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REVIEW

Newborn screening for primary congenital hypothyroidism: past, present and future

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Abstract

This manuscript reviews the evolution of newborn screening for primary congenital hypothyroidism (CH) and explores future strategies to enhance diagnostic accuracy. Over the past few decades, newborn screening has expanded globally, significantly reducing the incidence of severe forms of the disease. However, challenges persist, especially regarding the overdiagnosis of mild cases of primary CH, which may not require treatment. Omic sciences may help researchers to enhance the understanding of primary CH and to uncover new biomarkers to identify mild cases with altered proteomic and/or metabolic profiles associated with the need for treatment. Record-linkage studies can help deepen knowledge on the long-term outcomes of affected children identified through newborn screening. Nevertheless, despite 50 years of newborn screening for primary CH, a minority of newborns currently benefit from this critically important public health intervention. Efforts should be done to expand access to newborn screening globally, especially for those born in developing countries.

Keywords: primary congenital hypothyroidism; newborns; neonatal screening; prevention

Introduction

Primary congenital hypothyroidism (CH) is one of the most common preventable causes of adverse neurodevelopmental outcomes. It is a complex condition present at birth, resulting in severe to mild thyroid hormone deficiency and may be caused by abnormal development (dysgenesis) or dysfunction of the thyroid gland (*in situ* thyroid). There is a female predominance among cases with thyroid dysgenesis and a similar sex incidence among those with a normally shaped and located thyroid (1, 2, 3). Among these, primary CH may be permanent or transient.

Optimal management of primary CH requires early diagnosis and prompt treatment to avoid severe neurocognitive sequelae (4). This goal has been achieved by newborn screening (NBS), which represents one of the most important results of preventive medicine in childhood.

NBS for CH began in Quebec and Pittsburg. Specifically, in 1973, Jean Dussault developed a radioimmunoassay to measure T4 in dried blood filter paper specimens and then applied it to screening newborns for CH (5). In 1975, Dussault reported the results of screening 47,000 newborns in Quebec, detecting seven cases of CH (~1:7000) (6). In 1978, Fisher and collaborators reported the detection of 277 patients with CH in 1 million infants screened in North America, an incidence of 1:3684 (7). Since then, universal NBS for CH has been implemented in many countries around the world.

Here, we present how NBS for primary CH has evolved over time in terms of priorities, screening technology and

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populations screened. Considerations on the future development of NBS for primary CH are also discussed.

The priority of NBS for primary CH

In 1968, Wilson and Jungner published 'Principles and Practice of Screening for Disease', a fundamental work that identified ten principles that should be considered when making a screening decision (8). Although these principles (Table 1) referred to population-based screening in general and not to NBS in particular, they have been widely adopted for NBS policies.

At that time, it was already known that neurocognitive impairment due to CH was associated with the severity of hypothyroidism at birth and the age of initiation of replacement therapy (9, 10) and that improved developmental outcomes could justify the economic costs of mass NBS programs for CH (11, 12). However, CH did not meet the Wilson-Jungner criteria until the late 1970s when a validated screening test first became available.

Now, as then, the main objective of NBS for CH is the eradication of cognitive disability resulting from severe CH. However, the lesson learned in five decades of NBS for CH is that primary CH covers a spectrum of phenotypes, including mild, moderate and severe forms of hypothyroidism (4, 13). In 2014 the international guidelines promoted by the European Society for Paediatric Endocrinology stated that the priority of NBS for CH should be the detection of all forms of primary CH: mild, moderate and severe (14). This concept was also confirmed in the most recent ENDO-European Reference Network (ENDO-ERN) consensus guidelines on the diagnosis and management of CH (13).

Although both ESPE and ENDO-ERN guidelines confirm that the most sensitive test for detecting primary CH is the measurement of thyrotropin (TSH), ENDO-ERN guidelines also recommend adding the measurement of total or free thyroxine (fT4) to TSH to screen for central CH when financial resources are available. This recommendation, aimed at preventing the neurodevelopmental sequelae of neonatal thyroid hormone deficiency present at birth (13, 15), is also based on evidence that the reported incidence of central CH detected through neonatal screening is higher than that thought in the pre-screening era (1:100,000) and varies between 1:30,000 and 1:16,000, based on the screening strategy (16, 17, 18).

Similarly, the American Academy of Pediatrics recently reported that although the detection of infants with moderate or severe primary hypothyroidism is the chief priority of NBS programs, secondary priorities include the detection of infants with mild primary hypothyroidism, primary hypothyroidism of delayed onset ('delayed TSH elevation'), or central hypothyroidism (19).

Screening tests and strategies to screen for primary CH

As already mentioned, the first available test to screen for CH was the measurement of T4 from the eluate of filter paper blood samples (6, 7). However, at that time, it was already known that raised TSH had the advantage of being a more sensitive test of thyroid insufficiency, which is capable of detecting the disease even when the level of circulating thyroid hormones had not yet changed (20, 21, 22). Therefore, the measurement of TSH from the eluate of dried blood spots (DBS) was rapidly set up (23, 24). Furthermore, in 1977, Delange and colleagues suggested that a better separation between negative and positive results could be obtained with TSH measured on the 5th day of life because the variability of individual results

Table 1 Wilson and Jungner's principles of screening (8).

Concepts	Principles
Condition to screen	
	1 The condition should be an important health problem
	2 The natural history of the condition should be adequately understood
	3 There should be a recognizable latent or early symptomatic stage
Test used to screen	
	4 There should be a suitable test or examination
	5 The test should be acceptable to the population
Patients to screen and treatment	
	6 There should be an agreed policy on whom to treat as patients
	7 There should be an accepted treatment for patients with recognized disease
	8 Facilities for diagnosis and treatment should be available
Screening program features	
	9 The cost of case-finding (including diagnosis and treatment of patients diagnosed) should be economically balanced in relation to possible expenditure on medical care as a whole
	10 Case-finding should be a continuing process and not a 'once and for all' project

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on the 5th day was smaller than that observed at birth when measuring TSH in cord blood (25). Since then, TSH measurement in the first week of life has been progressively adopted in many screening programs worldwide.

Because most cases of CH are caused by abnormalities in thyroid gland formation or thyroid hormone synthesis, NBS strategies are designed to detect elevated levels of TSH and/or decreased concentrations of thyroxine (T4). Currently, TSH is used as the primary screening test in Canada, in more than 50% of the states in the US, and in most other countries where an NBS program for CH is active (19, 26, 27). In some NBS programs for CH, T4-TSH backup-based or T4+TSH-based strategies are also used (26). Only the Netherlands has chosen a more complex approach, which is T4-based with a TSH and TBG backup. This screening strategy is particularly effective in detecting cases of central CH (17).

It is worth noting that strategies employing primary-TSH testing or primary-T4 testing (with reflex TSH) have similar accuracy in detecting severe forms of primary CH, which show the greatest neurodevelopmental risk (28). However, while the primary-TSH strategy can detect forms with mild increases in TSH at birth (with normal T4) (29, 30, 31), either strategy employing primary-TSH or primary-T4 testing may be less sensitive for detecting specific subgroups of infants with delayed increase in TSH, particularly those born preterm or with low birth weight (LBW), twins and infants admitted to neonatal intensive care units (NICUs) (4, 32). For these infants, re-screening at 2–4 weeks of age is recommended (13). This issue is widely discussed in the sections below.

Changes in the neonatal population screened for CH

Over recent decades, the profile of newborns screened for primary CH in developed nations has undergone significant changes, with an increase in preterm and multiple births and infants admitted to NICU (33, 34, 35, 36).

From 1980 to 2014, there was an increase in preterm births (<37 weeks gestation), so that in 2014 approximately 10.6% of all births worldwide were preterm (37). It has been reported that the primary explanation for this increase globally may be changes in the registration of live births because previously many preterm infants were not recorded as live births unless they survived the first day of life (37). In the past decade, little changes have been observed in global preterm birth rates; however, some differences have been reported among countries, with the US reporting a 12% increase in preterm births from 2014 to 2022 (38). This increase can be attributed to various factors, including an increase in maternal age. Although both younger and advanced maternal age are associated with preterm birth (36, 39), in recent years, mothers aged over 40 years have the highest rates of preterm births (38).

Since the 1980s, twin pregnancy rates have also increased, particularly in high-income or middle-income countries (40, 41, 42, 43). It has been reported that twinning rates were recently peaking at a historical high, with rates of over 15 twin deliveries per 1000 deliveries in the USA, Canada, and the European Union. Only the poorer regions of Latin America and in South and Southeast Asia had lower twinning rates, often (well) below 10 twin deliveries per 1000 deliveries (35). While the important role of medically assisted reproduction is undisputed (44), also the rise in the age at childbearing, especially in high-income countries, has contributed to increased twinning rates (45).

Concerning critically ill infants, in the last decades, neonatal medicine has seen an increase in adopting better clinical practices to decrease complications and increase survival without major disability in these infants (46, 47). These improvements have led to a higher incidence of admissions to NICU in the USA (48) and Europe (49), subsequently increasing the number of sick infants who are now screened but who would have died in the past (50).

