PEDIATRIC ENDOCRINOLOGY AND DIABETES

2021 UPDATE

Editors:

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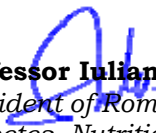
Preface

In the year we celebrate 100 years since the discovery of insulin, we unfortunately find ourselves in the second SARS-COV2 pandemic year and also in the middle of the 4th "wave" development, "wave" that at least in our country seems to be the most aggressive one so far. Under these circumstances the entire medical sector is subjected to unimaginable material and human efforts another time. When all the medical attention is focused on patients with Covid-19 who are desperately knocking on hospitals' doors, we must not lose sight of the small patients with chronic diseases in the records of endocrinology and pediatric diabetes services or endocrinology and adult diabetes services. Given the needs of these patients, we are often forced to provide remote consultations through telemedicine, which requires a continuous professional organization of the medical staff.

Respecting the principles stipulated in the Statute of the Romanian Society of Diabetes Nutrition and Pediatric Endocrinology in accordance with the recommendations to standardize the training of pediatricians in the field of endocrinology and pediatric diabetes even in these conditions of isolation and distancing in which the 8th National Congress of Diabetes Nutrition and Pediatric Endocrinology is taking place, we offer our "participants" a new "Pediatric Endocrinology and Diabetes – 2021 update" volume.



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President of Romanian Society of
Diabetes, Nutrition and Pediatric Endocrinology.

Contents

NEWBORN ENDOCRINOLOGY SCREENING
for
CONGENITAL HYPOTHYROIDISM
and
CONGENITAL ADRENAL HYPERPLASIA (CAH)
(ADRENOGENITAL SYNDROME [AGS]

Stuart J. Brink

Introduction

Both congenital hypothyroidism and congenital adrenal hyperplasia are two key congenital endocrine disturbances and able to be screened in order to prevent lifelong growth and mental retardation vis-à-vis congenital hypothyroidism and death in congenital adrenal hyperplasia from unrecognized severe adrenal insufficiency.

CONGENITAL HYPOTHYROIDISM[1]

Congenital hypothyroidism[2,3] can at times be overtly clinically obvious; but also, many times, can be very subtle so that clinical diagnosis is frequently missed unless there is a well-organized, neonatal screening program established and highly functional. Missed diagnosis of congenital hypothyroidism where such neonatal screening programs are not operational or unable to be afforded, results in longstanding growth abnormalities as well as lifelong mental retardation so that every analysis done regarding setting up neonatal screening facilities and cost benefit analysis as well

as quality of life issues have reiterated the importance of such screening shortly after birth.[4] Looking at pictures of newborns with congenital hypothyroidism can allow diagnosis under some (more severe physical abnormalities as shown in *figure 1*) but most instances of congenital hypothyroidism have minimal or very subtle abnormalities noted in the nursery or ambulatory facilities. Classical, yet often retrospective evaluation of neonatal findings[2] include "quiet" babies, hoarse crying, lethargy, constipation, feeding difficulties, dry skin, cold to touch, skin mottling, umbilical hernia, large/protruding tongues (macroglossia), large fontanels and wide cranial sutures, distended abdomen, slow reflexes. Despite this obvious list, all of which improves once thyroid hormone treatment is instituted, fewer than 5% of such neonates with documented congenital hypothyroidism were suspected of having this problem clinically.[2] *Figure 1* demonstrates one neonate with some these subtleties in hindsight knowing that there were abnormalities on screening compared to a neonate, presumably with more severe and/or earlier intrauterine hypothyroidism as the explanation proposed for "more severe" clinical findings.

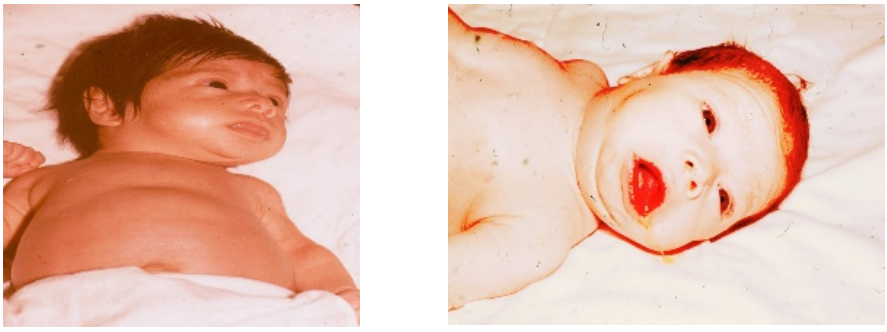

Figure 1: Classical (Severe) Newborn Congenital Hypothyroidism

Progression of these same subtle clinical findings in the untreated and undiagnosed neonate with congenital hypothyroidism generally progress during the early months of

postnatal life and also include excess weight gain as well as poor linear growth, further delayed bone age and worsening developmental landmarks.[1]

Early diagnosis and initiation of appropriate treatment allows thyroid hormone replacement with oral levothyroxine shortly after birth to begin correction of this endocrinopathy, avoids weight excess and linear height challenges as well as the obvious life-long severe mental retardation and brain problems associated with unrecognized/untreated hypothyroidism, cardiac problems, severe constipation and many other clinical issues. 1 year followup pictures of this same neonate and then late adolescent followup pictures of the same girl (*figure 2*) cared for by the author for all those years and diagnosed in a highly functioning screening program in which the author participates, demonstrate this well; absolutely no symptoms or signs of any problems all these years!

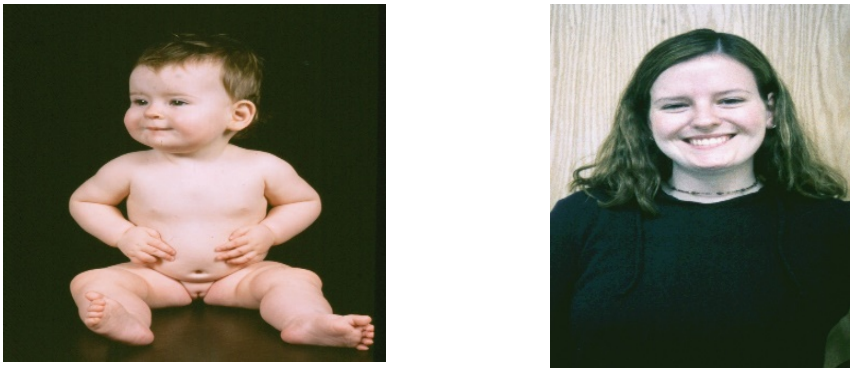

Figure 2: Normal one year old and adolescent well-treated congenital hypothyroidism

Congenital hypothyroidism can be grouped into five broad diagnostic categories as shown in *figure 3*. Dysgenesis is divided into athyreosis, hypoplasia and ectopia but specific incidence data varies from country to country with thyroid agenesis in the 20-50% range, 25-40% with ectopia, 4-15%

with dyshormonogenesis. Knowing about them provide the opportunity for discussions with parents and eventually as the child matures into adulthood, appropriate genetic counseling, and rationale for life-long treatment and any adjustments needed during normal child and adolescent progression and as needed for any co-morbidities in later years.[5,6]

- **Athyreosis**: absent thyroid gland; dysgenesis
- **Hypoplasia**: another type of dysgenesis
- **Ectopic**: usually along "path" from base of tongue to anterior neck associated with complete or partial dysgenesis
- **Dyshormonogenesis**: various disorders of biosynthesis with many genetically defined mutations
- **Transient congenital hypothyroidism**: secondary to maternal TSH or TSH-like suppression

Figure 3: Congenital Hypothyroidism Diagnostic Categories

Congenital hypothyroidism was previously thought to be mostly athyreosis but now is believed to be mostly hypoplastic or ectopic hypothyroid varieties when radioiodine or technetium scans are utilized as well as thyroid utrasonography. As such, thyroid dysgenesis accounts for approximately 85% of congenital hypothyroidism in parts of the world where regular neonatal thyroid screening is performed. It is estimated that 1:2500-4000 births involve congenital hypothyroidism. Disorders of hormone synthesis either full or partial dyshormonogeneis accounts for 1:40,000 births wherein thyroglobulin synthetic defects and iodothyronine deiodinase defects are the two most common forms of such dyshormonogenesis. Hypothalamic (TRH: thyroid releasing hormone) abnormalities and pituitary (TSH: thyroid stimulating hormone) deficiencies account for another 1:20,000 births.

Hypothalamic congenital hypothyroidism includes series of known diagnosis including septo-optic dysplasia, pituitary hypoplasia and HESX1 gene mutations abnormalities. Holoprosencephalic mutations involving SHH, SIX3 and ZIC1

mutations also go in this category as do other even more rare hypothalamic gene mutation syndromes. Pituitary gland genetic mutations involve embryogenesis with or without other hormone deficiencies and some of these types of mutations can be familial patterns. These include TTF1 with pituitary and thyroid aplasia as their hallmarks, LHX3 and LHX4 embryogenesis problems, Prop-1 and Pit-1 mutations associated with programing and pituitary cell mutations as well as thyroid embryogenesis factors identified so far: NK2.2 (TTF-1), FOX#-1 (TTF2) and PAX8. Specific conditions that include syndromic congenital hypothyroidism include Pendred, Bamforth-Lazarus, Ectodermal dysplasia, Kocher-Deber-Semilange, glaucoma associated with hypothyroidism, chorea and choreoathetosis associated with hypothyroidism, Down Syndrome and Turner Syndrome. (*Figure 4*)

- Pendred: hypothyroidism, deafness, goiter: chromosome 7
- Bamforth Lazarus: hypothyroidism, cleft palate, spiky hair
- Ectodermal dysplasia: hypohidrosis, hypothyroidism, ciliary dyskinesia
- Kocher-Deber-Semilange: muscular pseudohypertrophy, hypothyroidism
- Benign chorea or choreoathesis with hypothyroidism
- Congenital glaucoma associated hypothyroidism
- Down Syndrome associated congenital hypothyroidism (different from increased but later-onset autoimmunopathies associated with Down Syndrome including type 1 diabetes mellitus, Hashimoto's thyroiditis, celiac disease
- Turner Syndrome associated congenital hypothyroidism (also with unrelated and usually not congenital onset increased autoimmunopathies such as Hashimoto's thyroiditis)

Figure 4: Syndromic Congenital Hypothyroidism [1]

Neonatal screening for hypothyroidism

Routine newborn screening in Canada and the United States started with T4 first then followed by TSH on heel stick paper blots while most of Europe began with TSH primarily

then followed by T4. With improved technologies for TSH measurement, this has changed somewhat with more programs primarily checking TSH and then T4 followup. Some programs utilize both T4 and TSH together on spot testing for more specific diagnoses.[1,2,7] T4 screening is believed to detect the more rare secondary and tertiary congenital hypothyroidism variants whereas TSH alone screening would tend to miss such infants since screening would commonly detect low T4 values caused by all levels of abnormalities of the thyroxine synthesizing pathways whereas pituitary and hypothalamic thyroid abnormalities would not be detected when looking for elevation of TSH and not low levels of TSH in the neonate. Using absolute cutoff normal results as well as concomitantly repeat checks on those automatically below a certain cutoff for laboratory analyses by standard deviation comparisons helps to minimize day-of-testing variabilities in the normal changes in T4, TSH and TRH in the neonate.

It is understood that about 5% of congenital hypothyroid infants are missed by screening programs because of "human" errors [8] in specimen handling, missed screening errors or related to transfer from one hospital to another hospital, results reporting/notification or variabilities of thyroid functions dependent upon sample timing post-birth. Generally testing is done at 1-3 days after delivery to allow for current common early discharge from hospital units in recent years. Understanding effects on neonatal thyroid function and response to labor and delivery as well as effects associated with prematurity, SGA or neonatal sepsis on thyroid metabolism [9] also comes into question when thyroid screening results are evaluated. Pitfalls in such screening program were presented with recommendations, when adopted, which minimized such problems.[10]

Such conditions may be indeed, temporary effects and the otherwise normal neonate thyroid system makes such corrections on its own although identifying if this is about to happen can be challenging at times. Followup laboratory assessment of total T4, free T4 and TSH usually solve this

problem. A maternal or other family history of autoimmune thyroid disease also raises suspicions of the possibility of a transient congenital hypothyroid condition perhaps related to transplacental factor(s) transmission and/or autoantibodies themselves.

Familial thyroid diagnoses, especially those which are congenital conditions, could suggest thyroid dyshormonogensis although most of these conditions are believed to be autosomal recessive conditions rather than dominant inheritance. Other types of congenital hypothyroidism would also include with mostly elevated TSH levels and normal or only slightly low T4 levels so that the type of screening details may not allow all such conditions to be recognized without further followup levels and perhaps also assistance of a well trained and experienced pediatric endocrinologist/thyroidologist.

Transient congenital hypothyroidism is believed to occur in about 5-10% of screening results and reflect transplacental passage of autoantibodies effecting the neonate in about 1-2%. Some such transient problems may reflect how the infant's brain-hypothalamus-pituitary-thyroid system is functioning or may reflect such abnormalities linked to other diagnoses: very sick neonates "under stress", prematurity or an infant small for gestational age, for instance. Endemic iodine deficiency of mother and fetus also is not rare in many parts of the world.[11] Maternal iodine ingestion excess can cause similar problems when it occurs. Maternal anti-thyroid medication correctly used for maternal hyperthyroidism during pregnancy can cross the placenta and affect the fetus and neonate. Any seriously ill neonate can also have transient thyroid function abnormalities and therefore also require followup thyroid function tests.[12] Prematurity and small for gestational age (SGA) infants also should be evaluated as part of the entire neonatal thyroid screening program but both these latter two categories need followup assessments according to their progress, treatment needed and co-morbidities several times post-delivery. Specific attention to normative thyroid function values for the

premature infant is also now available.[13] Mandated automatic retesting protocols in many programs have allowed better identification of those with transient thyroid function abnormalities that self-correct on rechecking where these protocols exist and are faithfully followed.

Data from the Florida state newborn screening program for 2009-2010[5] where both T4 and TSH are routinely measured in all neonates and provide some interesting perspective (*Figure 5*):

	2010	2009
Live Births	214,936	221,391
High TSH	1,119	1,232
Confirmed Congenital Hypothyroidism	68	68
Congenital Hypothyroidism Incidence	1:3,161	1:3,256

Figure 5: Florida Newborn Screening Thyroid Incidence 2010/2009

Congenital hypothyroidism is one of the most common preventable causes of mental retardation since early diagnosis in the neonatal and post-neonatal period would allow earlier treatment and therefore sooner correction of any hypothyroid related cerebral damage.

