methods are not affordable to many in resource limited geographies and are available either as portable or wearable technologies.[25,24] Thus, an easy-to-use, readily available, simple non-invasive method is needed as a substitute to laboratory tests for the screening of anemia in a larger population. The WHO-HCS is a useful non-invasive screening tool that can be used for widespread screening of anemia at the population levels.[10,11]

We have reported the experience and outcomes for the use of the NISA card in four geographical regions of India. In a culturally diverse country like India, this helps to describe the risk screening for anemia in children with different skin tones and regional variations. Having said that community screening, detection of anemia in tertiary care settings is equally important.[8] Our data suggest that the colour-based shade card may be a suitable option as a risk assessment tool for anemia in children, specifically so in limited-resource settings, at the community levels with ease and convenience. This implies inclusivity for children from different socio-economic groups. This tool can enable planning and execution of decisions and strategies for optimal nutrition in children. It can be used to strengthen the understanding of feeding practices in infant and young children and generate awareness on nutritional requirements early in life in children.

In clinical settings, the NISA card may be supplemented with the medical history and general physical examination to assess the risk of anemia and deploy the appropriate strategies for its prevention. The NISA card is a simple, rapid, portable, accurate and reliable, inexpensive tool that could be deployed at grass root level as support to reduce the risk of anemia by generating the awareness on importance of nutrition.

To further improvise the tool, we have gathered experiences for the use of the NISA card in physician interviews in the current study as well the earlier report for 9 paediatricians.[13] Most paediatricians (87.5%) rated the NISA card as 'Good' to 'Excellent' and >90% of them considered it to be user-friendly for community settings.

The applicability of NISA card in a larger population and enhancing its usage with clinical examination at various levels of healthcare systems need public-private partnership in various government-initiated programs meant for reducing the prevalence of anemia. The NISA card finds applicability in early risk assessment as well as in early intervention options for prevention of anemia and also in educating the masses for nutritional requirements and feeding practices for healthy growth and development of children.

Limitations

Limitations of the study include the lack of data for the ethnicity of study participants. Only conjunctival pallor was assessed to screen for anemia. Nailbeds and palms were not assessed for pallor. Some of the challenges with the use of the NISA card included the possibility of misdiagnosis in conjunctival discoloration in presence of other reasons and congestion and the difficulty of use in non-cooperative young children. The influence & impact of skin tone on conjunctival pallor in the real-world, needs further well-designed randomized clinical trials for conclusive outcomes. The concerns for the use of the card included the non-utility in children with infections and jaundice when the conjunctival discolorations can limit the use of the card.

Conclusion

In conclusion, the timely risk assessment is an important aspect in preventing anemia. In this extended survey for physician-experience with the NISA card, the utility of the card was confirmed for early risk assessment of anemia

in larger populations across geographical regions in a culturally diverse country like India. The NISA card was used in community as well as tertiary care settings and this suggests a wider applicability of the tool in various socio-economic segments. The sensitivity, specificity, and accuracy of the NISA card for the diagnosis of anemia were 80.83%, 66.04%, and 81.28%, respectively. The real-world evidence of NISA in 2 publications suggests its usage in the need and importance of generating awareness on optimal feeding practices and nutrition in supporting overall growth and development of young children.

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The named author meets the International Committee of Medical Journal Editors (ICMJE) criteria for authorship for this manuscript, takes responsibility for the integrity of the work, and has given final approval for the version to be published. The authors thank Dr Punit Srivastava of Mediception Science Pvt Ltd (www.mediception.com) for supporting the preparation of the manuscript. The group of Health care practitioners contributing to the conduct of this survey is mentioned in Annexure.

Conflict of interest disclosure: None

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Annexure

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