Impact of changes in screened population on NBS for CH

Babies with primary CH who are born premature, as twins or with low birth weight, or who are sick in the neonatal period, may not show an adequate TSH response in the first weeks of life (33, 34, 51). Causes of this late rise in TSH include one or a combination of the following factors: developmental delay in the maturation of the hypothalamic–pituitary–thyroid (HPT) axis (52); exposure to medications frequently used in the intensive care setting that can suppress TSH concentrations, such as dopamine and glucocorticoids (53, 54); susceptibility to the thyroid-suppressive effects of iodine in topical antiseptics, especially in iodine-deficient areas (55, 56); recovery from sick euthyroid syndrome (57, 58, 59); and fetal blood mixing in monochorionic monozygotic twins (60).

Preterm infants exhibit a unique and dynamic pattern of thyroid hormone levels, which can complicate the diagnosis of primary CH. Besides the immaturity of the HPT axis, the withdrawal of maternal thyroxine (T4) after birth, exposure to iodine, especially in iodine-deficient areas, medications, and the persistence of fetal metabolism influence thyroid hormone physiology in these infants (61). Being small for gestational age has been strongly associated with higher TSH levels at screening and follow-up (62, 63). In addition, transient hypothyroxinemia, which is due to immature HPT function, is a frequent finding in these infants and is often aggravated by general illness in the preterm neonate (64, 65, 66).

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Based on these observations, screening results in these special categories of infants may be false negative either in TSH-based neonatal screening programs or in primary-T4 testing (67, 68, 69). To avoid missing cases among neonates at risk of delayed increase in TSH, ENDO-ERN guidelines recommend repeat sampling at 2 weeks of life or 2 weeks after the first screening test was carried out in the following situations: preterm birth, LBW and very low birth weight neonates, ill and preterm newborns admitted to NICU, specimen collection within the first 24 h of life, and multiple births (13). However, some US and Canadian programs also adopt routine repeat DBS collection and screening at 2 weeks of life (19).

Identifying delayed TSH rise is clinically important because it may be associated with overt hypothyroidism at diagnosis. Particularly, a study conducted on 333 infants (preterm and term infants) admitted to NICU and showing delayed TSH rise on second screening (TSH >15 mU/L) reported that 58% of these infants required levothyroxine treatment (70). Another recently published study found that 33% (15/45) of preterm infants with delayed TSH rise had TSH concentrations >100 mIU/L at diagnosis, and all infants with confirmatory serum TSH >20 mIU/L also showed low serum free T4 (71).

The impact of NBS on the incidence of primary CH

Changes in testing techniques and improvements in laboratory medicine

When NBS for CH was introduced, radioimmunological assays (RIAs) were used to measure both T4 and TSH (25). Over the years, laboratories moved towards the measurement of TSH as the primary screening test and progressively adopted more sensitive immunoradiometric assays (IRMAs) as testing techniques (72). Currently, most TSH test kits for DBS specimens are immunometric assays that use fluorescence labels (fluorescence immunoassay, FIA; time-resolved fluorescence immunoassay, TRFIA), although kits using colorimetric (enzyme-linked immunosorbent assay, ELISA) or radiometric labels (IRMA) are also commercially available (73).

Over the years, laboratory medicine has also improved and accurate methods to calculate cutoffs based on reference intervals of neonatal TSH have been progressively adopted (74, 75). In fact, to establish cutoffs, the screening laboratory must first establish a reference range for the disease marker(s) used. The reference range for the non-normally distributed TSH variable is generally calculated as values included between the 2.5th and 97.5th percentiles and the TSH cutoff is roughly set at the 99th percentile of the distribution (74).

It is worth noting that the selection of screening cutoffs is based on a balance of clinical sensitivity (i.e. a high rate of case detection) and clinical specificity (i.e. an acceptable recall rate) and that all screening programs might miss cases. Therefore, selecting relatively low TSH cutoff values reduces the number of false-negative screens (i.e. missed cases), although it increases the recall rate, which in turn increases costs and workload for the screening program. Some authors have underlined the negative impact of false-positive screening results on families (76), whereas others have suggested that parental stress and anxiety can be reduced with improved education and communication to parents, specifically at the time of follow-up screening (77).

Another factor to take into consideration when choosing cutoffs is that TSH (as well as T4) levels decrease with the age of the infant. In healthy term infants, birth stress causes a physiological increase in TSH concentrations to 50–80 mIU/L serum within 30–60 min after delivery, it remains elevated (10–15 mIU/L serum) for 24–48 h, and then declines to <10 mIU/L serum by 1 week of age (78). It is therefore important to adopt appropriate age-adjusted TSH cutoffs to avoid erroneous results, with the potential for missed cases or a too-high recall rate (27, 73, 79, 80). This issue has become particularly relevant with the introduction in many countries of expanded NBS programs for rare metabolic diseases. In fact, some of these diseases are time-critical and require the earliest diagnosis to avoid mortality, therefore causing a reduction of the time window to screen. For example, in Italy, the introduction in 2016 of the expanded NBS program reduced the age at screening from 3 to 5 days to 48–72 h of life (81), with implications for the calculation of the TSH cutoff.

Lowering TSH cutoff at screening and re-screening

Starting with 50 mU/L TSH blood as the cutoff in the early programs, which were able to identify severe cases of primary CH, TSH cutoff points at screening have been lowered over time to improve detection rates and because of changes made to the assay methods (30, 82, 83, 84, 85).

With the introduction of re-screening procedures in special categories of infants at risk of delayed rise of TSH and because of TSH and thyroid hormone levels change significantly within the first weeks after birth, in some 2-screen programs it has been shown the benefit of using differential TSH cutoffs, with a lower cutoff at re-screening (83, 86). In Quebec, the analysis of 20 years of screening for CH (1990–2009) showed that following an initial TSH between 15.0 and 30.0 mU/L blood, the reduction of the cutoff at the second test from 15.0 to 5.0 mU/L allowed to identify 49 additional cases on 620 diagnosed with primary CH in the period of the observation, and 10 of these 49 had thyroid dysgenesis in the form of ectopic thyroid (83). Similarly, the analysis of 8 years' experience with a 2-screen program for CH in the Lombardy region, Italy,

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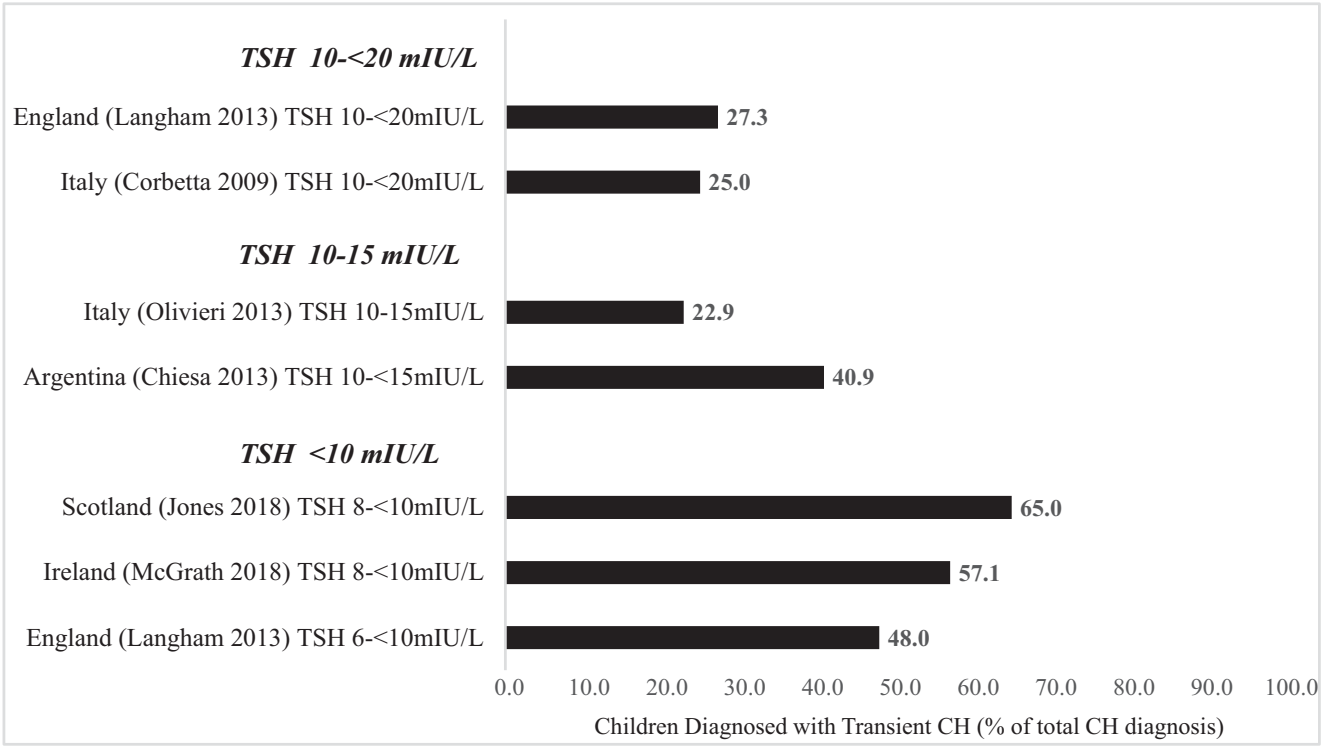

Figure 1

Percentage of children diagnosed with CH that had transient CH at different initial TSH levels.

using differential TSH cutoffs (10.0 mIU/L blood at the first screen and 5.0 mIU/L blood at the second one), has shown that 52 of 119 infants with primary CH who were identified at the second screening would have been missed if the same TSH cutoff of 10.0 mU/L blood had been used at the first and second screen. In addition, more than half of these babies showed subnormal serum FT4 levels at diagnosis, including cases with thyroid dysgenesis (86). Taken together, these findings suggest that 2-screen programs with the same screening cutoff for both screens risk missing infants because of the physiologic TSH concentration changes in relation to the age of the infant at specimen collection. This is particularly relevant in countries where the initial screening is obtained at age 24–72 h, because early timing of the first screening needs higher TSH cutoffs to compensate for the physiologic postnatal TSH surge (87, 88).