With the overall incidence of approximately 1:2500-4000 infants, there is an approximately 2:1 female:male ratio of congenital hypothyroidism for reasons still unexplained. There also is a slightly higher incidence in Latino populations (from the Americas and/or Caribbean) and slightly lower incidence in African-American populations compared to European based Caucasian populations. Down Syndrome[14] also is associated with slightly higher congenital hypothyroidism. World-wide with most infants born in low resource countries (LRC) and medium resource countries (MRC), iodine deficiency[1,2] remains the most common preventable cause of mental retardation worldwide.

In recognition of this fact, iodination of salt established by HRC (and some MRC) governments has corrected this now

preventable problem so hopefully this will continue to increase worldwide as more and more governments help to establish formal neonatal congenital screening as well with the knowledge that only identifying such problems in the neonatal period post-partum costs less than the lifelong costs that otherwise accrue.[15]

History of Newborn Screening [16]

Pilot newborn screening programs originally were only for PKU (phenylketonuria) but in 1974 Quebec, Canada and Pittsburgh, PA, USA began congenital hypothyroidism screening added to PKU neonatal programs already in existence and were soon followed by the New England Congenital Hypothyroidism Collaborative incorporating the neighboring states of Massachusetts, Connecticut, Rhode Island, New Hampshire, Vermont and Maine.

Similarly a multistate program was added in the northwestern United States as well and soon all parts of the United States had similar screening programs established. These programs hoped to prevent growth abnormalities as well as attempt identification and correction of missed congenital hypothyroidism with earlier institution of diagnosis and treatment in an effort to decrease or prevent so many of the commonly seen issues with congenital hypothyroidism before such screening. Now such programs, having shown enormous success in quality of life, CNS functioning, growth abnormalities and family distress throughout established in Europe, Asia, Australia, New Zealand, North, Central and South America and some parts of Africa. In North America (USA, Canada and Mexico) more than 5 million newborns are currently screened and approximately 1,400 infants with congenital hypothyroidism are detected annually! Cost of screening[17] is much lower than the cost of diagnosing and managing congenital hypothyroidism at an older age because the financial and social costs of mental retardation and its associated co-morbidities and education hurdles are minimized or avoided. For the child/teen/adult with congenital hypothyroidism and for that person's family,

quality of life significantly improves even with lifelong oral levothyroxine treatment necessary on a daily basis and appropriate followup medical lab testing and clinical followup also lifelong. Noncompliance with such daily medication especially surfaces in or around adolescence with less overt parental supervision and in adults who can become lax with such requirements and minimal symptoms when medication compliance persists until neurologic and/or cardiovascular problems – preventable - present.

Thyroid Physiology [1,2,7]

The thyroid gland develops from the buccopharyngeal cavity fourth brachial pouches between 4 and 10 weeks gestation. By 10-11 weeks' gestation, the fetal thyroid is capable of producing thyroid hormone but fetal thyroid hormone concentrations are low in the first half of pregnancy. By 18-20 weeks' gestation, blood levels of T4 have reached term levels. As a consequence, during a good portion of fetal development, the fetus is entirely or partially dependent on maternal thyroid hormone and iodine for effects of body development especially neurologic development. Supply to the fetus is believed to be controlled by the placenta as well as maternal thyroid-pituitary-hypothalamic feedback loops working in combination to maintain a "euthyroid" state.

Despite critical importance of thyroid hormone on multiple organ systems, especially the brain, most infants with congenital hypothyroidism appear overtly normal at birth; it is only the most severe neonates who can be readily diagnosed by examination in the delivery room or nursery: exceptionally quiet baby/dull affect, large anterior fontanelles, enlarged and protruding tongue (see *figure 1*). This missed diagnosis on clinical grounds reflects the subtle clinical abnormalities documented to be present, in retrospect, when clinical physical notes are reviewed following newborn screening notification.

However, except in cases of extremely severe thyroid function results at confirmation, when clinical suspicion may already have existed, clinical diagnosis is rare without weeks

or months of hypothyroid effects on the growing infant – then, more obvious symptoms present or there is some deterioration by history or exam. In general, the hypothyroid fetus appears to be protected somewhat by placental transfer of maternal thyroid hormone. Infants with some of the dysgenetic causes of their congenital hypothyroidism who are unable to synthesize their own thyroxine so that cord blood T4 levels are ⅓ to ½ that of normal infants. Also, some studies have demonstrated what appears to be increased intracerebral conversion of T4 to T3, resulting in increased local availability of cerebral T3 despite low serum levels of thyroid hormones under such circumstances that also may explain lack of prominent and diagnosable physical findings in the neonate.

Specific neonatal thyroid considerations are somewhat different than later periods of life. Marked neurologic impairment is seen if both maternal and fetal hypothyroidism are present despite adequate therapy shortly after birth. This is predominantly from conditions where severe iodine deficiency exists as is common around the world unless governments have regulations which are enforced to provide iodinated salt. In addition, and unrelated to such iodine deficiencies in the world's food chain, potent thyrotropin receptor blocking antibodies also exist, although quite rare, which affect the fetus. Also, rare maternal-fetal PIT-1 deficiency can occur with pituitary specific transcription factor errors for somatrope (growth hormone), lactotrope (prolactin) and thyrotrope (thyroid stimulating hormone) function.

Newborn Screen Protocols[1,2,7,18]

Hospitals usually obtain heel-stick blood samples and such small volumes can be sent to centralized laboratories established specifically for newborn screening analysis.

Many programs have added not only neonatal hypothyroidism screening program to PKU programs already in existence but multiple other endocrine and metabolic screening that can be done at relatively low cost vs. the extended and enormous life-long costs of missed diagnoses, missed opportunities for earlier treatment and the

improvement in health status as well as quality of life for the child and family that makes them worthwhile to consider.

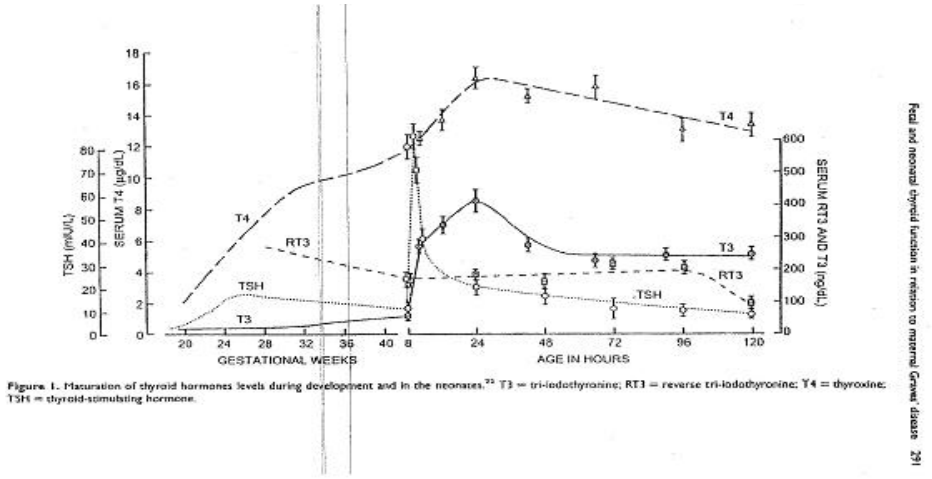

Figure 6: Intrauterine and neonatal TFT's [1,2,7]

Figure 6 - presents some study results evaluating thyroid functions during intrauterine and neonatal time periods. Understanding these stages of development and function allows for improved cutoff laboratory values based not only on individual assay variability and standard deviations, but also defining potential cutoff values based on gestational age, timing of birth and post-neonatal age for both screening protocols as well as evaluation of confirmatory thyroid functions.

Taking into account the normal perinatal changes that are now well recognized as shown in figure 6, there is some discussion ongoing about the optimal timing of sampling after birth. Most program protocols consider sampling of the neonate after 24 hours of life to try to miss the normal neonatal TSH surge and allow at least one or more feedings to take place. This is sometimes not always possible with early discharges of otherwise normal neonates in many hospitals as well as when there is some other medical urgency, ie. NICU admissions or other medical conditions that can interfere with optimized thyroid metabolism and function.

In the NICU, for instance, one such protocol suggests thyroid and other screening be done on admission to the NICU, day 7, day 21 as well as 48 hours after total parental

nutrition has been completed as well as prior to any blood transfusion in an attempt to minimize interpretation dilemmas. Of extreme importance when setting up and supervising such neonatal screening programs is to establish methods for oversight [8] to try to minimize neonate missing screening testing and to make sure that there are sufficient methods established for being able to communicate with primary care providers and parents if test results are delayed until after the newborn leaves the hospital unit. When neonates are transferred from local facilities to more specialized centers, sometimes the timing of the transfer also allows for missed screening of the infant.

Specifically, discharge orders must be established with protocols for nursing staff supervising newborn discharges to document that the newborn screen was performed and that the results obtained also were documented as normal or abnormal, needing followup with an endocrinologist or primary provider, thyroxine treatment or repeat blood work etc.

Thyroxine cutoff values are somewhat different in different centers and countries but approximately a T4 cutoff of 6.6 ugm/dl for the first week of life while a T4 cutoff of 5.0 ugm/if the neonatal sampling was obtained after 8 days of life. TSH cutoffs for such neonatal screening programs are approximately >23 uIU/ml. requiring followup assessment, repeat testing and formal decisions about treatment needs. Knowing the specific assay normative values and paying attention to sample timing postnatally as well as gestational age of the newborn, help to make such assessments better while using mathematical laboratory standard deviation analysis helps make the screening systems better as well as more cost-efficient.

If the T4 levels are below these cutoffs and/or the TSH level are too high (or inappropriately too low considering the T4 levels obtained, specific steps must be taken and documented per protocol with attention to quality control requiring periodic independent assessment of any such program. Individually established interpretation of lab

determinations should be based on normative data for the country, state, province and/or city by thyroid specialists so engaged when the programs and laboratories were set up and with due consideration for the specific laboratory methodology in use.

Primary congenital hypothyroidism	**Diagnostic Tests to Consider**
Core tests	Either primary T4 screen with followup TSH or primary TSH with followup T4 or combination T4 and TSH screening
Followup testing	T4, TSH free T4, T3 uptake
Supplemental testing	Serum TBG
	Anti-thyroid peroxidase and thyroglobulin antibodies
	I^{123} or Te^{99} thyroid uptake studies
	Thyroid ultrasonography if radioisotope scans negative
	Bone age
	Specific genetic and/or molecular evaluation

Figure 7 - Diagnostic Tests for Congenital Hypothyroidism[19]

As with all other such matters, specific protocol follow up should be reviewed and documented often with a committee of several interested pediatric endocrinologists, neonatologists and other specialties of interest. For any results from screening, follow up confirmatory T4 and TSH levels should be obtained as recommended. Some would also include T3 and resin T3 uptake levels as well as free T4 levels as well as thyroid ultrasonography to document presence and site of the actual thyroid gland, ectopia and perhaps pituitary/hypothalamic imaging as well if there are some suspicions of secondary or tertiary thyroid abnormalities.

Screen Results	**Probable Cause**	**Clinical Manifestations**	**Special evaluation/care needed**
T4 low, TSH high	Primary hypothyroidism	None → severe	Repeat TFTs & l-T4 Rx
	Maternal antibodies	Often none	Same + both Ab's
	Maternal anti-thyroid Rx	Often none	Same
	Maternal iodine	Often none	Repeat TFTs
T4 low, TSH ok	Possible 1 hypothyroidism ? 2 or 3 hypothyroidism	None → severe	Repeat TFTs Consider pituitary [20] or hypothalamic problem or other transient conditions, sick NB, SGA, prematurity, TBG deficiency etc.
T4 ok, TSH high	Possible hypothyroidism	None → severe	Repeat TFTs then consider other testing
TSH very high with or without low T4	Primary hypothyroidism	None → severe	Repeat TFTs and consider other testing
	TSH post-natal surge	None	Repeat TFTs and consider other testing

Figure 8. Interpretation of Initial Newborn Screening Results for Congenital Hypothyroidism [20]

Radioactive iodine and technetium scans [21] also can help to better define exact diagnoses accordingly. Ultrasonography and radioactive scanning should not be delayed for scheduling reasons, however, and protocols should define such considerations very specifically.

Levothyroxine Treatment[1,2,7,22]

Any infant with low T4 and elevated TSH is considered to have primary hypothyroidism and further evaluation should usually be the responsibility of a pediatric endocrinologist with experience and interest in pediatric

thyroid disorders. If no pediatric subspecialist is available, then discussions with an adult endocrinologist interested in thyroid disorders should be considered as well as any knowledgeable neonatologists or general pediatricians so interested.

With current international telephone communications, email and zoom meetings can be arranged with pediatric endocrinologists/thyroidologists to provide such consultation as individual situations warrant quite readily. Sometimes, genetic specialists are also involved especially when syndromic conditions are being considered and here, too, international cooperation works to fill the gaps in local expertise. Similarly, neurologists may be called for consultation although most endocrinologists would be able to obtain appropriate sampling for consideration of pituitary or hypothalamic dysfunction and testing. This is especially important of hypothalamic or pituitary insufficiency is being considered since subtle secondary or tertiary adrenal insufficiency under such circumstances can become rapidly lethal if thyroxine is instituted without prior cortisol. If there is any suspicion of adrenal insufficiency, then cortisol and ACTH levels, if available, should be obtained and appropriate cortisol treatment instituted before levothyroxine is provided. As before, emergency consultations with an adult endocrinologist and/or radiology, genetics, neonatology should be considered.

If such specialists are not available then consultation or referral to centers of excellent should be part of the treatment team. Delay in contacting home medical care teams and primary providers, especially in MRC and LRC situations where they may not be easily available because of financial barriers, socioeconomic or political barriers and simple nonavailability needs to be avoided.

Confirmatory testing if not done in the nursery/hospital, needs to be done without delay as well and sometimes treatment with levothyroxine should be started even before such confirmatory results are availability to prevent further treatment delay.

Initial neonatal levothyroxine dose is 10-15 ugm/kg/day given usually once each morning with a crushed pill by the parents prior to the first morning feeding. This be placed in the infant's cheek and then breast feeding proceeds to wash the levothyroxine into the stomach for absorption. If bottle feeding is the feeding method of choice, the same protocol can be used and the crushed tablet of levothyroxine can even be placed in the rubber nipple, and then the milk from the bottle washes the medication through the oral cavity for absorption.