Reducing the TSH cutoff (at screening and re-screening) has caused a progressive increase in detecting additional mild forms of primary CH, mostly with normally located and shaped thyroid, although more severe cases of primary CH have been also reported (31, 69, 82, 85). Particularly, it has been shown that around 20% of infants diagnosed with primary CH and mild increases at screening (<15 mIU/L blood) are due to some forms of thyroid dysgenesis such as hemiagenesis, hypoplasia and ectopic thyroid (31, 85). However, cases of mild primary CH with eutopic thyroid frequently show transient hypothyroidism. These forms have a number of possible causes, including iodine deficiency or excess, defects involving *DUOX2* pathogenic variants, maternal antithyroid drugs or maternal TSH-blocking antibodies, and will revert back to normal thyroid function in the first few years of life (89, 90).

Studies from the UK (29, 91), Italy (31, 92), Argentina (82), Ireland (93) and Serbia (94) have examined those diagnosed with permanent and transient CH at various thresholds for the first TSH screening test. Fig. 1 shows that for all children diagnosed with CH, the proportion diagnosed with transient CH increases at lower initial TSH thresholds. Among those who had an initial TSH between 10 and 20 mIU/L blood, 23–40% of diagnoses of CH had transient CH compared to 48–65% of children diagnosed with primary CH with an initial TSH level<10 mU/L blood. The day of bloodspot collection differs between NBS programs and may impact results; however, in all studies, children were diagnosed with transient CH after a trial discontinuation of levothyroxine treatment at approximately 3 years of age.

Impact of lowering TSH cutoffs and other factors on the reported incidence of primary CH

Over the last four to five decades, increasing incidence of CH has been reported internationally (30, 31, 82, 83). The

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increasing incidence of CH can be explained by a number of factors outlined above, with the most likely explanation being the lowering of TSH thresholds by NBS programs worldwide and increased survival of preterm infants (84). Table 2 summarizes studies reporting variations in primary CH incidence in relation to lowering TSH cutoffs and adoption of re-screening procedures in special categories of infants at risk of delayed TSH rise.

NBS TSH thresholds are selected to detect as much clinically significant disease as possible while minimizing harms caused by false-positive results and possible overdiagnosis. International experts disagree on whether to lower NBS TSH levels to identify infants with mildly elevated TSH levels (95). In the late 1990s and early 2000s, a number of countries in Europe (92, 94, 96) and North and South America (82, 83) reduced TSH cutoffs. A systematic review of global CH rates from the past 50 years identified that the global incidence increased more sharply during 2001–2010 than in the previous four decades, although after adjustment for different TSH cutoffs, the incidence of primary CH significantly increased over the last 40 years (97). This indicates that changes in TSH cutoffs alone did not drive the increasing primary CH rates.

The impact of an increased number of preterm infants on incidence estimates of primary CH is more complicated. Screening for CH in preterm infants is very challenging, as their thyroid glands are in various stages of development. As aforementioned, preterm infants differ from term infants with a high risk of delayed TSH rise due to the administration of drugs known to impact thyroid hormone production and the developmental delay of the hypothalamic-pituitary-thyroid axis (14, 98). All of these issues may be particularly pertinent at lower TSH cutoffs. The lowering of initial TSH thresholds identifies more preterm infants with CH (29, 31), and, in addition, the introduction of repeat TSH screening for high-risk and premature infants identifies more children with CH (84). The increased survival of preterm infants and changes in NBS strategies both contribute to the increased incidence of CH. An Italian study found that about half of the increased incidence of CH was attributable to increased numbers of preterm infants (84). Another recent study investigating increased CH rates in China during the period 2012–2019 found an increase in primary CH incidence from 1:2493 in 2012 to 1:1733 in 2019, with a parallel increment of 38% in premature infants diagnosed with primary CH (99). This study also hypothesizes that maternal characteristics and environmental exposures may impact CH incidence. However, studies examining the impact of maternal characteristics, such as diabetes, thyroid disease or alcohol use during pregnancy, on neonatal TSH levels yield inconsistent findings (100, 101).

Ethnic differences in CH incidence have been reported, with a higher risk for primary CH in infants of primarily non-European ancestry (102). For example, Hinton *et al.* reported that between 1991 and 2000, the highest incidence of CH in the USA was found in Hispanic newborns. However, the lowest incidence of CH in US newborns in that study was found among infants of African ancestry. The prevalence of consanguinity can help to explain certain differences. In a study conducted in Italy (84), consanguinity was found to be significantly higher among African (24%), Asian (13%) and Hispanic (9.0%) parents of CH babies than among Italian or East European parents (2%). Babies born to consanguineous parents showed a significantly higher frequency of normal/hyperplastic thyroid than of thyroid dysgenesis (65 vs 35%), suggesting a high occurrence of genetically determined dyshormonogenesis.

Overall, the incidence of primary CH in preterm infants has been reported to be around 1:900 live births (67), with the highest estimate (1:50) in very low birth weight infants (107). However, it is important to underline that although the risk of transient CH is three-fold higher in preterm than in term infants, only the frequency of preterm infants with permanent CH (not transient CH) has increased over time, mostly due to the progressive adoption of re-screening procedures (84).

Identifying children using NBS TSH cutoffs is only the first stage of ensuring at-risk children are diagnosed with CH and treated. The decision to treat infants at lower confirmed TSH levels depends upon clinical judgment and varies between clinicians. A recent survey of pediatric endocrinologists in Australia and New Zealand found that extensive variation among clinical practice of initiation of treatment with levothyroxine, particularly among children with mild thyroid deficiency (103). Changes in diagnostic practices could impact CH incidence rates and should be investigated.

The future of NBS for primary CH

To optimize outcomes minimizing over- and under-diagnosis

Improving detection of true cases of primary CH, particularly reducing the over- and under-diagnosis of mild cases, is a challenge for the future of NBS for CH. First, it is crucial to establish screening reference intervals for TSH across various preterm populations. TSH percentiles at birth can differ based on multiple factors in both term and preterm newborns. This variability is evident not only during initial screenings (71) but also in subsequent samples (86), underscoring the need for tailored reference ranges. Few studies have assessed postnatal trends in thyroid hormones, corrected for gestational age, beyond the first week of life, particularly for TSH concentrations in the vulnerable

Table 2 CH incidence at different NBS TSH cutoffs in TSH based screening programs.

		NBS details				TSH cut-off (mU/L blood)			
Reference	**Country**	**Assay**	**DOC, day**	**Study exclusions**	**Year cutoffs changes**	**Initial**	**At re-screening†**	**CH incidence**	**Re-screening in infants at risk of delayed TSH rise**
McGrath *et al.* (93)	Ireland	TRFIA	3rd–5th	Excluding preterm infants and infants exposed to iodine excess in the newborn period	1976–2016	>8	No	1:2220	No
Mitrovic *et al.* (94)	Serbia	IFMA	48–72 h	-	1997	>15	No	1:6223	No
					1998–2006	>10	No	1:3893	No
					2006–2014	>9	No	1:1872	No
Silvestrin *et al.* (134)	Brazil	TRFIA	3rd–5th	-	2010–2012	>20	No	1:4137	No
						>15	No	1:3603	No
						>10	No	1:3103	No
						>5	No	1:2539	No
Botler *et al.* (135)	Brazil	TRFIA	3rd–7th	Exclude NSRC B where samples collected from children over than 7 days, the critical level was 10.0 μIU/mL. This is a follow-up TSH cutoff	2005–2007	>20	No	1:3311	No
						>10	No	1:1014	No
Deladoey *et al.* (83)	Canada	TRFIA	2nd–3rd	-	<2001	>15	No	1:2898	No
					2001	>15	>5	1:2450	No
Heather *et al.* (136)	New Zealand	TRFIA	2nd–3rd	Excluding low birth weight (<1500 g) infants	2013–2014	>30	No	1:3670	No, requested second dried spot in infants older than 14 days and with initial TSH ≥15 mU/L blood
						>25	>8	1:3174	
						>20	>8	1:2796	
						>15	>8	1:2499	
Corbetta *et al.* (92)	Italy	TRFIA	3rd*	-	<1998	>20	No	1:2654	No
					1999–2002	>12	No	1:1816	No
					2003–2005	>10	>5	1:1154	Yes, infants born at <37 weeks gestation retested between the 14th and 21st day of life
Mengreli *et al.* (30)	Greece	RIA	3rd–5th	-	2000–2002	>20	No	1:2162	Yes, infants born <37 weeks gestation and infants admitted to NICU
						>10	No	1:1557	Yes
Chiesa *et al.* (82)	Argentina	TRFIA	1.5–5th	-	1997–2002	>15	No	1:2726	Yes, only babies born at <32 weeks gestation
					2003–2010	>10	No	1:2088	Yes
Langham *et al.* (29)	UK	TRFIA	5th	Excluding infants screened in neonatal units	2006–2007	>20	No	1:2513	Yes, infants born at <32 weeks gestation
						>10	No	1:2015	Yes
						>6	No	1:1645	Yes
Anastasovska & Kocova (137)	Macedonia	TRFIA	48–72 h	-	2002–2010	>15	No	1:2489	Yes, infants born <37 weeks gestation, infants with birth weight <2500 g, infants admitted to NICU
					2011–2015	>10	No	1:1585	Yes
Teixera Palla Braga *et al.* (85)	Brazil	TRFIA	3rd–5th	-	2021–2022	>10	No	1:2229	Yes, infants born at <32 weeks gestation or with birth weight <1500 g, infants with hemodynamic instability, infants who received a transfusion before sample collection for screening
						>6	No	1:1323	Yes

RIA, radioimmunological assay; TRFIA, time-resolved fluorescence immunoassay; IFMA, two-site immunofluorometric assay; NSRC, Neonatal Screening Reference Centers; DOC, day of collection.
*Mean. †Differential TSH cut-off.