More recently there have been some liquid formulation of levothyroxine that may have some place in the therapeutic armamentarium of neonatal hypothyroidism as more studies are done of safety and efficacy as well as bioequivalency and proper absorption. Soy formulas may be associated with decreased thyroxine medication absorption and, similarly, for later consideration, high fiber and higher iron content of food provided so further testing and dose adjustment may be recognized and made. There remain some controversies over the equivalency of generic levothyroxine tables compared to more expensive brand-name products especially with cost considerations around the world.[23]

Goals of Newborn Thyroid Therapy[24]

Ideally, normalizing T4 by 2 weeks and normalizing TSH by 1 month of age would be reasonable therapeutic goals and there are several studies which suggest that earlier and more rapid correction with documented normalization of T4, free T4 and TSH is associated with better neurodevelopmental outcomes with or without screening identification and diagnosis. If a higher initial dose of levothyroxine is used, 12-17 ugm/kg/day, serum T4 normalizes within 3 days and TSH can return to target by 2 weeks in some published studies. Some studies have also suggested that rapid correction of TSH has cognitive benefits but one study also showed overcorrection with higher T4 results has some deleterious effects. Most experienced pediatric thyroidologists tend to rapidly correct neonatal hypothyroidism since this lone study

has not been replicated elsewhere and clinical evaluations do not show any obvious harm from more rapid rather than slower corrections in these infants. T3 (liothyronine) should not be used for treatment, even though T3 is more biologically active than T4, since most brain T3 is derived from local monodeiodination of T4 rather than from circulating T4 itself.

Follow up should ideally be by a board certified pediatric endocrinologist, assuming one is available soon after diagnosis, if not already evaluated by a pediatric endocrinologist in the nursery. An adult endocrinologist or specially trained and interested general pediatricians can also provide such services if a pediatric endocrinologist is unavailable if they have appropriate experience.

Follow up free T4 and TSH should be obtained with a complete history and physical exam at 2 and 4 weeks after initiation of levothyroxine treatment. Some would also include total T4 as well as free T4 and TSH and a flow chart should be part of the computerized medical record (or handwritten medical/lab chart) to facilitate ongoing analysis and act as a critical guide for dose adjustments as well as compliance issues.

Thereafter follow up[1-3,7-8,10-13,25] should usually occur about every 1-2 months for the first 6 months of life, every 3-4 months between 6 months and 3 years of life and then about every 4 months to follow normal growth and development needs and adjustments of dosing, optimize compliance. Side effects with levothyroxine are extremely rare unless an inadvertent overdose is provided either by parental error or pharmacist error. When generic levothyroxine is used instead of brand name levothyroxine products, changes from one company to another sometimes produce a need for dose adjustments so this too should be routinely questioned at followup visits and becomes a potentially important education issue for parents. Quality control of generic medications around the world even in HRC sites does not always guarantee optimal quality control. More frequent followup should often be arranged whenever dose adjustments are recommended to document improved lab results and/or

associated symptoms amelioration. Compliance issues [26] with any chronic illness, congenital hypothyroidism included, must address financial barriers, insurance barriers and all other issues involving compliance of a daily task like pill taking.

Developmental outcomes should include expectation for normal weight and height progress documented by weight and height charts kept in the medical computer record systems and/or with written well documented weight and height charts.

Twenty years ago, there were minor differences in IQ, school achievement and neuropsychological tests in congenital hypothyroid adults when congenital hypothyroidism was treated earlier compared to unaffected classmates and siblings. Nowadays with more screening programs including congenital hypothyroidism around the world, more recent studies testing such youngsters at school entry showed no difference compared to siblings with the protocols discussed and with "more rapid" correction of circulating free T4, total T4 and TSH reached and maintained.[27,28]

CONGENITAL ADRENAL HYPERPLASIA (CAH): ADRENOGENITAL SYNDROME (AGS)[29]

Congenital adrenal hyperplasia[30,31] in its most common form, 21-hydroxylase deficiency, can be suspected in children with three clinically overt problems: in female neonates with ambiguous genitalia or enlarged clitoris with evidence of sodium deficiency or dehydration and in any male or female child with progressive and/or early virilization.[32,33] Because 21-hydroxylase deficiency CAH is autosomal recessive for the CYP21A gene (also called P450c21)[34], incidence should be the same for boys and girls but the absence of genital ambiguity in males and often the subtlety of any male neonatal or early infancy virilization is not so apparent on clinical examination compared to changes in genetic females of the clitoris and/or labia. Decrease of this loss of 21-hydroxylase enzyme function interferes with ability to make cortisol and aldosterone so that

the adrenal cortex is stimulated by hypothalamic and pituitary factors and overproduces various cortisol precursors. Some of these precursors are diverted to the biosynthesis and overproduction of sex hormones which are associated with androgen excess including the ambiguous genitalia in newborn girls and rapid postnatal growth in both sexes as well as early/excessive virilization.

Some but not all forms of the 21-hydroxylase deficiency also interfere with mineralocorticoid pathways and that explains the salt wasting and consequent failure to thrive, hypovolemia, shock and deaths. There is a milder nonclassic form that may be asymptomatic or associated only with signs of postnatal androgen excess. Assessment of sodium and potassium levels as well as acid/base status, usually with bicarbonate levels as well as hydration and BP status can be lifesaving, particularly in the more severely affected patients. Assessing cortisol (low results), ACTH (high results) and 17-hydroxy-progesterone results (high levels), assuming that these measurements are available, can lead to early consideration of this diagnosis; closer clinical monitoring as well as emergency institution not only of salt and water replacement but also emergency cortisol replacement corrects the electrolyte and volume deficits to prevent imminent death.

Congenital adrenal hyperplasia[35,36] can be caused by a variety of enzyme deficiencies in the adrenal cortex. There are six major forms of CAH [37] each felt to be caused by a mutation of one of the six enzymes required for the biosynthesis of cortisol. The enzymatic processes are involved with very specific clinical signs and symptoms[29-37] that depend on whether the specific enzyme absence is full or partial and whether the enzyme deficiency results in high or low production of the various steroid products in the mineralocorticoid, androgen or glucocorticoid systems. The decrease in cortisol secretion results in a decrease in negative feedback at the level of the hypothalamus-pituitary system such that the resulting increase in hypothalamic CRF (corticotropin releasing factor) and pituitary ACTH (adrenocorticotropic hormone) attempts to produce more

cortisol – if the mutations allow some degree of enzymatic activity in the adrenal gland. At the same time, such increases in CRF and/or ACTH produce elevated production of the usual cortisol precursors before the blocked pathway site, and this can add to different clinical manifestations and difficulties and modified by whether there is a full or only partial enzymatic block present.

The pathways of steroid biosynthesis in the adrenal cortex (*Figure 9*) are now well recognized and confirmed. Understanding this sequence and which enzymes are involved allows for better understanding of the pathophysiology and clinical findings in each circumstance on presentation as well as which measurements are needed to assess treatment compliance and efficacy.

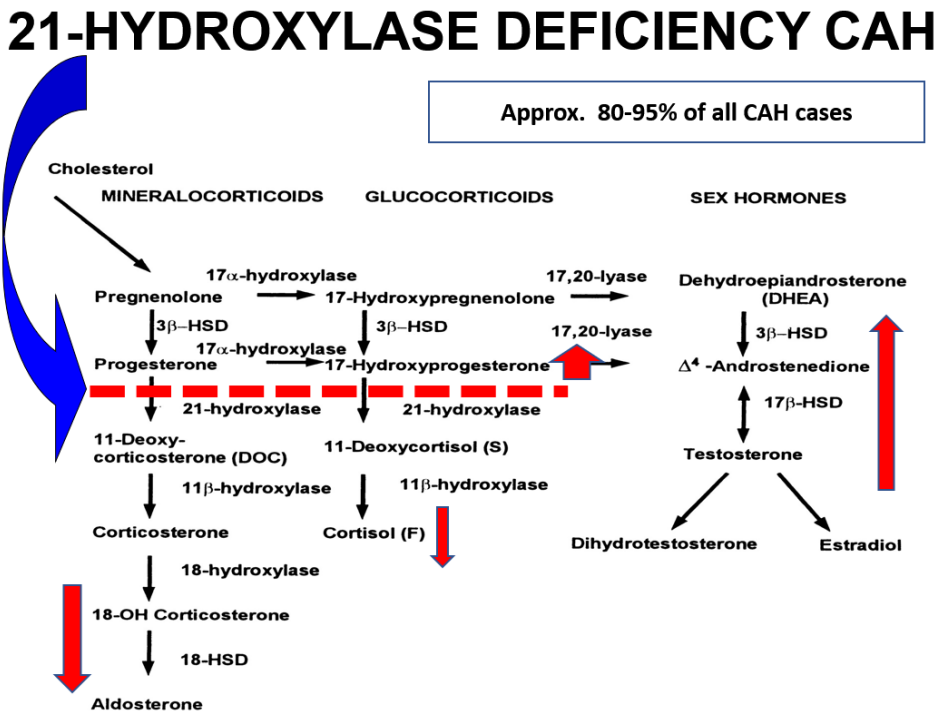

Figure 9: Pathways of Adrenal Cortex Steroid Biosynthesis [29-45]

21 hydroxylase deficiency presents in girls with classical CAH having clitoral enlargement, labia majora which can be partially or totally fused and/or a common urogenital sinus instead of separate urethra and vagina. Variable extent of virilization from nearly full male appearance, and thus the risk of misapplication of gender assignment as a boy with absent testes, when the newborn is actually a genetic female with excess androgen causing the problem. More severe salt-wasting variants show up "sooner" and often in those females with obvious genital ambiguity - but still with a great deal of variability from patient to patient.

Electrolyte decompensation and hypovolemia often does not become apparent clinically until the second week of life so in the more subtle male clinical presentation there is no ambiguous genitalia issues to "warn" health care providers of what's brewing. As a result, recognition of salt-wasting clinically seems to occur "earlier" in girls vs boys once a differential diagnosis of virilization occurs. Since the peak of such mineralocorticoid insufficiency usually occurs post-nursery discharge, the males, otherwise usually unrecognized as virilized clinically, get sick while at home, become dehydrated, progress to hypovolemia shock and may die before the diagnosis is actually considered or before treatment can occur, whereas the girls get diagnosed earlier because of their more overt virilization. As a result, the diagnosis at birth of a female with simple virilizing CAH often is made sooner and more often on clinical grounds because of the apparent and obvious genital ambiguity.

The clitoris is often enlarged in such genetic females because of the androgen excess and has an appearance similar, but not quit a normal penis, at the same time that there appears to be hypospadius and/or cryptorchidism if the labia are greatly fused.

Nonclassical partial or later onset females with 21 hydroxylase CAH may present without salt losing but with unexplained increased pubic hair, clitoral enlargement, oligomenorrhea, hirsutism and excessive or early acne as well as accelerated growth and bone age skeletal maturation since

there is some mineralocorticoid and some cortisol production possible but still relative androgen excess.

Males with 21 hydroxylase deficiency, even when they have significant excess androgens measurable on lab testing, sometimes will appear to have an enlarged penis but males will usually have normal scrotal sacs and normal appearing and size of testes. Many time such genital changes externally are only recognizable in retrospect and after the when the chemical diagnosis has occurred - or the salt losing crisis has brought them back to the hospital.

In the nursery most examinations are not sufficiently abnormal to raise the possibility of early clinical diagnosis and most of the salt wasting crises don't happen the first few days or week of life but rather 1-4 weeks after birth. Such salt wasters are mostly males, appear as failure to thrive infants, unable to regain their birth weight, frequently with vomiting, significant dehydration and hypotension and too often in shock due to the severe mineralocorticoid and glucocorticoid deficiency.

Nonclassical males may not always be salt wasting under usual circumstances but under stress conditions (illness, surgery) this may appear. They also may present with unexplained early pubic hair, hirsutism, early or sever acne and have phallic enlargement, accelerated growth and bone age skeletal maturation.

CAH/AGS Incidence and Pathophysiology

More than 80-90% of CAH is thought to be caused by a deficiency of 21-hydroxylase with an incidence of approximately 1:10,000-1:15,000. It is also thought to be quite similar all around the world but where there is increased consanguinity, as in some more isolated tribal communities, the incidence of rarer forms of CAH increases. For example, 1:300-700 in Alaskan Yupik Eskimos but unknown if this also occurs in Canadian or Russian indigenous populations. Classical 21-hydroxylase deficiency, 21OHD CAH, is characterized by too low cortisol levels, increased ACTH levels

as the body tries to boost cortisol production in standard fashion and is associated with increased androgens so that testosterone, dihydrotestosterone and androstenedione levels are increased with virilization present.

The androgen increase is a consequence of the ACTH increase and pushing the cortex to make more of these androgens is "allowed" because of where the 21 hydroxylase enzymatic block occurs in the synthetic pathway ("towards the right side").

The virilization in females can be rather severe and the more so because 21OHD CAH in its most severe manifestations not only decreases cortisol secretion but also because the enzymatic block allows exposes of the very early female fetus to masculinize the genital precursor tissues. There are also salt losing varieties but with nonspecific demarcation between the mineralocorticoid deficient varieties and those who can more or less maintain sodium and potassium balance unless there are severe condition of stress that intervene. Someone with a simple virilizing form of 21OHD CAH may only show mineralocorticoid imbalance under such extreme conditions, ie. severe illness, surgery. Non-classical 21OHD CAH may escape detection at birth (without neonatal screening programs) particularly in males where the virilization can be significantly more subtle compared to females; the females may appear more like males with underdeveloped penises (in fact, virilized clitoral bodies), labioscrotal sacs devoid of testes since they are, in reality, females with ovaries overwhelmed by the excessive internally secreted androgenic hormones.

Usually, such females can be easier to identify clinically in the nursery compared to the males and so early mineralocorticoid deficiency crises will then be detected earlier in females than males when there is no screening taking place. The virilization of newborn girls with 21OHD CAH may be severe enough that the baby is thought to be a boy – but without palpable testes (see below).

CAH due to 21OHD is the most common cause of ambiguous genitalia. As reviewed in figure 9, classical 21

hydroxylase CAH prevents sufficient cortisol and often also mineralocorticoids to be available and the 17-OH progesterone used as a screening measurement rises since the hypothalamus and pituitary gland feedback loops notice the lack of cortisol presented to them.

Classical salt wasting forms of CAH are thought to have virtually no 21 hydroxylase activity present in about 75% of cases while the classical simple virilizing form of CAH has about 1% enzymatic activity and that appears to be "sufficient" to prevent routine salt wasting unless a stress situation like infection or surgery coexists.