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preterm population (104). Second, establishing accurate reference intervals for thyroid hormones for the screened population that consider both gestational and postnatal age is essential for the precise diagnosis and management of thyroid dysfunction. Clinical features suggestive of thyroid dysfunction are often present in preterm infants regardless of their thyroid profile: they are often admitted to NICU with cardiac dysfunction, temperature instability, coagulation disorders, bradycardia, apnea and hypotonia. These are therefore not useful to differentiate children at risk of hypothyroidism. Particularly during a critical period of brain development, thyroid hormone ranges that consider not only gestational age at birth but also postnatal age are needed.

The decision to treat children with a certain thyroid hormone profile must incorporate the potential long-term neurodevelopmental and cognitive impairment of children with mildly elevated TSH levels. However, the question of whether these infants with mild primary CH can benefit from early treatment remains a topic of debate (95).

Outcome studies of duration and cognitive consequences of thyroid hormone abnormalities in preterm infants are scarce and confusing, as threshold selection for TSH or T4 and primary outcomes vary among studies. In neonatal hyperthyrotropinemia, a systematic review including 82% of infants who received levothyroxine did not show adverse developmental outcomes during infancy or childhood (105). Hyperthyrotropinemia in preterm infants is typically a mild and transient condition characterized by delayed TSH elevation. These infants often have known risk factors associated with this late rise in TSH levels (71, 106), leading to variable neurocognitive outcomes. Some studies indicate no long-term developmental consequences (107). A Taiwanese cohort study noted that preterm newborns with TSH levels consistently in the upper quartiles at birth and before discharge did not show an increased risk of neurodevelopmental disturbances at 2 years of age. This contrasts with infants who maintained lower TSH concentrations or whose TSH levels rose from middle to higher quartiles (108). In contrast, another small follow-up study reported that 3% of infants consistently in the top decile of gestationally age-adjusted TSH levels had a significant reduction in cognitive, motor and fine motor scores when compared with those not in the top decile (109). Conversely, in a study involving 74 preterm infants born before 32 weeks, no significant correlation between TSH percentiles from the last newborn screen and neurodevelopmental outcomes, including the Bayley-III cognitive composite score, or growth metrics at 18–22 months corrected age were observed (110).

Studies in term infants in Australia have reported associations of mildly elevated neonatal TSH levels with being exempt from school testing due to significant or complex disability (111) and poor educational and developmental outcomes (112). In contrast, a Belgian study found that although school-aged children with neonatal TSH concentrations between 10 and 15 mIU/L initially displayed lower verbal IQ scores, this association was no longer significant after adjusting for household income, maternal education and bilingualism (101). The hypothesis that such patterns might reflect differences in population iodine status (95) is consistent with the findings of a recent meta-analysis of 13 studies that found high newborn TSH levels negatively impacted cognitive development in iodine-deficient areas, with no significant relationship between these variables in iodine-sufficient populations (113).

NBS for primary CH and omic sciences

The integration of biochemical data from NBS with omics sciences – particularly genomics, proteomics and metabolomics – offers significant potential to enhance our understanding of primary CH and to uncover new biomarkers able to identify mild cases with altered proteomic/metabolic profiles associated with the need of treatment. Over the past few decades, genomics has helped clarify the genetic underpinnings of thyroid dysgenesis and thyroid function defects (114, 115, 116, 117). However, genetic variations account for a small minority of CH cases. As mentioned above, a key challenge for the future of NBS for CH lies in improving detection of true cases of primary CH, reducing both over- and underdiagnosis of mild cases of CH. Mild cases with true primary CH may exhibit distinct circulatory and excretory metabolic profiles.

Wright *et al.* conducted a study on the thyroglobulin (Tg) interactome using multiplexed quantitative affinity purification-mass spectrometry (118). They defined the Tg proteostasis interactome and observed changes in proteostasis between wild-type Tg and several CH variants. Their findings demonstrated that mutant Tg processing is associated with common proteostasis imbalances, including increased chaperoning, oxidative folding and enhanced engagement of factors targeting proteins for endoplasmic reticulum-associated degradation (118).

Metabolomics may also aid in identifying infants with mild TSH elevations who could benefit from early treatment by predicting hypothyroidism through metabolic changes. Shao *et al.* showed that both subclinical and clinical hypothyroidism in adults are associated with metabolic alterations, including changes in bile acid biosynthesis, steroid hormone biosynthesis and amino acid metabolism. These findings could potentially be extended to infants with mild TSH elevation (119).

Moreover, discrepancies in thyroid hormone levels and actions across different tissues in specific clinical contexts highlight the need for detailed metabolic profiling and tissue-specific biomarkers to more accurately assess thyroid function. Future approaches may include

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tissue-specific delivery systems and isoform-specific targeting of thyromimetics or thyroid hormone metabolites, providing a minimally invasive treatment option (120).

NBS for primary CH and linkage-record studies

In the past decade, the linkage of NBS data to routinely collected health, development and education data have been used to examine long-term outcomes for children with various initial TSH levels (112). There are many advantages to using linked administrative data to identify long-term outcomes compared to other traditional outcome collection methods such as patient interviews or medical record reviews. As administrative data are collected for other purposes, in jurisdictions where these data sources are readily available, data linkage may be a cost- and time-efficient method of outcome collection. Data linkage minimizes loss to follow-up as patients, particularly for long-term outcomes where individuals have moved and cannot be contacted (121) and reduces the burden of being involved in research studies on children and families.

However, routinely collected administrative data lack clinical detail of diagnosis, treatment and comorbidities. This limitation can be overcome by the linkage between bespoke clinical datasets and administrative data for outcome identification. A project in the UK is linking routinely collected health, vital statistics and education data to NBS data. These data are also being linked to a clinical dataset of 1800 children who have screened positive for CH, which includes longitudinal data on blood tests, thyroid scans and treatment doses (122). However, these clinical datasets require substantial time and input from clinicians and parents for initial data collection.

Electronic health records (EHRs) are a potentially rich source of clinical data. An EHR is a digital version of a patient's medical records updated in real time by clinicians. The EHR can contain structured clinical information recorded using clinical coding systems; however, it can also include large amounts of free text, imaging and laboratory data. While the amount of data being stored in EHRs is increasing, there are a number of barriers to the use of EHR in research, including lack of completeness of the records (123), high amounts of unstructured text (124), and lack of standardization between systems (125). However, advances in analytic techniques, particularly in machine learning and natural language processing (124), can help in realizing the full potential of EHRs for research purposes. Using EHRs and other forms of 'big data' for research is an exciting prospect for researchers and clinicians; however, it is important to balance releasing confidential medical information for research and ensuring individuals' privacy and confidentiality are protected (124).

NBS for primary CH in all the countries worldwide

NBS for CH is an established public health measure that significantly reduces the risk of cognitive disabilities and other severe consequences if treatment is initiated promptly. Despite its critical importance, only about 30% of newborns globally have access to this potentially life-changing screening (26). Universal NBS facilitates the early detection and timely administration of levothyroxine, reducing the burden of intellectual disability, which justifies NBS (126).

Low- and middle-income countries (LMICs) face numerous challenges in implementing NBS programs, which can arise at various stages, including pre-analytical, analytical and post-analytical phases (127, 128). In settings where most deliveries occur in hospitals and mothers are discharged within 24–48 h, using cord blood for CH screening is a feasible option. However, in regions where most births happen at home, alternative and innovative strategies for NBS need to be developed. For instance, a pilot project in Nigeria successfully offered screening for sickle cell disease to infants up to 9 months old attending immunization clinics, achieving coverage of up to 90% (129). Since most infants in sub-Saharan Africa receive Bacillus Calmette-Guerine (BCG) and polio vaccination, 71.7 and 76.1%, respectively, by day 28 of life (130), a similar proportion of infants could be screened within the first month of life if NBS and immunizations were linked. In these cases, point-of-care testing (POCT) devices could be used to perform the screening, identifying children who may need referral for confirmatory testing if the initial screening result is positive. POCT involves laboratory tests performed near the patient care site, offering rapid results to initiate appropriate treatment once the condition has been confirmed. This might be particularly beneficial in resource-limited settings where access to medical care is limited. These straightforward tests are suitable for use in primary care and remote areas without traditional laboratory facilities. Conventional laboratory tests typically require a multistep process, including sample collection, transportation to a distant centralized laboratory, and various processing steps. The absence of healthcare providers, transportation or adequate resources can delay clinical decision-making. With these considerations in mind, we proposed a strategy to enhance screening coverage for CH in LMICs by using POCT for TSH at tuberculosis vaccination clinics (131). Recent research has demonstrated high sensitivity and specificity of TSH detection using POCT compared to a reference method, with consistent results across venous, capillary and serum samples. The study also showed high accuracy for diagnosing thyroid disorders and a strong correlation between POCT and the reference method (132). Furthermore, an Italian study confirmed the robust performance of a different bedside test relative

to the standard reference method, underscoring POCT's potential for NBS (133). However, costing analyses of POCT implementation in LMICs, similar to those already conducted for sickle cell disease screening (138), are needed to assess feasibility and inform policy decisions for the adoption of this strategy in NBS.

Conclusion

NBS for primary CH is a successful public health measure that, in combination with early treatment, has prevented cognitive impairment in large numbers of infants worldwide. An important lesson learned from decades of experience with programs screening for this disease is that primary CH is a complex condition with phenotypes including mild, moderate and severe forms. Over the decades, NBS for primary CH has evolved to increase the sensitivity of screening tests, which has led to changes in the population of newborns diagnosed with CH. A key challenge for the future lies in improving detection of true cases of primary CH while reducing the overdiagnosis of infants for whom treatment is not required. Although the debate about optimal TSH cutoffs continues, omic sciences may help researchers to enhance understanding of primary CH and to uncover new biomarkers to identify mild cases with altered proteomic and/or metabolic profiles associated with the need for treatment. Record-linkage studies can help to deepen knowledge on long-term outcomes of children with primary CH identified through NBS. Nevertheless, despite 50 years of NBS for primary CH, only 30% of the world's newborns currently benefit from this essential public health intervention. It is therefore imperative to make concerted efforts to expand access to NBS globally and make possible a better life for all affected newborns, especially those born in LMICs.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the work reported.

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References

1 Cheetham T & Wood C. Paediatric thyroid disease. *Clin Endocrinol* 2024 **101** 223–233. (https://doi.org/10.1111/cen.15110)

2 Grüters A & Krude H. Detection and treatment of congenital hypothyroidism. *Nat Rev Endocrinol* 2012 **8** 104–113. (https://doi.org/10.1038/nrendo.2011.160)

3 Rastogi MV & LaFranchi SH. Congenital hypothyroidism. *Orphanet J Rare Dis* 2010 **5** 17. (https://doi.org/10.1186/1750-1172-5-17)

4 Cherella CE & Wassner AJ. Update on congenital hypothyroidism. *Curr Opin Endocrinol Diabetes Obes* 2020 **27** 63–69. (https://doi.org/10.1097/med.0000000000000520)

5 Dussault JH & Laberge C. Thyroxine (T4) determination by radioimmunological methods in dried blood dried blood eluate: new diagnostic method of neonatal hypothyroidism? *Union Med Can* 1973 **102** 2062–2064.

6 Dussault JH, Coulombe P, Laberge C, *et al.* Preliminary report on a mass screening program for neonatal hypothyroidism. *J Pediatr* 1975 **86** 670–674. (https://doi.org/10.1016/s0022-3476(75)80349-0)

7 Fisher DA, Dussault JH, Foley TP Jr, *et al.* Screening for congenital hypothyroidism: results of screening one million North American infants. *J Pediatr* 1979 **94** 700–705. (https://doi.org/10.1016/s0022-3476(79)80133-x)

8 Wilson J & Jungner G & World Health Organization. *Principles and Practice of Screening for Disease*. Geneva: World Health Organisation, 1968. (https://iris.who.int/handle/10665/37650)

9 Klein AH, Meltzer S & Kenny FM. Improved prognosis in congenital bypothyroidism treated before age three months. *J Pediatr* 1972 **81** 912–915. (https://doi.org/10.1016/s0022-3476(72)80542-0)

10 Smith DW, Blizzard RM & Wilkins L. The mental prognosis in hypothyroidism of infancy and childhood: a review of 128 cases. *Pediatrics* 1957 **19** 1011–1022. (https://doi.org/10.1542/peds.19.6.1011)

11 LaFranchi S. Newborn screening for congenital hypothyroidism. *Pediatr Ann* 1980 **9** 54–65. (https://doi.org/10.3928/0090-4481-19801001-08)

12 Layde PM, Von Allmen SD & Oakley GP. Congenital hypothyroidism control programs: a cost-benefit analysis. *JAMA* 1979 **241** 2290–2292. (https://doi.org/10.1001/jama.1979.03290470040024)

13 Van Trotsenburg P, Stoupa A, Léger J, *et al.* Congenital hypothyroidism: a 2020–2021 consensus guidelines update—an endo-European reference network initiative endorsed by the European society for pediatric Endocrinology and the European society for Endocrinology. *Thyroid* 2021 **31** 387–419. (https://doi.org/10.1089/thy.2020.0333)

14 Leger J, Olivieri A, Donaldson M, *et al.* European society for paediatric Endocrinology consensus guidelines on screening, diagnosis, and management of congenital hypothyroidism. *J Clin Endocrinol Metab* 2014 **99** 363–384. (https://doi.org/10.1210/jc.2013-1891)

15 Zwaveling-Soonawala N, van Trotsenburg ASP & Verkerk PH. The severity of congenital hypothyroidism of central origin should not be underestimated. *J Clin Endocrinol Metab* 2015 **100** E297–E300. (https://doi.org/10.1210/jc.2014-2871)

16 Adachi M, Soneda A, Asakura Y, *et al.* Mass screening of newborns for congenital hypothyroidism of central origin by free thyroxine measurement of blood samples on filter paper. *Eur J Endocrinol* 2012 **166** 829–838. (https://doi.org/10.1530/eje-11-0653)

17 Kempers M, Lanting C, Van Heijst A, *et al.* Neonatal screening for congenital hypothyroidism based on thyroxine, thyrotropin, and thyroxine-binding globulin measurement: potentials and pitfalls. *J Clin Endocrinol Metab* 2006 **91** 3370–3376. (https://doi.org/10.1210/jc.2006-0058)

18 Persani L, Brabant G, Dattani M, *et al.* 2018 European Thyroid Association (ETA) guidelines on the diagnosis and management of

central hypothyroidism. *Eur Thyroid J* 2018 **7** 225–237. (https://doi.org/10.1159/000491388)

19 Rose SR, Wassner AJ, Wintergerst KA, *et al.* Congenital hypothyroidism: screening and management. *Pediatrics* 2023 **151** e2022060420. (https://doi.org/10.1542/peds.2022-060420)

20 Barnes N. Serum TSH measurement in children with thyroid disorders. *Arch Dis Child* 1975 **50** 497–499. (https://doi.org/10.1136/adc.50.7.497)

21 Gordin A, Saarinen P, Pelkonen R, *et al.* Serum thyrotrophin and the response to thyrotrophin releasing hormone in symptomless autoimmune thyroiditis and in borderline and overt hypothyroidism. *Eur J Endocrinol* 1974 **75** 274–285. (https://doi.org/10.1530/acta.0.0750274)

22 Hayek A, Maloof F & Crawford JD. Thyrotropin behavior in thyroid disorders of childhood. *Pediatr Res* 1973 **7** 28–38. (https://doi.org/10.1203/00006450-197301000-00005)

23 Dussault JH, Coulombe P & Laberge C. Neonatal thyroid screening. In *Perinatal Thyroid Physiology and Disease*, p 221. Eds DA Fisher & B GN. New York: Raven Press, 1975.

24 Foley T, Klein AH, Agustin AV, *et al.* Screening for congenital hypothyroidism by determination of thyrotropin levels. In *Perinatal Thyroid Physiology and Disease*, p p255. Eds D Fisher & G Burrow. New York: Raven Press, 1975.

25 Delange F, Camus M, Winkler M, *et al.* Serum thyrotrophin determination on day 5 of life as screening procedure for congenital hypothyroidism. *Arch Dis Child* 1977 **52** 89–96. (https://doi.org/10.1136/adc.52.2.89)

26 Ford G & LaFranchi SH. Screening for congenital hypothyroidism: a worldwide view of strategies. *Best Pract Res Clin Endocrinol Metab* 2014 **28** 175–187. (https://doi.org/10.1016/j.beem.2013.05.008)

27 Kilberg MJ, Rasooly IR, LaFranchi SH, *et al.* Newborn screening in the US may miss mild persistent hypothyroidism. *J Pediatr* 2018 **192** 204–208. (https://doi.org/10.1016/j.jpeds.2017.09.003)

28 Selva KA, Harper A, Downs A, *et al.* Neurodevelopmental outcomes in congenital hypothyroidism: comparison of initial T4 dose and time to reach target T4 and TSH. *J Pediatr* 2005 **147** 775–780. (https://doi.org/10.1016/j.jpeds.2005.07.024)

29 Langham S, Hindmarsh P, Krywawych S, *et al.* Screening for congenital hypothyroidism: comparison of borderline screening cut-off points and the effect on the number of children treated with levothyroxine. *Eur Thyroid J* 2013 **2** 180–186. (https://doi.org/10.1159/000350039)

30 Mengreli C, Kanaka-Gantenbein C, Girginoudis P, *et al.* Screening for congenital hypothyroidism: the significance of threshold limit in false-negative results. *J Clin Endocrinol Metab* 2010 **95** 4283–4290. (https://doi.org/10.1210/jc.2010-0057)

31 Olivieri A, Corbetta C, Weber G, *et al.* Congenital hypothyroidism due to defects of thyroid development and mild increase of TSH at screening: data from the Italian National Registry of infants with congenital hypothyroidism. *J Clin Endocrinol Metab* 2013 **98** 1403–1408. (https://doi.org/10.1210/jc.2012-3273)