Later-onset nonclassical 21-OH CAH is thought to have about 20-50% of enzymatic activity as its explanation for much later clinical symptoms and signs. Premature infants, low birth weight infants can sometimes present with "stress" false positives on screening and so follow up assessment should include repeat cortisol and ACTH as well as electrolyte measurements.

Infants born to mothers actively treated with chronic or high dose glucocorticoids may also need reassessment of screening results.

17 hydroxy progesterone (17OHP) screening

The goals of newborn screening for 21-OH CAH include detecting boys at risk of salt-losing crises and preventing the associated hypovolemic shock and death that can result as well as preventing incorrect gender assignment of virilized girls.

Development of methodology to measure whole blood 17-OH-progesterone as a marker of the most common type of CAH eluted from a dried filter-paper blood spot [38] either by radioimmunoassay or more recently by use of polyclonal and monoclonal antibodies and the use of either radioactive, enzymatic or fluorometric markers of antibody-antigen interaction.

The immunological measurement of 17OHP in neonates is somewhat nonspecific because the fetal adrenal cortex secretes a large number of compounds that are

immunologically similar and cross-react.[38] Ln addition to the problem of assay specificity, circulating levels of 17OHP burst at delivery, decline rapidly after birth and also decline with gestational maturity so that normal whole blood screening vales of 17OHP must be determined for neonates of varying birth weight and gestational duration as well as for postnatal age, "stress" exposure and assay variability from day-to-day.[39]

Examples of 17OHP neonatal cutoff values that can be considered change a bit when considering weight of the neonate, age when the testing is obtained: if 17OHP >120 ng/ml in an infant weighing more than 2000 grams and less than 12 hours; if > 40 ng/ml in an infant weighing more than 2000 grams but specimen obtained > 12 hours of life; if >153 ng/ml for infants weighing less than 2000 grams at any time then emergency consultation with pediatric endocrinologist and repeat blood work including cortisol, 17OHP, ACTH if available as well as BP monitoring, measurement of electrolytes and re-evaluation of eternal genitalia perhaps also with karyotyping. Many sch screening programs now exist adding on to phenylketonuria and congenital hypothyroidism screening and adding specific genetic analyses for CYP21 as well.[40, 41]

CAH Screening statistics

Screening data are presented in figure 10 for the Florida statewide program for two years as reported as an example of such results.

Too early timing of the blood screening sample may lead to false positives because 17OHP after birth can stay elevated for up to 48 hours. This is especially true for premature infants or very sick infants. Confirmatory testing and detailed physical examination by neonatologists and/or pediatric endocrinologists help to make a correct assessment.

As the program became more sophisticated, the confirmed diagnoses numbers don't change a huge amount but the assessments of the low weight infants (premature and SGA) improve with followup examination and lab results.

	2010	2009
Live Births	214,936	221,391
CAH <12 hours	175	169
CAH >12 hours	220	214
CAH <2000 grams	24	206
CAH	31	94
Confirmed CAH	11	5
Incidence	1:19,539	1:44,278

Figure 10: Florida Newborn 17OHP CAH Screening 2010/2009 [7]

Reports from around the world,[42,43,44,45,46,47] confirm what was originally learned in the Quebec, New England and then Pittsburgh and Toronto programs: about half of the CAH patients diagnosed because of screening programs would have been missed on the basis of clinical presentation and significantly fewer go into adrenal crisis and hypovolemic/shock situations as a result of these successful screening efforts.

Salt losing crisis and potential death

Classical hyponatremic crises typically occur in males with 21OHD CAH about 1-3 weeks after birth, out of the hospital. In circumstances around the world where there is no available, knowledgeable medical care, such infants in LRC and/or MRC usually die in coma from severe hypoglycemia, hyponatremia and hypovolemia and without specific diagnoses unless detailed autopsies are performed. This also can occur throughout the more developed nations (HRC).

Over the past decades in most Western countries, newborn screening has been expanded to include not only congenital hypothyroidism but also screening for elevated levels of 17 hydroxyprogesterone (17OHP) on the second or third day of life to detect 21OHD CAH before serious salt loss appears. The test is not available in most countries of the world or in parts of the world where most births do not take place in hospitals but at home. After the first few days to a

week or so after birth, both boys and girls with salt losing forms of 21OHD CAH then may progress to worsening hyponatremia, hyperkalemia, dehydration, failure to thrive, vomiting, hypotension, hypovolemic shock, coma and death if not diagnosed and treated not only with salt and fluids but with glucocorticoids and mineralocorticoids.

Screening has decreased the occurrence of such preventable situations by making the diagnosis in many males which were not being recognized while still in the nursery and by providing earlier identification of females as well. With such earlier diagnoses now feasible and well documented in screening programs around the world, early institution of cortisol replacement therapy often avoids significant salt losing crises or makes earlier sodium replenishment possible with earlier electrolyte evaluation even when neonates remain asymptomatic.

Because CAH screening is with 17OH progesterone measurements, all of the other kinds of CAH require other methods for their more rare diagnoses. Screening also produces a "false positive" rate of about 1:100 even with statistical methods of interpreting cutoff points for consideration normal and abnormal results.

Genital ambiguity in females and virilization in males

The finding of genital ambiguity at birth is a medical and social emergency with family distress, shock, embarrassment, anger made especially difficult by social conventions where the first questions after is the baby healthy involve whether or not the baby is a boy or a girl. So, under such circumstances, sometimes there can be no apparent answer at first.

Medical personnel must be trained and equipped to deal with the familial, social manifestations of this angst in addition to the best ways to biochemically and genetically figure out the appropriate strategies for diagnosis and treatment. Examples of 46XX females with ambiguous genitalia are presented in figure 11A: clitoral enlargement and some labia prominence. 10B: more significant clitoromegaly,

fused labia but absent gonads. 10C: male-appearing external genitalia but not as prominent clitoral enlargement and absent scrotal gonads from labial fusion.

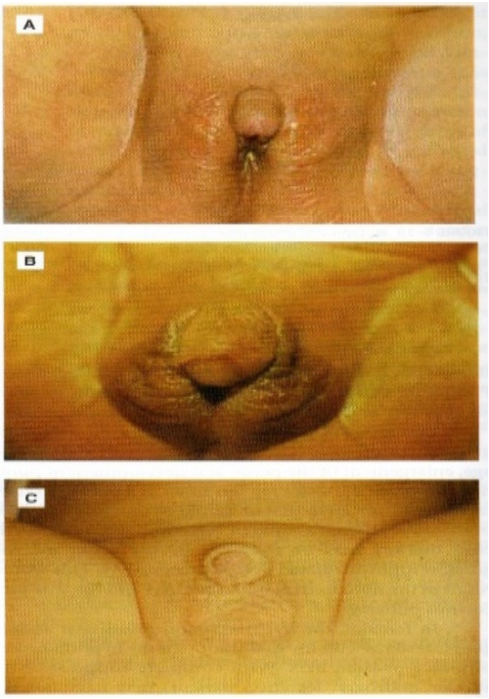

Figure 11 - 46XX Ambiguous Genitalia

CAH is inherited in an autosomal recessive fashion where parents are usually asymptomatic carriers of the gene mutations responsible. The carrier incidence of 21OHD is thought to be about 1:50 and usually parents are healthy carriers of the mutated CYP21A genes.

Both sexes are affected equally from a genetic standpoint despite the fact that more males die in adrenal crisis because of nonrecognition in the neonatal period without 17OHP screening protocols.

Misidentified gender

Without neonatal 17OHP screening available, one of the most distressing situations can arise when a newborn female is clinically diagnosed as a “male with a micropenis and

undescended testicles" if the labia have fused from fetal androgenization. So, the announcement from the midwife or obstetrician to the parents is that they have a "deformed baby boy" and they may or may not have available knowledgeable consultation provided to allow a correct diagnosis.

The newborn boy otherwise has not yet presented with salt losing since that traditionally doesn't occur until several weeks later. Only then does the possibility of a correct diagnosis take place if the family and the baby is fortunate enough to get treatment for the salt losing disaster. With or without a salt losing crisis as described, such an "announced female" when correctly diagnosed and treated must have the family and community, and especially the parents, introduced to the idea that the sex presented to them at birth was incorrect and their baby boy is, in reality, a baby girl with AGS because of excess androgens. Difficult for the health care providers and especially difficult for the parents and family.

With well organized and quality run neonatal screening programs for CAH, such difficulties can be minimized since results of the neonatal screening are generally available with a few days to a week postnatally.

Other tests to consider

Presence of a uterus on ultrasonography in a virilized newborn without palpable testes should raise the diagnostic possibility of virilizing 21OHD CAH and emergency sodium and potassium levels should be checked as should blood pressure monitoring.

Blood urea nitrogen and/or serum creatinine should be checked to rule out renal disease related to electrolyte and acid-base disturbance.

Ultrasonography can also potentially identify ovaries as well as inguinal testes. A buccal smear looking for X chromatin Barr bodies can be done rather quickly but needs an experienced examiner and may be unreliable in neonates. Formal karyotypes would be diagnostic but also take some time for results to return to help established XX or XY situations.

Results of measurements of 17 hydroxyprogesterone that are elevated would be diagnostic if available and if results can be reported without long delays. Tests for mineralocorticoids are expensive and likely will be difficult to interpret or not available with quick turnaround times from laboratories under most circumstances. Other tests to be considered would include assays of other components of the steroid pathways, ACTH concomitant with cortisol levels, urinary 17 ketosteroid and urinary cortisol levels as well as thinking about formal ACTH stimulation testing (if available) particularly looking at 17 hydroxyprogesterone response and other intermediary metabolic products of the steroid pathways, especially the androgens.

Adrenal imaging to rule out adrenal tumors both benign and malignant - but hormone producing - should be considered as appropriate in older children and adolescents who may present only with virilization or premature puberty without salt-wasting forms of CAH.

Testicular adrenal rest tumors[48] in males with CAH do not typically show up until much later in life, although frequently present, so are of more concern for followup endocrinology visits in adolescents and adult male CAH patients.

Variants and missed diagnosis without screening

Those with simple virilizing but not salt losing forms of CAH have intermediate forms of 21 hydroxylase deficiencies affecting more of the glucocorticoid pathways than the mineralocorticoid pathways. Some of these patients are even milder and only present for diagnostic consideration after a severe illness, trauma event, anesthesia or surgical event where the demands are increased and then mineralocorticoid insufficiency shows up. Without neonatal screening for 17OHP, some do not come to medical attention until later in childhood or adolescence because of premature adrenarche or puberty, tall stature otherwise unexplained by family patterns or accelerated height velocity associated with clinical androgen excess. Such early adrenarche or full pubarche does

not typically occur in very rapid fashion but in an early yet progressive pattern so that detailed history may be important to elucidate from the child, teen and/or parents. With the variants of CAH, where 17OP levels are not elevated, then current screening methodology is not likely to facilitate specific mutations and variant CAH situations.

Genital surgery

Surgery to change the appearance of the external genitalia of female infants with the salt-losing and non-salt-losing but virilizing forms of CAH is available but experienced surgeons with support staff that include social workers and psychologists are deemed very important to assist with such complex decisions. Initial approaches included clitorectomy as well as clitoral size reductions but with earlier diagnosis and more introspection, current impetus allows for the realization that with appropriate treatment, androgen excess will be markedly decreased and clitoral size should respond accordingly.

Changes to the vagina and labia themselves, often require some gynecological surgical approach with more attention lately to more preservation of the vascular and neural supply to enhance sexual responsiveness.[49] In girls who are more completely masculinized, often because of subtle situations, medication unavailability or noncompliance, there may be more significant internal anomalies required multiple step surgical procedures.

In some patients, more virilization occurs than would otherwise be expected presumably because there is more intermittent androgenization taking place even with ongoing glucocorticoid and mineralocorticoid therapy or related to missed doses. The reasons for such “refractory androgen suppression” are not well understood but medication noncompliance especially in adolescents and young adults may be explanatory.

Therapeutic approaches have considered bilateral adrenalectomy[50] as a means to reduce the androgen excess, peripheral androgen and blockade accompanied by provision

of estrogen but optimal treatment is not known with a great deal of individual variability.

As such youngsters grow and begin to better understand their CAH and such complex situations, discussion of details of their diagnosis, treatment options, self-care involvement with age and maturity often respond well to psychosocial supportive intervention in addition to nurturing relationships with the physician and nurse clinical experts. There is also some discussion of the prenatal exposure to these excess androgens and whether this "masculinizes" the fetal brain influencing postnatal, adolescent and adult sexuality and behavior but there are no definitive studies and there is much variability in such outcomes to date.

Types of mutation and surgical procedures which took place both need consideration for long-term quality of life in women so affected.[51]

Some studies[52] have documented as much as 50% of CAH women claimed that the disease affected their sex life, were less satisfied with their genitals and felt that clitoral size and function were affected. Many had narrow vaginal canals so that sexual intercourse was uncomfortable or even avoided. This was associated in these women with history of later sexual debut, fewer pregnancies, fewer children and higher than expected incidence of homosexuality.[53]

Non-classical 21-hydroxylase-deficient CAH

Those with non-classical 21OHD CAH are thought to have milder mutations of the CYP21A alleles on chromosome 6 with signs of androgen excess at varying ages and into adolescence or even adulthood. And as with all CAH, salt losing is also variable and sometimes only expressed under stress conditions as previously stated. Anyone with increased linear height velocity otherwise unexplained and associated with even mild androgen excess or irregular periods as well as poor breast development in females should also be considered as having some variant of CAH with appropriate

endocrinologic testing.

Sometimes there is positive family history of such premature adrenarche.

Girls are not necessarily otherwise virilized at birth and in both sexes, the variety of clinical presentation is extremely variable especially if there is no clitoral enlargement or vaginal/labial changes noted.

Differential diagnosis of older children and adolescents as well as adults include virilizing carcinomas of the gonads and adrenal glands, polycystic ovarian syndrome and varying forms of precocious puberty as well as exogenous steroid exposure.

Unilateral testicular enlargement as result of testicular adrenal rest tumors have been reported and routine examination of testes in males with CAH should be taught for self-examination by patients as they reach adolescence and move into adulthood – as well as for health care providers documenting their discussions and examinations in the medical records. This is especially true in transition periods from adolescence when they might normally be cared for by pediatricians and pediatric endocrinologists compared to late teenage years and adulthood when they move to new internists or family physicians as well as adult endocrinologists.

During such transition periods, there is a higher degree of medication noncompliance as well as missed office followup assessments but there are ways to raise awareness of such problems and to begin discussions and facilitate smoother transitions.

Some studies have suggested specific high risk ethnic populations where such late androgen excess conditions may be as common as 1 in 25-30, ie. Ashkenazi Jews or as common 1:1000 individuals (PCOS-like, menstrual irregularities, hirsutism, clitoral enlargement, severe forms of acne, etc.)[54] with those of African descent may have decreased CAH.

Other rarer forms of adrenal insufficiency differentiate from 21-hydroxylase-deficient CAH

Presence of hypertension rather than hypotension should prompt consideration of alternative types of CAH such as classical forms of 11β-hydroxylase deficiency, 17,20 lyase deficiency, 17α-hydroxylase deficiency with excessive mineralocorticoids (hypertension and hypokalemia) at the same time that there is deficient glucocorticoid production contrasts with other variants where some of these CAH forms may only present with sexual infantilism or other forms of gonadal abnormalities (incomplete male differentiation sometimes to the extent of incorrect female gender assignment) depending exactly in which tissues and to what extent such abnormal enzyme aberrations exist.

These rarer forms of enzymatic genetic abnormalities wouldn't be detected with 17OHP screening of newborns but may still present with adrenal insufficiency since they increase mineralocorticoids while decreasing cortisol and decreasing androgens.

Earlier enzymatic pathway deficiencies such as cholesterol desmolase deficiency (lipoid CAH) have low levels of all steroid hormones with decreased or absent ACTH responses but high circulating levels of ACTH. The absence of this very early enzyme causes failure of conversion of cholesterol to pregnenolone very early in the steroid pathway in the adrenals and gonads.

Very early presentation is associated with cardiovascular collapse following increasing severity of adrenal insufficiency and as with many other of these enzyme disorders, milder variants are also reported. Low cortisol, high ACTH levels may offer a clue to this diagnosis.

3β-OH steroid dehydrogenase deficiencies felt to reside in mutations in chromosome 1 in classical salt wasting forms (with decreases in both mineralocorticoids and glucocorticoids as well as androgens) and in non-classical forms (affecting more as abnormalities of puberty, menses with some virilization) also need to be considered under such

circumstances.

Most of the other enzymatic abnormalities in the adrenal synthesis pathways including enzymatic abnormalities in StAR defects of cholesterol synthesis and P450 SCC would be associated with cortisol deficiency and/or mineralocorticoid deficiencies as well as elevated ACTH but related specifically to CAH 17OHP neonatal screening, these would not overlap since 17OHP would not be elevated in those other conditions. When being considered, repeat cortisol and ACTH levels as well as other testing will be helpful in addition to genetic mutation analyses if available.

Diagnostic approach to the infant with suspected CAH is presented in *Figure 11* adapted from Henwood and Levitt Katz[55]

CAH Treatment[33,55,56,57]

In the asymptomatic neonate with confirmed diagnosis, initial treatment would be with hydrocortisone at a dose of 10-15 mg/m^2/day divided in two or three doses although under certain clinical conditions may require up to 25 mg/m^2/day to start treatment with the dose then tapered downward clinically.

Caution with higher doses for too sustained length of time may interfere with growth.

Prednisone can be used at a dose of 2-4 mg/m^2/day usually in two doses per day and dexamethasone can be used at a dose of 0.25-0.375 mg/m^2/day once daily. Many pediatric endocrinologists have experienced problems with prednisone and dexamethasone providing excessive glucocorticoid replacement as well as weight excess and decreased height velocity or growth cessation. However, availability of hydrocortisone cannot always be guaranteed around the world.

Fludrocortisone as mineralocorticoid replacement can be added if sodium levels cannot be maintained with only glucocorticoid replacement and when there is good compliance documented at appropriate dosage with a dose range of 0.05-0.20 mg/day and this can be given only once each morning

but some clinicians prefer smaller doses given equally twice-a-day.

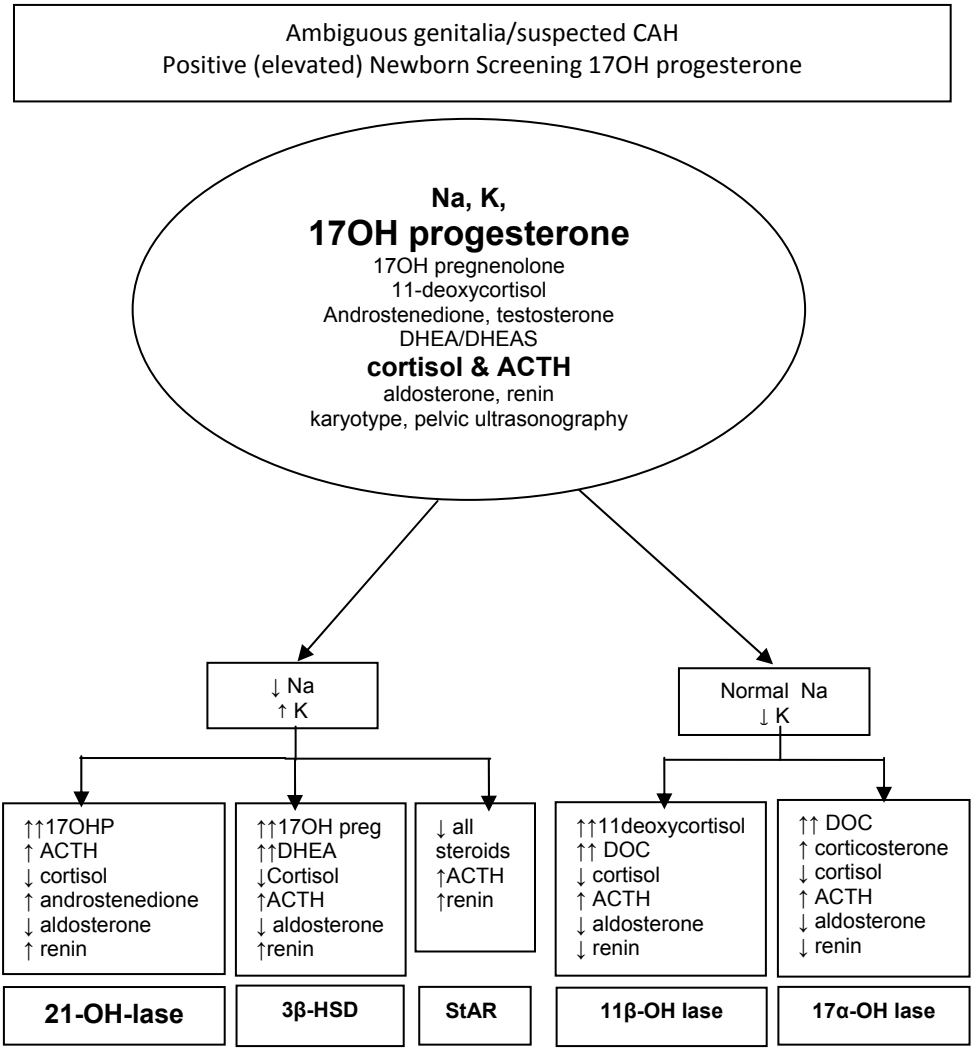

Figure 11: Suspected CAH Diagnostic Approach

Salt supplements can be supplied intravenously in the nursery and sometimes are needed at a range of 17-51 mEq/day but again this is highly variable so that clinical followup and assessment is mandatory.

All infants with ambiguous genitalia should be closely monitored for signs of salt wasting until a definitive final diagnosis is established with appropriate clinical response either intravenously or orally.

Treatment then would depend upon the exact deficiency and its severity and for the classical and variant types of AGS and 21 hydroxylase deficient CAH, screening, awareness, appropriate lab confirmation and repeat lab electrolyte and BP followup will be needed.

Attention to potential for salt losing crisis and high risk of death are the critical issues and these require special education of parents and other family members with periodic review and reminders documented at least yearly in the medical record by health care providers. Poor outcomes and death occur without adequate medical education and sophistication, without adequate special (and expensive) laboratory hormone measurements and without appropriate pharmacy quality control ongoing.

Cortisol and mineralocorticoid medication replacement often can be not available, unduly expensive or burdensome so that these additional barriers also need to be addressed with the parents and eventually with the adolescent and young adult on followup. The special risks of noncompliance with medical followup and with daily medications require specific attention as previously mentioned.

On sick days, with any significant stress event or with planned or unplanned surgery, “sick day rules” should be taught and strictly observed including when and how to contact health care providers, when to come directly to the emergency room (or other urgent care facilities) in an effort to avoid future morbidity and mortality. This includes how and when to provide extra salt intake, home blood pressure monitoring as well as when and how to boost glucocorticoid doses.

Vomiting is an especially problematic issue that also must be discussed and addressed at least annually.

In any or all of these variants of CAH, if there is mineralocorticoid and or glucocorticoid abnormalities, then

sodium chloride, mineralocorticoid replacement, intravenous fluid blood pressure management and/or glucocorticoids all are needed as emergency or urgent treatment measures with appropriate monitoring of all such parameters. Once the emergency situation is under control and parenteral administration is no longer required, then maintenance oral hormone replacement treatment (often with some extra sodium chloride for awhile) will be needed forever as well as education for dealing with intercurrent illness, surgery and other stress situations where extra hormone replacement is warranted. All of these issues must be specifically addressed and written into the paper or computer generated medical records.

When an adrenal crisis occurs, the protocol might include 100 mg/m^2 intravenous hydrocortisone or cortisol, fluid resuscitation with 20 ml/kg normal saline bolus repeated as needed and then with maintenance saline according to clinical response. 5% dextrose with boluses or running as maintenance may also be required if there is concomitant hypoglycemia until the glucocorticoid replacement becomes operative. Repeat doses of hydrocortisone may be needed every 4-6 hours until clinical stabilization.

As a guide, it should be assumed that Solucortef® has an approximate mineralocorticoid activity of about 0.1 mg of fludrocortisone.

Treatment of CAH depends upon the age of the patient, the severity of presentation, whether or not there is need for only glucocorticoid replacement or also mineralocorticoid treatment. This would be directly dependent on the site of the enzyme abnormality and its severity and be reflected in the known variability of clinical presentation as well as lab parameters. Glucocorticoids can be replaced with cortisone acetate, hydrocortisone, prednisone, prednisolone or dexamethasone alone or in combination depending upon availability, cost and individual therapeutic considerations and response.

Overtreatment with glucocorticoids in children can

compromise height but this is no longer a concern in adults and so longer acting glucocorticoids often are used in adolescents and adults compared to children with open epiphyses.

The lowest doses of glucocorticoid to produce the desired beneficial improvement in day-to-day quality of life, optimizing energy and growth while at the same time avoiding the common virilization from underdosing of glucocorticoids (purposeful or accidental) is the therapeutic goal. Similarly, paying attention to mineralocorticoid (and salt) needs particularly in younger babies, infants and children is important and often there is a need for fludrocortisone in addition to glucocorticoids replacement to treat CAH. Here, too, there is great individual variation in need and efficacy; compliance factors with some patients and families are another potential treatment barrier.

Specific salt supplementation is often used in neonates, infants and very young children especially before and up to about 1-2 years of age depending upon such individual idiosyncrasies of that child's type of CAH.

Sex hormone replacement at or around puberty aimed to mimic normal pubertal progression and optimize final height also is required in those types of CAH where sex steroid abnormality co-exists.

Surgical correction of severe ambiguous genitalia (ie. virilization of genetic females) depends upon surgical experience, degree of abnormalities and ability to predict and optimize adult sexual functioning. There remains much debate about what should be offered and done, when it should be done and the entire decision making process under such circumstances in addition to the debates about sexual identify, subtle effects of excess or deficiencies of androgens and estrogens on the fetal and developing brain as well as societal restrictions, religious and cultural norms. Some experiments have shown positive effects using combinations of growth hormone, gonadotropin blockade, aromatase inhibitors in specific circumstances where there is significant height compromise, precocious puberty with demonstrated

improvement in final height and pubertal effects accordingly. With further scientific progress about identifying specific mutations and possibly being able to "correct" the specific genetic defect directly rather than just provide the missing hormones, this too may prove possible with ongoing research endeavors in the coming years.

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PRECOCIOUS PSEUDO-PUBERTY
CAUSES, POSSIBILITIES FOR DIAGNOSIS AND TREATMENT

Camelia Alkhzouz

Introduction

Precocious pseudo-puberty is defined as a plurietiological syndrome that consists in the appearance of secondary sexual characteristics, without activation of the hypothalamic-pituitary-gonadal axis.

The appearance of puberty signs is either due to the autonomous secretion of sexoid hormones in the adrenal cortex or gonads or is of iatrogenic origin. [1]

Precocious pseudo-puberty, depending on the type of secreted hormones, may be iso (secondary sexual characteristics are concordant with the sex of the patient) or heterosexual (secondary sexual characteristics are discordant with the sex of the patient).

Hypersecretion/excess of androgen hormones induces isosexual pseudo-puberty in males and heterosexual pseudo-puberty in females and that of estrogen - isosexual pseudo-puberty in females and heterosexual in males. [2]

Most often, precocious pseudo-puberty is secondary to hypersecretion of androgen hormones from the adrenal cortex and rarely gonadal.

1. Congenital adrenal hyperplasia

Is the best known cause of precocious pseudo-puberty and is due to the deficiency of enzymes involved in the bio-synthesis of steroid hormones.[3] At the level of the adrenal cortex, the bio-synthesis of steroid hormones takes place, starting from a common metabolite, cholesterol. Aldosterone is synthesized in the zona glomerulosa, cortisol in the zona fasciculata, and androgen hormones in the zona reticulata of the adrenal gland.

1.1 21-Hydroxylase deficiency is the most common cause of congenital adrenal hypertrophy (90%) [4]

The incidence of the disease is 1:15,000-20,000 newborns (in some areas the incidence is much higher, even 1:450).[4]

21-Hydroxylase deficiency is an autosomal recessive disease. The CYP21A2 gene, which encodes the enzyme 21-hydroxylase, is located on the short arm of chromosome 6 (6p21.3), near major histocompatibility genes, class C. There are 2 genes in the same locus, an active CYP21A2 gene and a pseudogene CYP21A1.[5] Mutations in the CYP21A2 gene can be deletions/insertions or point mutations. Depending on the remaining enzymatic activity, we can distinguish severe mutations that do not allow the synthesis of a functional enzyme and induce severe forms of disease with salt loss syndrome or milder mutations that allow the synthesis of a variable quantity of functional enzyme and thus inducing a more attenuated phenotype - simple virilizing form, or late onset form.[6]

21-Hydroxylase enzyme deficiency impedes the synthesis of cortisol and aldosterone. Intermediate metabolites accumulated before the metabolic blockade are diverted towards the synthesis of androgen hormones, thus inducing hyperandrogenism.