32 Mandel SJ, Hermos RJ, Larson CA, *et al.* Atypical hypothyroidism and the very low birthweight infant. *Thyroid* 2000 **10** 693–695. (https://doi.org/10.1089/10507250050137770)

33 Hyman SJ, Greig F, Holzman I, *et al.* Late rise of thyroid stimulating hormone in ill newborns. *J Pediatr Endocrinol Metab* 2007 **20** 501–510. (https://doi.org/10.1515/jpem.2007.20.4.501)

34 Larson C, Hermos R, Delaney A, *et al.* Risk factors associated with delayed thyrotropin elevations in congenital hypothyroidism. *J Pediatr* 2003 **143** 587–591. (https://doi.org/10.1067/s0022-3476(03)00332-9)

35 Monden C, Pison G & Smits J. Twin Peaks: more twinning in humans than ever before. *Hum Reprod* 2021 **36** 1666–1673. (https://doi.org/10.1093/humrep/deab029)

36 Seetho S, Kongwattanakul K, Saksiriwuttho P, *et al.* Epidemiology and factors associated with preterm births in multiple pregnancy: a retrospective cohort study. *BMC Pregnancy Childbirth* 2023 **23** 872. (https://doi.org/10.1186/s12884-023-06186-0)

37 Chawanpaiboon S, Vogel JP, Moller A-B, *et al.* Global, regional, and national estimates of levels of preterm birth in 2014: a systematic review and modelling analysis. *Lancet Global Health* 2019 **7** e37–e46. (https://doi.org/10.1016/s2214-109x(18)30451-0)

38 Martin JA & Osterman MJ. Shifts in the distribution of births by gestational age: United States, 2014–2022. *Natl Vital Stat Rep* 2024 **73** 1–11. (https://www.cdc.gov/nchs//data/nvsr/nvsr73/nvsr73-01.pdf)

39 Fuchs F, Monet B, Ducruet T, *et al.* Effect of maternal age on the risk of preterm birth: a large cohort study. *PLoS One* 2018 **13** e0191002. (https://doi.org/10.1371/journal.pone.0191002)

40 Torres C, Caporali A & Pison G. The Human multiple births database (HMDB). *Demographic Research* 2023 **48** 89–106. (https://doi.org/10.4054/DemRes.2023.48.4)

41 Collins J. Global epidemiology of multiple birth. *Reprod Biomed Online* 2007 **15** 45–52. (https://doi.org/10.1016/s1472-6483(10)62251-1)

42 Hall JG. Twinning. *Lancet* 2003 **362** 735–743. (https://doi.org/10.1016/s0140-6736(03)14237-7)

43 Martin JA, Hamilton BE & Osterman MJ. Three decades of twin births in the United States, 1980–2009. *NCHS Data Brief* 2012 1–8. (https://www.cdc.gov/nchs/data/databriefs/db80.pdf)

44 Luke B. Pregnancy and birth outcomes in couples with infertility with and without assisted reproductive technology: with an emphasis on US population-based studies. *Am J Obstet Gynecol* 2017 **217** 270–281. (https://doi.org/10.1016/j.ajog.2017.03.012)

45 Pison G, Monden C & Smits J. Twinning rates in developed countries: trends and explanations. *Popul Dev Rev* 2015 **41** 629–649. (https://doi.org/10.1111/j.1728-4457.2015.00088.x)

46 Bourque SL, Murthy K, Grover TR, *et al.* Cutting into the NICU: improvements in outcomes for neonates with surgical conditions. *Neoreviews* 2024 **25** e634–e647. (https://doi.org/10.1542/neo.25-10-e634)

47 Pearlman SA. Advancements in neonatology through quality improvement. *J Perinatol* 2022 **42** 1277–1282. (https://doi.org/10.1038/s41372-022-01383-9)

48 Gamber RA, Blonsky H, McDowell M, *et al.* Declining birth rates, increasing maternal age and neonatal intensive care unit admissions. *J Perinatol* 2024 **44** 203–208. (https://doi.org/10.1038/s41372-023-01834-x)

49 Olivo S, Venier D, Zannier M, *et al.* A two-year retrospective study of the neonatal emergency transport service in Northeast Italy. *J Matern Fetal Neonatal Med* 2023 **36** 2199907. (https://doi.org/10.1080/14767058.2023.2199907)

50 Myrhaug HT, Brurberg KG, Hov L, *et al.* Survival and impairment of extremely premature infants: a meta-analysis. *Pediatrics* 2019 **143** e20180933. (https://doi.org/10.1542/peds.2018-0933)

51 LaFranchi SH. Thyroid function in preterm/low birth weight infants: impact on diagnosis and management of thyroid dysfunction. *Front Endocrinol* 2021 **12** 666207. (https://doi.org/10.3389/fendo.2021.666207)

52 Fisher DA. Thyroid system immaturities in very low birth weight premature infants. *Semin perinatology* 2008 **32** 387–397. (https://doi.org/10.1053/j.semperi.2008.09.003)

53 De Zegher F, Van den Berghe G, Dumoulin M, *et al.* Dopamine suppresses thyroid-stimulating hormone secretion in neonatal hypothyroidism. *Acta Pædiatrica* 1995 **84** 213–214. (https://doi.org/10.1111/j.1651-2227.1995.tb13614.x)

54 Re R, Kourides I, Ridgway E, *et al.* The effect of glucocorticoid administration on human pituitary secretion of thyrotropin and prolactin. *J Clin Endocrinol Metab* 1976 **43** 338–346. (https://doi.org/10.1210/jcem-43-2-338)

55 Brown RS, Bloomfield S, Bednarek FJ, *et al.* Routine skin cleansing with povidone-iodine is not a common cause of transient neonatal hypothyroidism in North America: a prospective controlled study. *Thyroid* 1997 **7** 395–400. (https://doi.org/10.1089/thy.1997.7.395)

56 Gordon CM, Rowitch DH, Mitchell ML, *et al.* Topical iodine and neonatal hypothyroidism. *Arch Pediatr Adolesc Med* 1995 **149** 1336–1339. (https://doi.org/10.1001/archpedi.1995.02170250042006)

57 Goldsmit GS, Valdes M, Herzovich V, *et al.* Evaluation and clinical application of changes in thyroid hormone and TSH levels in critically ill full-term newborns. *J Perinat Med* 2011 **39** 59–64. (https://doi.org/10.1515/jpm.2010.120)

58 Golombek SG. Nonthyroidal illness syndrome and euthyroid sick syndrome in intensive care patients. *Semin Perinatology* 2008 **32** 413–418. (https://doi.org/10.1053/j.semperi.2008.09.010)

59 Williams FL, Ogston SA, van Toor H, *et al.* Serum thyroid hormones in preterm infants: associations with postnatal illnesses and drug usage. *J Clin Endocrinol Metab* 2005 **90** 5954–5963. (https://doi.org/10.1210/jc.2005-1049)

60 Perry R, Heinrichs C, Bourdoux P, *et al.* Discordance of monozygotic twins for thyroid dysgenesis: implications for screening and for molecular pathophysiology. *J Clin Endocrinol Metab* 2002 **87** 4072–4077. (https://doi.org/10.1210/jc.2001-011995)

61 Grob F. Approaching the diagnosis of thyroid disorders in preterm infants. *Pediatr Res* 2022 **91** 1021–1022. (https://doi.org/10.1038/s41390-022-01951-x)

62 Grob F, Gutiérrez M, Leguizamón L, *et al.* Hyperthyrotropinemia is common in preterm infants who are born small for gestational age. *J Pediatr Endocrinol Metab* 2020 **33** 375–382. (https://doi.org/10.1515/jpem-2019-0461)

63 Nolan B, Uy C, Stablein L, *et al.* Screening for delayed thyroid stimulation hormone rise and atypical congenital hypothyroidism in infants born very preterm and infants with very low birth weight. *J Pediatr* 2024 **269** 113974. (https://doi.org/10.1016/j.jpeds.2024.113974)

64 Rapaport R, Rose SR & Freemark M. Hypothyroxinemia in the preterm infant: the benefits and risks of thyroxine treatment. *J Pediatr* 2001 **139** 182–188. (https://doi.org/10.1067/mpd.2001.116934)

65 Simpson J, Williams FL, Delahunty C, *et al.* Serum thyroid hormones in preterm infants and relationships to indices of severity of intercurrent illness. *J Clin Endocrinol Metab* 2005 **90** 1271–1279. (https://doi.org/10.1210/jc.2004-2091)

66 Van Wassenaer AG, Kok JH, Dekker FW, *et al.* Thyroid function in very preterm infants: influences of gestational age and disease. *Pediatr Res* 1997 **42** 604–609. (https://doi.org/10.1203/00006450-199711000-00009)

67 Cavarzere P, Camilot M, Popa FI, *et al.* Congenital hypothyroidism with delayed TSH elevation in low-birth-weight infants: incidence, diagnosis and management. *Eur J Endocrinol* 2016 **175** 395–402. (https://doi.org/10.1530/eje-15-1233)

68 LaFranchi SH. Screening preterm infants for congenital hypothyroidism: better the second time around. *J Pediatr* 2014 **164** 1259–1261. (https://doi.org/10.1016/j.jpeds.2014.02.031)

69 McGrath N, Hawkes CP, Mayne P, *et al.* Optimal timing of repeat newborn screening for congenital hypothyroidism in preterm infants to detect delayed thyroid-stimulating hormone elevation. *J Pediatr* 2019 **205** 77–82. (https://doi.org/10.1016/j.jpeds.2018.09.044)

70 Zung A, Yehieli A, Blau A, *et al.* Characteristics of delayed thyroid stimulating hormone elevation in neonatal intensive care unit newborns. *J Pediatr* 2016 **178** 135–140. e1. (https://doi.org/10.1016/j.jpeds.2016.07.022)

71 Kaluarachchi DC, Allen DB, Eickhoff JC, *et al.* Increased congenital hypothyroidism detection in preterm infants with serial newborn screening. *J Pediatr* 2019 **207** 220–225. (https://doi.org/10.1016/j.jpeds.2018.11.044)

72 Spencer CA. Laboratory thyroid tests: a historical perspective. *Thyroid* 2023 **33** 407–419. (https://doi.org/10.1089/thy.2022.0397)

73 Clinical and Laboratory Standards Institute. *Newborn Screening for Congenital Hypothyroidism*, 1st edn, 2024. CLSI guidelines NBS10.