The virilization process begins in the fetal period (from week 6-10). In genetic female fetuses, masculinization of the

external genitalia occurs; thus, at birth, girls have ambivalent external genitalia, from simple clitoral hyperplasia and posterior fusion of the labia majora (Prader grade 1), to hypoplastic male appearance (Prader grade 5). In genetic male fetuses, the external genitalia are normally developed at birth or slightly pigmented, which may delay the diagnosis [7] Postnatally, virilization continues in both sexes, causing the emergence of pubic hair between 6 months and 2 years of age and of axillary hair between 2 and 4 years of age (*Figure 1*).

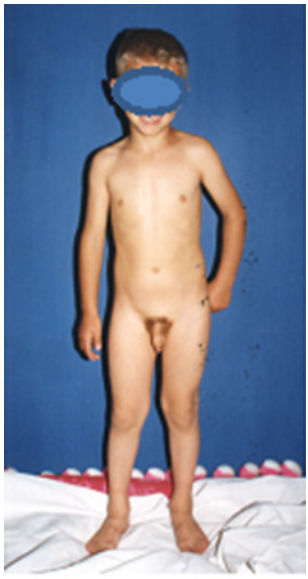

Figure 1. 4 year and 6 months old boy with type I 21-hydroxylase deficiency

The rate of bone growth and maturation is rapid, leading to premature closure of the growth cartilage. These patients are "tall children and short adults." Muscle mass also develops and the tone of voice becomes lower. The final height for girls is between 140-150 cm and 145-155 for boys. In the absence of adequate treatment in girls, female secondary sexual characteristics do not appear at puberty.[8]

In type II, with salt loss, acute adrenal crisis occurs in the first 4-15 days of life through loss of appetite, vomiting, diarrhea, adynamia, severe dehydration syndrome, dyselectrolithemia with hyponatremia and hyperkalemia, acidosis, arrhythmias. [5]

1.2. 11-β-Hydroxylase deficiency is the second leading cause of congenital adrenal hyperplasia (5%).

Its incidence is 1:100,000 newborns. The gene responsible for the enzyme synthesis is located on the long arm of chromosome 8.[9]

The enzyme 11-β-hydroxylase intervenes in the last step of cortisol synthesis.

In the absence of the enzyme, deoxycortisol does not convert to cortisol.

Cortisol deficiency induces negative feedback and, therefore, stimulation of the gland by ACTH.

The enzyme is also involved in the synthesis of aldosterone. In its absence, deoxycorticosterone does not convert to 18-hydroxy-deoxycorticosterone, the intermediate metabolite located before the metabolic block has a strong mineralocorticoid effect causing sodium retention and hypertension.

Due to gland hyperplasia and deviation of intermediate metabolites in androgen synthesis, hyperandrogenism occurs, responsible for virilization.[10]

The clinical picture is similar to that described in the deficiency of 21 hydroxylase, the simple virilizing form (*Figure 2.*) associated with hypertension, with values usually between 150 mmHg (systolic) and 90 mmHg (diastolic). In some cases the values may exceed 200 mmHg.[10,11]

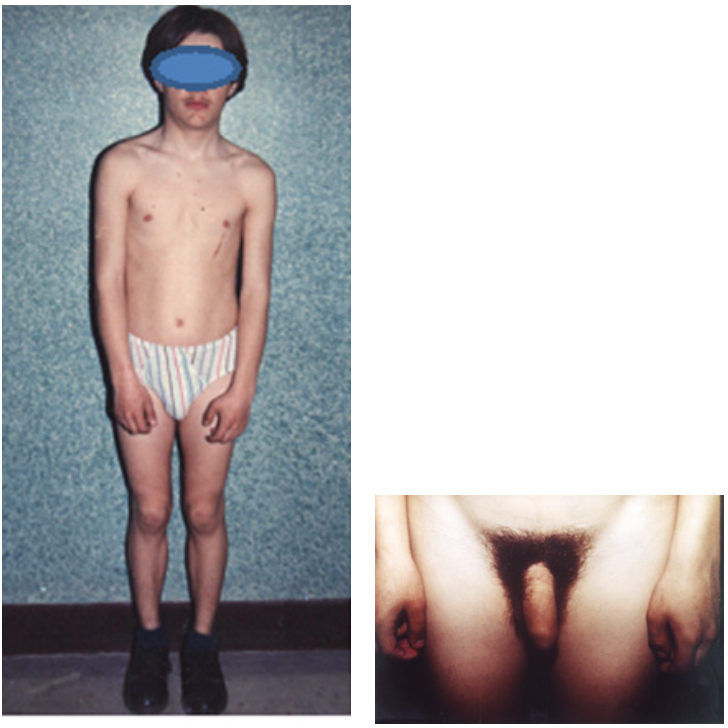

Figure 2. 5 years 8 months old boy with 11 ß-hydroxylase deficiency

Diagnosis.

Cytogenetic analysis is important to be performed immediately after birth in children with ambivalence of external genitalia (SRY gene, karyogram), to determine genetic sex.

Hormonal workup reveal elevated values of androgenic hormones of adrenal origin: 17-hydroxy-progesterone, deoxycortisole and 11-deoxycorticosterone (in the deficiency of 11 beta hydroxylase), dehydroepiandrosterone (DHEA) and dehydroepiandrosterone sulfate (DHEA-S), androstendione and testosterone.

Serum levels of morning and evening cortisol are low and ACTH is high; free urinary cortisol/24h is low; plasma

levels of aldosterone are low and renin is elevated in 21-hydroxylase deficiency.

The dexamethasone suppression test is positive (indicated for non-classical forms).

The parameters of hydro-electrolytic and acid-base metabolism can be modified in the form with "salt loss" (hyponatremia; hyperkalemia; hypoglycemia, decompensated metabolic acidosis).

Imaging assay the ultrasound of the adrenal glands shows their hyperplasia; ultrasound of internal genital organs - in girls/testicular in boys; genitography.

Hand and wrist x-ray reveals the advance of bone age in the simple virilizing/non-classical form.

Molecular analysis of the CYP21A2 and CYP11A1 gene, respectively, allows the identification of the causal mutation.

Treatment goals:

1. Substituting cortisol deficiency and, therefore, interrupting the gland stimulation feedback, thus reducing androgen hypersecretion.
2. Prevention of premature closure of growth cartilage by improving the final height.

Glucocorticoid substitution (hydrocortisone, prednisone, dexamethasone- is used only after the end of growth. Substitution therapy with mineralocorticoids (9 α-fludrocortisone) in 21-dehydroxylase deficiency.[7] Antiandrogens such as cyproterone acetate or spironolactone may also be associated in to reduce hyperandrogenism.

2. Adrenocortical tumors

Adrenocortical tumors are rare in pediatric age group, usually appearing under the age of 3 most of them being malignant (adrenocortical carcinoma) and hormonally active. They constitute about 0.2 % of all pediatric tumors and 5–6 % of all adrenal tumors, with a reported incidence of 0.2–0.3 new cases per 1 million children per year. There is

geographical variation in incidence as it occurs remarkably more frequently in Southern Brazil (12–18 times more than in United States of America or Europe) probably due to the high prevalence of a specific germ line p53 mutation in that population (12).

2.1. Virilizing adrenocortical tumors they induce masculinization of the external genitalia in girls (clitoral hyperplasia but without posterior fusion of the lips); hair growth occurs in hormone-dependent areas (male insertion of pubic hair) (*Figure 3,4*).

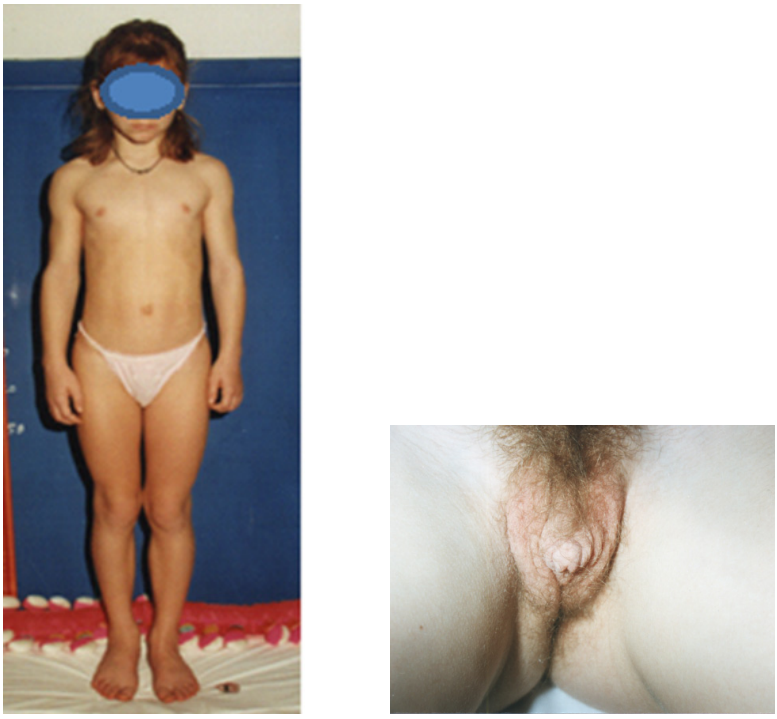

Figure 3. 7-year-old patient with a right androgen secreting adrenal carcinoma

In boys, it causes isosexual pseudo-puberty (adrenarche and penile hyperplasia without testicular growth).

In both sexes, bone maturation and growth rate are accelerated.[13] *(Figure 4*).

Hormonal workup reveals much higher levels of androgen hormones of adrenocortical origin (DHEA, androstenedione) in serum and urine. The dexamethasone suppression test is negative.

Imaging tests (ultrasound, computed tomography and magnetic resonance imaging) may reveal the tumor.[14]

Carcinomas are suggested by the presence of metastases and/or vascular invasion on the pathology analysis.

Large tumor size, internal necrosis, and/or hemorrhage are also features that suggest of malignancy.[15] The Wieneke criteria can accurately predicts the clinical course in childhood adrenocortical tumours and could be considered the gold standard in their pathological characterization.

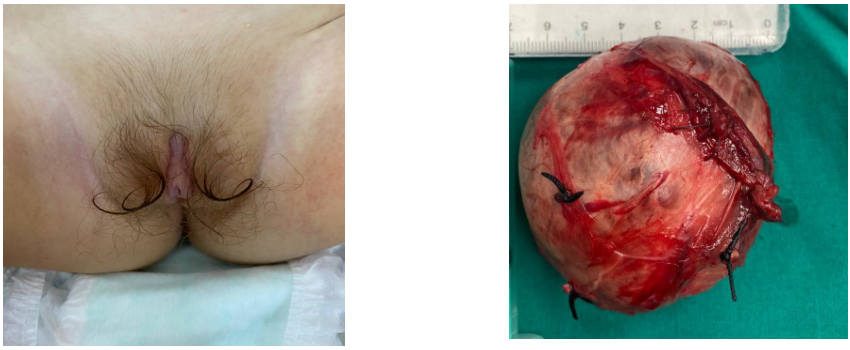

Figure 4. 2-year 2 months-old girl with a left androgen secreting adrenal adenoma

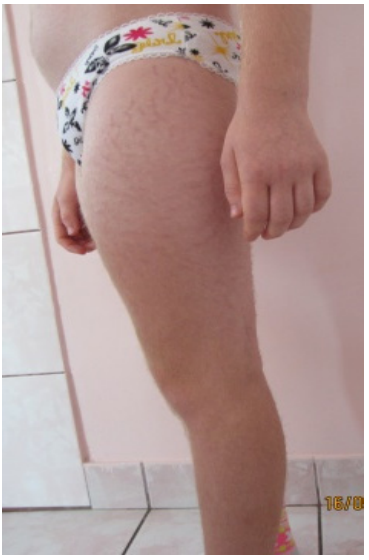

Figure 5. 11-year-old girl with right adrenal reticulata zone hyperplasia

The treatment of these tumors is surgical. After complete removal of the tumor, the signs of puberty regress rapidly [16,17]

2.2. Feminizing adrenal tumors they are very rare and can be benign or carcinomas. Estrogen-secreting adrenocortical tumors in boys induce precocious heterosexual puberty (with the appearance of gynecomastia); growth is normal; signs of virilization such as acne, change in tone of voice and rapid bone maturation may also be associated. The testicles are developed according to chronological age. In girls, these tumors induce isosexual puberty. The ovaries are normal in appearance and have no follicles. [18] Serum estrogen and testosterone levels are elevated.

The dexamethasone suppression test is negative.

Imaging tests (ultrasound, computed tomography) that can reveal the tumor are needed to clarify the diagnosis.

The treatment is surgical. Symptoms regress rapidly after tumor removal.

3. Testicular tumors

Leydig cell tumor: is a rare cause of gonadotropin-independent precocious puberty; it is usually unilateral and benign.[19] The clinical picture is that of isosexual pseudo-puberty that begins between 5-7 years, sometimes gynecomastia can be observed; the unaffected testicle is of normal size.

Testicular ultrasound may reveal the tumor.[20]

Histological examination of testicular biopsy reveals the presence of Reinke crystalloids. Hormone workup reveals low levels of gonadotropins, but testosterone has a high value.

The treatment is surgical and consists of removing the affected testicle.

4. Ovarian tumors

4.1. Virilizing ovarian tumors they are very rare. They can be gonadoblastomas, lipoid cell tumors or tumors of the sexual cord stroma cells.

4.2. Juvenile granulosa cell tumor of the ovary it is a tumor found on the border between benign and malignant, in children. In 2009, Shah et al first reported the existence of a somatic mutation in the FOXL2 gene (C402G; Cys134Trp) in patients with an adult type of ovarian granulosa cell tumor.[21] Curiously, a decrease and/or loss of gene expression of FOXL2 has been reported in advanced/aggressive juvenile granular cell tumors. [22] The FOXL2 gene in the female gonad is a key factor for the correct differentiation of granular cells during folliculogenesis, and its expression persists in the ovary after birth. [23]

Clinically, the signs of isosexual puberty appear at an early age (thelarche, menarche, pubarche), associated with

abdominal pain syndrome; an abdominal mass can be palpated (*Figure 6*.)

Hormonal workup reveals elevated estrogen values and prepubertal values for the gonadotropines.

From a histological point of view, there is a nodular architecture, follicle formation, microcysts and necrosis areas.[23]

The treatment is surgical. After the tumor is removed, the signs of puberty regress rapidly.

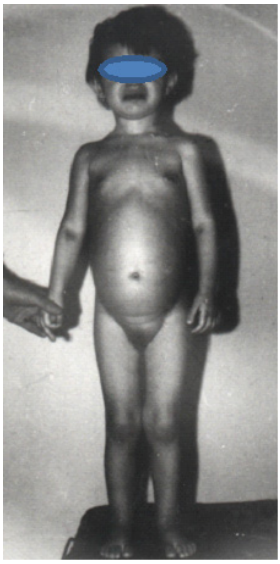

Figure 6. 2-year-old patient with juvenile granulosa cell tumor

Gonadoblastoma

Gonadoblastoma occurs in the dysgenetic gonad in subjects with a female phenotype who have a Y chromosome (46XY, 45, X / 46, XY) in the karyogram or Y chromosome sequences expressing specific testis Y-encoded protein 1 (TSPY1), but it can very rarely also be observed in people without sexual development abnormalities (normal karyotype). The tumor can be uni or bilateral.[23] If the dysgenetic gonad contains delayed-maturing germ cells and has the TSPY1 gene,

the cells may undergo a transformation into classical gonadoblastoma.[25]

The clinical picture includes virilization (clitoral hyperplasia and the appearance of hirsutism), accelerated bone growth and maturation, acne. Hormonal workup reveals an increase in testosterone and androstenedione. Imaging examinations allow the diagnosis to be made. Classical gonadoblastoma can be interpreted as a non-invasive or in situ neoplasm that is the precursor of germinoma, a much more aggressive form of neoplasm in some cases. [26]

The treatment is surgical. After the tumor is removed, the signs of puberty regress rapidly.

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HYPOGLYCEMIA & GLUCAGON TREATMENT OF TYPE 1 DIABETES MELLITUS

Stuart J. Brink

Introduction

As Professor Dr. Mark Sperling has written, the dark side of treatment regimens for type 1 diabetes, especially with intensified management programs and tighter glycemic targets, is that they also increase the risk of hypoglycemia.[1] Hypoglycemia is the most frequent morbid event in type 1 diabetes.[2]

Hypoglycemia is often a limiting factor in the glycemic management of diabetes as we strive toward improved overall glycemia to safely reduce time when hyperglycemic and increase time in (target) range (TIR).[3]

Hypoglycemia occurs in all patients with Type 1 diabetes mellitus, partly because of the imprecise manner in which insulin deficiency is corrected, and partly because of a series of potential estimations based upon the interaction of food, activity and insulin.

In children and adolescents, as in adults, it is often bothersome, sometimes embarrassing, always inconvenient and potentially extremely dangerous and life-threatening. Healthy respect for hypoglycemia is always warranted. Attention to frequency and severity of hypoglycemia is critically important for the overall well-being of youngsters with diabetes and how their family as well those at school, at after school and summer times perceive the PWD.

Hypoglycemia definitions in textbooks, manuals, research projects and clinical as well as research papers remain extremely varied related to type 1 diabetes treatment with differing definitions, and also different ways of counting how often and how severe such episodes really are, and under what circumstances they occur.

The International Society for Pediatric and Adolescent Diabetes (ISPAD), the American Diabetes Association (ADA), the European Association for the Study of Diabetes (EASD), Pediatric Endocrine Society (PES), European Society for Pediatric Endocrinology (ESPE) among other academic societies have had numerous definitions of hypoglycemia (approximately ≤ 70 mg/dl or ≤3.8 mmol/l) and different ways to determine how severe an episode of hypoglycemia has occurred.

One of the big benefits of the hugely successful Diabetes Control and Complications Trial (DCCT)[4] was the enormous collaboration that took place between 29 centers in the USA and Canada. Investigators from each of those centers spent almost a full year writing the treatment protocol and debating about definitions to which all these investigators could agree even though strict scientific definitions did not actually exist. This would become the basis for the standard treatment provided to the control group and the multidose or insulin pump treatment in the investigational group while maintaining the needed randomization.

The ultimate goal of the DCCT was trying to establish some of the causes of the short and long term complications associated with type 1 diabetes in adolescents as well as young and middle aged adults. Because such definitions existed but were so varied and because there was no single definition for a lot of these problems, the consensus that was forged and the process really was a triumph of the medical establishment.

One of the key ingredients of the DCCT debates was the decision to include three agreed-upon definitions of hypoglycemia <70 mg/dl: mild, moderate and severe

hypoglycemia according to symptoms and especially neuroglycopenia findings and need for assistance by others.

Mild hypoglycemia involves *self-recognized* symptoms:[5] heart pounding, fast pulse, shaky, sweaty, not speaking or responding clearly, tingling of extremities, headache, hunger, mild confusion are considered to be neurogenic symptoms and blurry vision, incoordination, lightheaded or dizzy, nausea, sleepiness, trouble concentrating, slurred speech, slowed thinking, lightheaded or dizziness but able to tell/agree something is amiss and able to self-treat with rapid-acting sugary solution like juice, soda or non-fat (not chocolate) sugar-containing hard candies or table sugar or sugar/glucose tablets.

Moderate hypoglycemia still with *self-recognition but needs treatment assistance* of family or peers, hospital staff (if inpatient) or even a passing stranger or police officer and usually can be treated with oral sugar, tablets or solution, able to follow specific instructions with prompting. The same symptoms as listed for mild hypoglycemia although they may be more or less intense. No loss of consciousness or seizures.

Severe hypoglycemia *without self-recognition and unable to self-treat so needs assistance*, same overall list of possible symptoms with more intensity or greater duration, shaky, sweaty, worse confusion, may have convulsions, may lose consciousness. Any loss of consciousness or any seizures automatically produce a severe classification of hypoglycemia. With such increased severity, may stop breathing if prolonged and severe enough, risk of death or neurologic damage much higher → oral treatment risky since may aspirate → **glucagon** (intranasal, SQ or IV) or IV **glucose** immediately required if available.

The exact level of blood glucose is often not correlated with blood glucose levels either in a hospital setting, home monitoring results or with sensor values. This may reflect the difference between peripheral glycemia and central nervous system glucose levels. It also may reflect differences in one's perceptions and the vagaries of when different symptoms occur physiologically, counterregulatory responsivity and

exact cause of the hypoglycemic event (peak of analog or regular insulin vs slower-acting insulins, exercise-induced, complicated by concomitant alcohol etc.) Altered behavior is commonly recognized by others but difficult to self-determine. Without education, practice and attention to the possibility that hypoglycemia can alter one's sense of well-being or functioning, hypoglycemia is often not detected in its earliest stages. Especially in the infant or young child, hypoglycemia (with presumed neuroglycopenia occurring) may take the form of a temper tantrum, misbehavior, aggressive play behavior or naughtiness, drowsiness, apathy or inattentiveness. Of course these can occur at any age and are extremely variable – and may occur completely unrelated to hypoglycemia!

Extreme vigilance is needed most in these youngest patients or any patient with mental compromise (ie. Down Syndrome) and obviously for those PWD who also have hypoglycemia unawareness. A working definition or hypoglycemia unawareness might simply be inability to always detected documented hypoglycemia with a presumed lack of symptoms, inability of the brain when neuroglycopenia occurs to recognize such symptoms or physiologic inability of the body to produce such symptoms. Parents and baby-sitters as well as teachers, coaches, peers and family members should all be taught how to recognize hypoglycemia even if the patient him/herself does not acknowledge the possibility. This may allow checking capillary blood glucose levels or sensor readings and offering oral fact-acting carbohydrate rather than waiting for self-treatment or ultimate self-recognition after symptoms worsen or become dangerous.

Causes of hypoglycemia are myriad and frequently, but not always, there was an obvious (human) error such as taking the wrong amount of insulin, mixing up the type of insulin injected, inappropriate food or insulin compensation for added activity, alcohol or other drugs taken, sleeping later than usual or delaying food or snacks. The first trimester of pregnancy also is a time when hypoglycemia can be severe and recurrent since the developing fetus get "priority" for use of maternal energy resources.[6] Health care providers need to

be aware of this when seeing pregnant patients, especially adolescents who don't necessarily always plan getting pregnant and how this may impact their diabetes needs. New onset of more severe hypoglycemia in adolescent or young adult females with PWD should always raise the possibility of pregnancy as the explanation when reviewed by health care professionals dealing with such populations.

Those PWD with more episodes of hypoglycemia are also prone to have further episodes and past history of more severe types of hypoglycemia also maintains higher risk for the PWD with recurrent issues. Nocturnal hypoglycemia is especially worrisome since there is the potential for more missed warning signals when asleep. Lack of routine bedtime monitoring is especially worrisome as a predictor of nocturnal hypoglycemia and not recognizing such patterns even with insulin pumps and continuous sensor monitoring providing more and more data for analysis. Of course if the data available from self-blood glucose monitoring and/or continuous monitoring systems is not being reviewed regularly at home by the PWD, if age appropriate and/or family members, then the potential for recognition of repetitive episodes of hypoglycemia, as well as repetitive patterns of hyperglycemia, can't be prevented and education efforts should focus on this too.

Further modifications of hypoglycemia classification were proposed by a consensus committee of the American Diabetes Association, American Association of Clinical Endocrinologists, American Association of Diabetes Educators, the Endocrine Society, Helmsley Charitable Trust, Pediatric Endocrine Society and the T1D Exchange [6] with many review articles over the years as referenced therein. Absolute blood glucose levels cannot be used to describe the severity of hypoglycemia because glycemic thresholds for the onset of symptoms differ greatly across individual PWDs and episodes depending on various mediating variables. Hypoglycemia can cause acute harm to the PWD or others especially if it causes falls, motor vehicle accidents or other types of injury. In addition, there are multiple research

studies suggesting increased barriers to "tight" glucose control, hypophobia (fear of hypoglycemia) sufficient to withhold full needed insulin doses and/or overeating as compensation for such hypophobia (see below). Risks of moderate or severe hypoglycemia are higher in very young children as well as the elderly taking insulin because of reduced ability to recognize hypoglycemic symptoms and effectively communicate their needs.

Concomitant alcohol[7] and/or drug use[4,8] as well as beta blockers further interfere with hypoglycemic symptom recognition and contribute to episode severity. Marijuana and other drugs which affect awareness and brain function don't directly cause hypoglycemia but contribute by interfering with proper timing of insulin and food, and decision making as well as appropriate responses. Alcohol also does not cause hypoglycemia directly (despite myths to the contrary) but has an inhibiting effect on gluconeogenesis, the ANS response to hypoglycemia, so that if alcohol is present, then self-correction of hypoglycemia is partially or totally blocked in the PWD imbibing as well as interfering with decision and responses similarly to marijuana and other drugs.

American Diabetes Association Standards of Care 2021[9] included a modification of hypoglycemia classification into three "levels" as follows:

- Level 1: glucose <70 mg/dl (3.9 mmol/L) and ≥54 mg/dl (3.0 mmol/L)
- Level 2: glucose <54 mg/dl (3.0 mmol/L)
- Level 3: a severe event characterized by altered mental and/or physical status requiring assistance of another person for treatment of hypoglycemia

This was based on research that suggested 70 mg/dl (3.9 mmol/L) as the threshold for neuroendocrine responses to falling glucose in people without diabetes. So, 70 mg/dl was considered clinically important independent of severity of acute hypoglycemic symptoms.

Level 2 hypoglycemia is the threshold at which neuroglycopenic symptoms begin to occur and requires immediate action to resolve the hypoglycemic event. If a PWD

has level 2 hypoglycemia without adrenergic or neuroglycopenic symptoms, they likely have hypoglycemia unawareness and requires further investigation and review of the medical regimen focusing on prevention of recurrence, analogs if available and affordable instead of older insulin preparations, education review, behavioral intervention and consideration for use of available/affordable diabetes technology such as CGM, sensors, HCL, CSII.

Oral treatment by the patient him/herself as proposed by this author[8] involves approximately 10-20 grams of glucose, fructose or sucrose (not with chocolate or high fiber foods or complex foods with slower absorption of metabolites). LifeSaver ® candies typically contain 2 grams of sugar so treatment would be 5-10 LifeSavers ®. Typical hard candies contain about 5 grams of sugar so 2-4 candies would suffice. Typical marketed glucose tablets are 5 grams each, so 2-4 tablets. A typical teaspoon of table sugar has about 4-5 grams of sucrose, so 2-4 teaspoons or 1-1½ tablespoons would be comparable; about 20 grams of sugar in 6 oz of orange juice.[5,8]

By 15 minutes after such treatment, ideally, capillary blood glucose should be rechecked and with continued hypoglycemia, the treatment should be repeated.[9]

Once SMBG or glucose pattern is trending upwards, the person with diabetes (PWD) should consume a small meal or snack to prevent hypoglycemia recurring.

The same treatment would also apply for both mild and moderate hypoglycemia as defined above per the DCCT with the only exception that moderate hypoglycemic episodes, by definition, are more severe since they require assistance of someone beside the PWD themselves. Exact levels of blood glucose by meter or sensor are very variable at all three levels although there is an obvious tendency for lower BG values to be associated with more moderate or severe hypoglycemic episodes.

Treatment of severe hypoglycemia[8-9] involves emergent intravenous glucose, if available, intravenous, intramuscular or subcutaneous glucagon (see below) as well as intranasal

glucagon (see below) with common expectation of some nausea and/or vomiting, headache and sometimes exacerbation of prior symptoms as recovery occurs post hypoglycemia and post glucagon. Once awake and responsive, so there is no apparent danger of aspiration, oral feeing with simple fast-acting carbohydrates plus some slower acting carbohydrates like crackers or bread plus available protein and fat (such as peanut butter, cheese, slice of meat) if able to tolerate. As with all episodes of symptomatic or asymptomatic hypoglycemia, ideally blood glucose levels should be rechecked afterwards to make sure that no further treatment is needed.

Asymptomatic hypoglycemia also is well documented especially with more extensive capillary home (or hospital) blood glucose monitoring and even more so with more recently available sensors providing continuous glucose monitoring. Asymptomatic hypoglycemia refers to the PWD who does not report any symptoms; however, any observant family member, friend or health care provider, camp staff of even passersby, may notice some of these symptoms and signs of hypoglycemia even if not self-recognized or even acknowledged by the PWD. Exact levels of blood glucose don't exist to attribute asymptomatic hypoglycemia, like with the mild, moderate or severe episodes as described previously, although with CGM [10,11,12,13,14] providing more retrospective analysis of such levels, some further information that is useful for prevention discussions often arises.