74 Association of Public Health Laboratories. Overview of cutoff determinations and risk assessment methods used in dried blood spot newborn screening- role of cutoffs and other methods of data analysis. 2022. (https://www.aphl.org/programs/newborn_screening/Documents/Cutoff-Determinations-and-Risk-Assessment-Methods.pdf)

75 Peters C, Brooke I, Heales S, *et al.* Defining the newborn blood spot screening reference interval for TSH: impact of ethnicity. *J Clin Endocrinol Metab* 2016 **101** 3445–3449. (https://doi.org/10.1210/jc.2016-1822)

76 Krude H & Blankenstein O. Treating patients not numbers: the benefit and burden of lowering TSH newborn screening cut-offs. *Arch Dis Child* 2011 **96** 121–122. (https://doi.org/10.1136/adc.2009.174466)

77 Hewlett J & Waisbren S. A review of the psychosocial effects of false-positive results on parents and current communication practices in newborn screening. *J Inherit Metab Dis* 2006 **29** 677–682. (https://doi.org/10.1007/s10545-006-0381-1)

78 Lem AJ, de Rijke YB, van Toor H, *et al.* Serum thyroid hormone levels in healthy children from birth to adulthood and in short children born small for gestational age. *J Clin Endocrinol Metab* 2012 **97** 3170–3178. (https://doi.org/10.1210/jc.2012-1759)

79 Allen DB, Sieger JE, Litsheim T, *et al.* Age-adjusted thyrotropin criteria for neonatal screening for hypothyroidism. *J Pediatr* 1990 **117** 309–312. (https://doi.org/10.1016/s0022-3476(05)80553-0)

80 Lott JA, Sardovia-Iyer M, Speakman KS, *et al.* Age-dependent cutoff values in screening newborns for hypothyroidism. *Clin Biochem* 2004 **37** 791–797. (https://doi.org/10.1016/j.clinbiochem.2004.05.019)

81 Ruoppolo M, Malvagia S, Boenzi S, *et al.* Expanded newborn screening in Italy using tandem mass spectrometry: two years of national experience. *Int J Neonatal Screen* 2022 **8** 47. (https://doi.org/10.3390/ijns8030047)

82 Chiesa A, Prieto L, Mendez V, *et al.* Prevalence and etiology of congenital hypothyroidism detected through an argentine neonatal screening program (1997–2010). *Horm Res Paediatr* 2013 **80** 185–192. (https://doi.org/10.1159/000354409)

83 Deladoey J, Ruel J, Giguere Y, *et al.* Is the incidence of congenital hypothyroidism really increasing? A 20-year retrospective

population-based study in Quebec. *J Clin Endocrinol Metab* 2011 **96** 2422–2429. (https://doi.org/10.1210/jc.2011-1073)

84 Olivieri A, Fazzini C & Medda E. Multiple factors influencing the incidence of congenital hypothyroidism detected by neonatal screening. *Horm Res Paediatr* 2015 **83** 86–93. (https://doi.org/10.1159/000369394)

85 Teixeira Palla Braga N, Vilela Antunes JM, Colosimo EA, *et al.* Impact of lowering TSH cut-off on neonatal screening for congenital hypothyroidism in Minas Gerais, Brazil. *Int J Neonatal Screen* 2024 **10** 52. (https://doi.org/10.3390/ijns10030052)

86 Caiulo S, Corbetta C, Di Frenna M, *et al.* Newborn screening for congenital hypothyroidism: the benefit of using differential TSH cutoffs in a 2-screen program. *J Clin Endocrinol Metab* 2021 **106** e338–e349. (https://doi.org/10.1210/clinem/dgaa789)

87 Jones DE, Hart K, Shapira SK, *et al.* Identification of primary congenital hypothyroidism based on two newborn screens – Utah, 2010–2016. *MMWR Morb Mortal Wkly Rep* 2018 **67** 782–785. (https://doi.org/10.15585/mmwr.mm6728a4)

88 Shapira SK, Hinton CF, Held PK, *et al.* Single newborn screen or routine second screening for primary congenital hypothyroidism. *Mol Genet Metabol* 2015 **116** 125–132. (https://doi.org/10.1016/j.ymgme.2015.08.003)

89 Baz-Redón N, Antolín M, Clemente M, *et al.* Patients with thyroid dyshormonogenesis and DUOX2 variants: molecular and clinical description and genotype–phenotype correlation. *Int J Mol Sci* 2024 **25** 8473. (https://doi.org/10.3390/ijms25158473)

90 Parks JS, Lin M, Grosse SD, *et al.* The impact of transient hypothyroidism on the increasing rate of congenital hypothyroidism in the United States. *Pediatrics* 2010 **125** (Supplement 2) S54–S63. (https://doi.org/10.1542/peds.2009-1975f)

91 Jones JH, Smith S, Dorrian C, *et al.* Permanent congenital hypothyroidism with blood spot thyroid stimulating hormone< 10 mU/L. *Arch Dis Child* 2018 **103** 65–67. (https://doi.org/10.1136/archdischild-2015-309564)

92 Corbetta C, Weber G, Cortinovis F, *et al.* A 7-year experience with low blood TSH cutoff levels for neonatal screening reveals an unsuspected frequency of congenital hypothyroidism (CH). *Clin Endocrinol* 2009 **71** 739–745. (https://doi.org/10.1111/j.1365-2265.2009.03568.x)

93 McGrath N, Hawkes CP, Mayne P, *et al.* Permanent decompensated congenital hypothyroidism in newborns with whole-blood thyroid-stimulating hormone concentrations between 8 and 10 mU/L: the case for lowering the threshold. *Horm Res Paediatr* 2018 **89** 265–270. (https://doi.org/10.1159/000488288)

94 Mitrovic K, Vukovic R, Milenkovic T, *et al.* Changes in the incidence and etiology of congenital hypothyroidism detected during 30 years of a screening program in central Serbia. *Eur J Pediatr* 2016 **175** 253–259. (https://doi.org/10.1007/s00431-015-2630-5)

95 Lain S, Trumpff C, Grosse SD, *et al.* Are lower TSH cutoffs in neonatal screening for congenital hypothyroidism warranted? *Eur J Endocrinol* 2017 **177** D1–D12. (https://doi.org/10.1530/eje-17-0107)

96 Jones JH, Mackenzie J, Croft G, *et al.* Improvement in screening performance and diagnosis of congenital hypothyroidism in Scotland 1979–2003. *Arch Dis Child* 2006 **91** 680–685. (https://doi.org/10.1136/adc.2005.088427)

97 Liu L, He W, Zhu J, *et al.* Global prevalence of congenital hypothyroidism among neonates from 1969 to 2020: a systematic review and meta-analysis. *Eur J Pediatr* 2023 **182** 2957–2965. (https://doi.org/10.1007/s00431-023-04932-2)

98 American Academy of Pediatrics. Update of newborn screening and therapy for congenital hypothyroidism. *Pediatrics* 2006 **117** 2290–2303. (https://doi.org/10.1542/peds.2006-0915)

99 Yao Y, Deng K, Zhu J, *et al.* Increased incidence of congenital hypothyroidism in China: an analysis of 119 million screened newborns. *Eur J Pediatr* 2023 **182** 4477–4486. (https://doi.org/10.1007/s00431-023-05108-8)

100 Herbstman J, Apelberg BJ, Witter FR, *et al.* Maternal, infant, and delivery factors associated with neonatal thyroid hormone status. *Thyroid* 2008 **18** 67–76. (https://doi.org/10.1089/thy.2007.0180)

101 Trumpff C, De Schepper J, Vanderfaeillie J, *et al.* Thyroid-Stimulating hormone (TSH) concentration at birth in Belgian neonates and cognitive development at preschool age. *Nutrients* 2015 **7** 9018–9032. (https://doi.org/10.3390/nu7115450)

102 Hinton CF, Harris KB, Borgfeld L, *et al.* Trends in incidence rates of congenital hypothyroidism related to select demographic factors: data from the United States, California, Massachusetts, New York, and Texas. *Pediatrics* 2010 **125** (Supplement 2) S37–S47. (https://doi.org/10.1542/peds.2009-1975d)

103 Lain S, Nassar N & Jack M. Clinical survey of current practice regarding treatment of children with borderline thyroid abnormalities. *J Paediatr Child Health* 2023 **59** 129–133. (https://doi.org/10.1111/jpc.16259)

104 Ziegler GM, Slaughter JL, Chaudhari M, *et al.* Preterm to term infant postmenstrual age reference intervals for thyroid-stimulating hormone and free thyroxine. *Pediatr Res* 2022 **91** 1130–1135. (https://doi.org/10.1038/s41390-021-01838-3)

105 Chiesa AE & Tellechea ML. Update on neonatal isolated hyperthyrotropinemia: a systematic review. *Front Endocrinol* 2021 **12** 643307. (https://doi.org/10.3389/fendo.2021.643307)

106 Odenwald B, Fischer A, Röschinger W, *et al.* Long-term course of hypothyroidism detected through neonatal TSH screening in a population-based cohort of very preterm infants born at less than 32 weeks of gestation. *Int J Neonatal Screen* 2021 **7** 65. (https://doi.org/10.3390/ijns7040065)

107 Woo HC, Lizarda A, Tucker R, *et al.* Congenital hypothyroidism with a delayed thyroid-stimulating hormone elevation in very premature infants: incidence and growth and developmental outcomes. *J Pediatr* 2011 **158** 538–542. (https://doi.org/10.1016/j.jpeds.2010.10.018)

108 Chen L-W, Chu C-H, Lin Y-C, *et al.* The quartile levels of thyroid-stimulating hormone at newborn screening stratified risks of neurodevelopmental impairment in extremely preterm infants: a population cohort study. *J Epidemiol* 2024 **34** 419–427. (https://doi.org/10.2188/jea.JE20230253)

109 Williams FL, Lindgren A, Watson J, *et al.* Thyroid function in preterm infants and neurodevelopment at 2 years. *Arch Dis Child Fetal Neonatal Ed* 2020 **105** 504–509. (https://doi.org/10.1136/archdischild-2018-316742)

110 Nicksic V, Allen D, Stanley M, *et al.* Lack of association between gestational age adjusted TSH percentiles and neurodevelopmental outcomes among preterm infants. *J Neonatal Perinat Med* 2022 **15** 243–247. (https://doi.org/10.3233/npm-210910)

111 Lain SJ, Wiley V, Jack M, *et al.* Association of elevated neonatal thyroid-stimulating hormone levels with school performance and stimulant prescription for attention deficit hyperactivity disorder in childhood. *Eur J Pediatr* 2021 **180** 1073–1080. (https://doi.org/10.1007/s00431-020-03828-9)

112 Lain SJ, Bentley JP, Wiley V, *et al.* Association between borderline neonatal thyroid-stimulating hormone concentrations and

educational and developmental outcomes: a population-based record-linkage study. *Lancet Diabetes Endocrinol* 2016 **4** 756–765. (https://doi.org/10.1016/s2213-8587(16)30122-x)

113 Wassie MM, Smithers LG & Zhou SJ. Association between newborn thyroid-stimulating-hormone concentration and neurodevelopment and growth: a systematic review. *Biol Trace Elem Res* 2021 **200** 473–487. (https://doi.org/10.1007/s12011-021-02665-7)

114 de Filippis T, Gelmini G, Paraboschi E, *et al.* A frequent oligogenic involvement in congenital hypothyroidism. *Hum Mol Genet* 2017 **26** 2507–2514. (https://doi.org/10.1093/hmg/ddx145)

115 Kara C, Mammadova J, Abur Ü, *et al.* Genetic testing can change diagnosis and treatment in children with congenital hypothyroidism. *Eur Thyroid J* 2023 **12** e220212. (https://doi.org/10.1530/etj-22-0212)

116 Stoupa A, Kariyawasam D, Polak M, *et al.* Genetics of congenital hypothyroidism: modern concepts. *Pediatr Invest* 2022 **6** 123–134. (https://doi.org/10.1002/ped4.12324)

117 Szinnai G. Genetics of normal and abnormal thyroid development in humans. *Best Pract Res Clin Endocrinol Metab* 2014 **28** 133–150. (https://doi.org/10.1016/j.beem.2013.08.005)

118 Wright MT, Kouba L & Plate L. Thyroglobulin interactome profiling defines altered proteostasis topology associated with thyroid dyshormonogenesis. *Mol Cell Proteomics* 2021 **20** 100008. (https://doi.org/10.1074/mcp.ra120.002168)

119 Shao F, Li R, Guo Q, *et al.* Plasma metabolomics reveals systemic metabolic alterations of subclinical and clinical hypothyroidism. *J Clin Endocrinol Metab* 2023 **108** 13–25. (https://doi.org/10.1210/clinem/dgac555)

120 Sinha RA & Yen PM. Metabolic messengers: thyroid hormones. *Nat Metab* 2024 **6** 639–650. (https://doi.org/10.1038/s42255-024-00986-0)

121 James S, Pryke A, Cusumano J, *et al.* 20-year outcomes of childhood-onset type 1 diabetes: the CANDID incident cohort survey. *Diabet Med* 2021 **38** e14473. (https://doi.org/10.1111/dme.14473)

122 Ruiz Nishiki M, Cabecinha M, Knowles R, *et al.* Establishing risk factors and outcomes for congenital hypothyroidism with gland in situ using population-based data linkage methods: study protocol. *BMJ Paediatrics Open* 2022 **6** e001341. (https://doi.org/10.1136/bmjpo-2021-001341)

123 Weiskopf NG, Hripcsak G, Swaminathan S, *et al.* Defining and measuring completeness of electronic health records for secondary use. *J Biomed Inf* 2013 **46** 830–836. (https://doi.org/10.1016/j.jbi.2013.06.010)

124 Ross M, Wei W & Ohno-Machado L. "Big data" and the electronic health record. *Yearb Med Inform* 2014 **23** 97–104. (https://doi.org/10.15265/iy-2014-0003)

125 Jensen PB, Jensen LJ & Brunak S. Mining electronic health records: towards better research applications and clinical care. *Nat Rev Genet* 2012 **13** 395–405. (https://doi.org/10.1038/nrg3208)

126 Grosse SD & Van Vliet G. Prevention of intellectual disability through screening for congenital hypothyroidism: how much and at what level? *Arch Dis Child* 2011 **96** 374–379. (https://doi.org/10.1136/adc.2010.190280)

127 Borrajo GJ. Newborn screening in Latin America: a brief overview of the state of the art. *Am J Med Genet C Semin Med Genet* 2021 **187** 322–328. (https://doi.org/10.1002/ajmg.c.31899)

128 Therrell BL & Padilla CD. Newborn screening in the developing countries. *Curr Opin Pediatr* 2018 **30** 734–739. (https://doi.org/10.1097/mop.0000000000000683)

129 Nnodu OE, Sopekan A, Nnebe-Agumadu U, *et al.* Implementing newborn screening for sickle cell disease as part of immunisation programmes in Nigeria: a feasibility study. *Lancet Haematol* 2020 **7** E534–E540. (https://doi.org/10.1016/S2352-3026(20)30143-5)

130 Bassoum O, Kimura M, Tal Dia A, *et al.* Coverage and timeliness of birth dose vaccination in sub-Saharan Africa: a systematic review and meta-analysis. *Vaccines* 2020 **8** 301. (https://doi.org/10.3390/vaccines8020301)

131 Grob F, Odame I & Van Vliet G. Worldwide newborn screening and early immunizations: aligning advances in preventive pediatrics. *J Pediatr* 2024 **264** 113732. (https://doi.org/10.1016/j.jpeds.2023.113732)

132 Shurbaji S, Al Tamimi F, Al Ghwairi MM, *et al.* High-sensitive detection and quantitation of thyroid-stimulating hormone (TSH) from capillary/fingerstick and venepuncture whole-blood using fluorescence-based rapid lateral flow immunoassay (LFIA). *Heliyon* 2023 **9** e20589. (https://doi.org/10.1016/j.heliyon.2023.e20589)

133 Di Cerbo A, Quagliano N, Napolitano A, *et al.* Comparison between an emerging point-of-care tool for TSH evaluation and a centralized laboratory-based method in a cohort of patients from Southern Italy. *Diagnostics* 2021 **11** 1590. (https://doi.org/10.3390/diagnostics11091590)

134 Silvestrin SM, Leone C & Leone CR. Detecting congenital hypothyroidism with newborn screening: the relevance of thyroid-stimulating hormone cutoff values. *Jornal de Pediatria* 2017 **93** 274–280. (https://doi.org/10.1016/j.jped.2016.07.006)

135 Botler J, Camacho LAB & Cruz MM. Phenylketonuria, congenital hypothyroidism and haemoglobinopathies: public health issues for a Brazilian newborn screening program. *Cad Saude Publica* 2012 **28** 1623–1631. (https://doi.org/10.1590/s0102-311x2012000900002)

136 Heather NL, Hofman PL, de Hora M, *et al.* Evaluation of the revised New Zealand national newborn screening protocol for congenital hypothyroidism. *Clin Endocrinol* 2017 **86** 431–437. (https://doi.org/10.1111/cen.13250)

137 Anastasovska V & Kocova M. Impact of lower screening TSH cutoff level on the increasing prevalence of congenital hypothyroidism. *Int J Neonatal Screen* 2017 **3** 7. (https://doi.org/10.3390/ijns3020007)

138 Mvundura M, Kiyaga C, Metzler M, *et al.* Cost for sickle cell disease screening using isoelectric focusing with dried blood spot samples and estimation of price thresholds for a point-of care test in Uganda. *Hematol Res Rev* 2019 **10** 59–67. (https://doi.org/10.2147/jbm.s186528)