Negative consequences [5,8] of severe as well as moderate or mild hypoglycemia include:

- impaired cognition with daily activities, exam taking, work/school tasks, sports
- embarrassment out in the world
- timing of food, sleeping late, partying with peers
- emotional trauma, anxiety, bullying
- family conflict, overparenting, spouse worries
- financial costs of emergency treatment, ambulance, emergency room costs, glucagon

- accidents especially when driving as a teenager or adult, sports injuries, falls
- convulsions and death associated with known severe hypoglycemia as well as dead-in-bed syndrome

Education about hypoglycemia, likely causes of hypoglycemia, ways to prevent or minimize severity or frequency of hypoglycemia and treatment options all are critically important for the PWD and their family. Knowledge of symptoms and the common occurrence of missed symptom recognition either because symptoms themselves have not occurred or merely not been able to be recognized should be taught. Everyone should be taught and everyone should have this reviewed at least annually. Our own practice at NEDEC, living in a part of the world (Boston) where there were very cold winters with generally less activity than when the sun returned with warmer weather in Spring and Summer, we routinely used the this as a reminder for the annual Spring hypoglycemia review discussions with our patients. Parents and older siblings as well as all caregivers (babysitters, grandparents, other relatives) need to be included in this process and the same is true for the school situation (teachers, teaching assistants, lunchroom attendants, coaches, band and orchestra leaders, school bus drivers, fellow students) [15] and when the PWD participates in diabetes camping programs. [16] Encouraging and discussing age-appropriate information sharing by the PWD themselves and/or parents' involvement in such educational efforts also needs ongoing discussions and support from health care providers. When the PWD is old enough to be working either part-time, summer-time or full-time, then discussions should also include why and how to pass along information to co-workers and bosses about living with diabetes, emergency situations and how assistance may be needed under which circumstances, contacting 911 etc. All this is a routine part of our Spring followup visits [8] with the PWD and their family and is documented very specifically as having been discussed in the medical record as part of quality care provision. This is also the time to review glucagon prescriptions, Medic-Alert

identification necklaces or bracelets and any other related issues that arise.

In adolescents learning to drive and in older PWD, driving [17,18,19,20,21,22] safety should also be routinely discussed vis-à-vis hypoglycemia awareness and preventive aspects. Specifically, monitoring should be done to know actual blood glucose readings prior to beginning to drive. With CGMS and sensors this is quite easy, of course, but it doesn't take very much time to do a quick capillary blood glucose finger test either if the importance is understood and the PWD is compliant with making sure that this is a safety priority.

At every followup visit[5,8] there should also be specific questions about severity and frequency as well as self-recognition and treatment of hypoglycemia to help maintain awareness and the importance of hypoglycemia in analysis of treatment systems being provided. This needs specific and mandatory documentation in the medical records whether handwritten or computer generated to ensure better quality control. The importance of self-blood glucose monitoring whether by capillary sticks and meters or by the newest continuous glucose sensors also needs discussions vis-à-vis hypoglycemia recognition and especially the importance of home analysis of such data - not just at ambulatory followup visits - for identifying potential patterns which may be amenable to prevention efforts.

Benefits of artificial intelligence (AI, see below) with automatic low glucose suspend (LGS) features and preventive computer programs now in use show great promise in achieving more time in range and less hyperglycemia while at the same time focusing on decreasing nocturnal, overall frequency and severity of hypoglycemia, increasing safety and decreasing barriers associated with hypoglycemia and/or hypophobia.

Education about hypoglycemia should emphasize these common reasons for hypoglycemia: late meals, missed snacks, insulin dosing mistakes, association with activity bursts especially when activity is intermittent or unpredictable and known peak effects of different insulin preparations.

Discussions about ongoing use of older NPH and non-analog regular insulins and how the newer analogs offer more predictable and uniformity from day-to-day are important. Also to be reviewed are how multidose insulin regimens and insulin pumps provide even more physiologic availability of insulin so that, at least theoretically, there should be less hyperglycemic bursts and fewer hypoglycemia events occurring. Options for moving from older insulin preparations to analogs as well as hybrid closed loop pumps and the artificial intelligence progress that has been made the past decade with such increased data sources all suggest that major barriers vis-à-vis hypoglycemia and glycemic control are indeed being decreased quite substantially.[23] While there is more hypoglycemia occurring as near normal glycemia is approached, there is still significant amount of hypoglycemia with out-of-control diabetes because of greater glycemic variability on a day-to-day basis. Motivational interviewing and focus on preventive education,[24] ways to self-identify potential patterns with home blood glucose monitoring, graphing with paper and pencil or with meters downloading to computer programs that highlight such issues and the more sophisticated data analysis programs now available with pump and sensor usage increasing all help to identify and prevent as well as minimize hypoglycemia for the PWD.

It should also be emphasized that there are some other conditions that may co-exist in the PWD that also increase susceptibility and severity of hypoglycemia. If the PWD has concomitant celiac disease, which may be as much 5-8% of a Type 1 diabetes mellitus cohort, [25] there may also be some increased problems with hypoglycemia because of food absorption issues. And this can occur in the asymptomatic or mildly symptomatic but yet undiagnosed PWD who also has undiagnosed celiac disease. Thyroid autoimmune disorders [ibid] are also increased in the PWD but don't directly impact hypoglycemia incidence or symptoms. However, just like there is more hypothyroidism and hyperthyroidism associated with autoimmune related type 1 diabetes mellitus and more celiac disease as well, there also is more of the rarer condition,

Addison's disease[26], where there is autoimmune destruction of the adrenal cortex and positive adrenal autoantibodies[27] so that there are mild, moderate or severe versions of Addison's with glucocorticoid insufficiency/deficiency that may have a large impact on causing or worsening hypoglycemia. Especially when Addison's disease exists but is not yet diagnosed [28] or when it occurs in a more subtle format, hypoglycemia risks increase with or without Type 1 diabetes mellitus. When celiac disease as well as Addison's disease are appropriately treated, then the associated increased hypoglycemia markedly decreases under such circumstances. Even more rare would be Type 1 diabetes mellitus in a patient with hypothalamic or hypopituitary insufficiency especially affecting adrenal[29] and growth hormone pathways and responses.

Work done by the hypoglycemia research group at the University of Virginia has been extensive and detailed over the past several decades and includes a hypoglycemia questionnaire/survey which they developed to help assess patient risk factors and hypoglycemia beliefs.[30] These are included as helpful queries to consider:

Followup studies from the DCCT/EDIC have continued and some specific attention has been paid to the increase in hypoglycemia originally reported in the intensively treated subgroup compared to the control group who continued on human insulin but without specific blood glucose targets. Both groups received educational support about minimizing hypoglycemia although the experimental group had more frequent contact and educational opportunities with study health care providers than the control group as part of the study protocol and efforts to minimize the excess hypoglycemia, when it became apparent, were also provided as part of the DCCT. Originally the DCCT [3] reported about a 3x increase in hypoglycemia associated with intensified treatment, either multidose insulin regimens or insulin pump regimens but with targeted blood glucose levels compared to the control group on standard pre-analog insulin once or

twice-a-day regimen. This was true for severe as well as mild and moderate episodes of hypoglycemia.

To what extent do you:	Not at all		Somewhat		Great Deal
1. Always carry some type of food or drink with sugar?	1	2	3	4	5
2. Skip meals?	1	2	3	4	5
3. Skip snacks?	1	2	3	4	5
4. Worry about hypoglycemia?	1	2	3	4	5
5. Try to keep BG <100 mg/dl/~5.5 mmol/L?	1	2	3	4	5
6. Delay eating when trying to finish a task?	1	2	3	4	5
7. Think low BG is a sign of good control?	1	2	3	4	5
8. Eat extra food when gong to be more active?	1	2	3	4	5
9. Recognize low BG symptoms?	1	2	3	4	5
10. Eat as little as possible to avoid gaining weight?	1	2	3	4	5
11. Increase your insulin when your BG is too high?	1	2	3	4	5
12. Wait until you feel strong symptoms to treat low BG?	1	2	3	4	5
13. Only treat very low BG levels (between 40-50 mg/dl/~ 2-3 mmol/L?	1	2	3	4	5
14. Believe you can function fine with a BG <50 mg/dl/~<3 mmol/L?	1	2	3	4	5

Other investigators have confirmed the increase in hypoglycemia following successfully adapted intensified diabetes treatment regimens[31,32,33] although more recent studies using analogs, more adaptable pumps and sensors have shown that excess hypoglycemia and more severe types of hypoglycemia can be decreased and minimized (compared to what was available during the actual 9 years of DCCT protocol.) [34,35] EDIC has continued to follow this same cohort

but with the control group offered the same options for intensified treatment the end of the original 9 year study period. At the 30 year followup point, DCCT/EDIC[36] analysis of risk of severe hypoglycemia was carried out and one half of the DCCT/EDIC cohort reported episodes of severe hypoglycemia. During EDIC rates of severe hypoglycemia fell in the former DCCT intensive treatment group but rose in the former conventional treatment group, resulting in similar rates (36.6 vs 40.8 episodes per 100 patient-years, respectively.

During the EDIC followup period all diabetes care was provided outside of DCCT facilities in the community and participants have adopted many advances in diabetes technology including use of raid and long acting insulin analogs instead of animal or human insulins, improved insulin pumps, meters, sensors and advances in artificial intelligence. During EDIC, A1c levels have been ~8.0% in both groups so it is believed that the key differences are the approximately 9 years of significantly improved A1c levels in the experimental vs the control group even though subsequent levels for A1c were similar in the EDIC extension period. Interestingly, the control group only opted for insulin pump treatment in 1.6% of their cohort while the original experimental group opted for pump use in 41%.

Preceding episode of severe hypoglycemia was the most powerful predictor of subsequent hypoglycemic episodes. The adolescent subgroup of the DCCT also persisted with increased risk of severe hypoglycemia while insulin pump use was associated with a lower risk. Severe hypoglycemia rates increased with lower hemoglobin A1c levels similarly among participants in both treatment groups. Lower overall hypoglycemia in EDIC is consistent with the results of randomized clinical trials of analog insulins and improved methods of continuous glucose monitoring (CGM)[37] that have demonstrated lower A1c could be obtained without increasing the risk of severe hypoglycemia compared with control subjects and similarly with intensified insulin pump therapy[38,39] over more conventional insulin injections even

when multiple insulin injections were utilized. Sensor augmented insulin pumps [40] improved the situation further as hybrid closed loop (HCL) systems were developed, adopted and further improved over the past decade.[8,13]

Hypophobia (fear of hypoglycemia) is a rather common phenomenon although not often brought up for discussion by the PWD unless volunteered as a topic of discussion by health care providers.[41,42,43,44] In both children and adolescents, fear of hypoglycemia [29,45] is related to a history of severe hypoglycemia and there is some relationship with generalized anxiety. In the child compared to the adolescent, however, there are some differences with respect to their parents' concerns. Glycemic control remains a delicate balancing act because of the more immediate concerns about hypoglycemia and its implications for overall glycemic control.[46] The parents of a child with diabetes have specific concerns about whether or not their child can learn and utilize behaviors to prevent low sugar episodes (ie. finish meals, not skip meals or snacks, check BG levels and respond) and this is also true for parents of adolescents as their teens move away from direct parental oversight. Parents with anxiety issues on their own also tend to have more fears of hypoglycemia episodes when it comes to children with type 1 diabetes but parents of adolescents also worry about their teenagers not carrying rapid acting glucose treatment with them more than is reported in parents of preteens. Some studies have also shown in adults that fear of hypoglycemia was significantly associated with more negative diabetes-related quality of life and psychological well-being.[47] Despite such concerns, we know that only 8% of future severe hypoglycemic episodes are predicted from known variables in the DCCT reports.[4,8,29] We also know that only 18% of future severe hypoglycemic episodes are predicted from previous history of hypoglycemia, hypoglycemic unawareness or known autonomic nervous system abnormalities. For the PWD, hypophobia can lead to unconsciously reducing insulin doses, omitting insulin doses especially at bedtime or when with peers away from the family as well as purposeful overeating with an unstated or denied goal of keeping actual blood

glucose levels so high that low sugar levels don't occur. Even though we know that most hypoglycemic episodes, when evaluated closely retrospectively, would be predicted to occur because of obvious mistakes as already mentioned: too much insulin for the situation, not adapting insulin and/or food for activity changes, missed snacks or meals, under-eating without decreasing insulin dosing, sleeping late etc. This means that with open discussions it may be possible to avoid the extreme thoughts that lead to (surreptitious) hypophobia and with motivational interviewing and honest, nonjudgmental discussions, allow learning preventive steps to minimize hypoglycemia without losing control of the diabetes situation, ie. more time in range and less time hyperglycemic without excessive or severe hypoglycemia as well. Hypophobia as a concept should be discussed in educational settings on a regular basis, brought up when there is no obvious explanation for high A1c persistence, persistent absent glucose monitoring, missed or too low insulin doses, higher time in range and hyperglycemia than ideal. It also should be discussed when there is a PWD with recurring hypoglycemia especially the moderate and severe episodes which require outside assistance.

Hypoglycemia unawareness[8,48,49] is likely a more common problem than assumed, not only in the very youngest infants and toddlers as well as anyone with dementia or genetic/neurologic compromise of CNS function, but in all patients with type 1 diabetes mellitus.[50,51]

The counterregulatory hormone response to hypoglycemia and the autonomic symptoms tend to decrease after several years of diabetes so that neuroglycopenic symptoms become the predominant first manifestation for many.[8,52] Beta blockers as well as alcohol or any drugs that impair brain function/decision making also have similar properties. Recurring hypoglycemia[53], mild, moderate or severe, also contribute to inability to produce appropriate basic counterregulatory and autonomic responses so that reducing hypoglycemic events of any kind, in anyone having problems with recurring severe hypoglycemia or hypoglycemia

unawareness, can recover and return to a more basic response standard if efforts are successful in avoiding hypoglycemic events of any kind; this may take several weeks to accomplish. The more blood glucose testing (or CGMS/sensor use) that is done, the more asymptomatic or minimal symptomatic hypoglycemia can be identified as long as the actual data obtained is reviewed by the PWD and/or family members or health care providers. Doing so only at office followup visits, however, isn't likely to be sufficient except to point out how often it is occurring and now acknowledged. Many PWD believe that they can "know" when they are hypoglycemic but with increased glucose testing this is obviously not a correct assessment.[54] Why some can be better at self-identification and others have convulsions or lose consciousness is not totally understood scientifically. Some have convulsions only in the middle-of-the-night whole others have aggressive or other behavioral changes during the daytime hours. Dangerous outcomes for such patients occur when driving, participating in very active or prolonged sports, sleeping late or also using drugs/medications which have an impact on hypoglycemia symptoms.

Correct initial and repeat education of about hypoglycemia [55] is critically important for decreasing severity and frequency of hypoglycemia especially if there is also unawareness. Queries about concerns from the perspective of the PWD but also parents[56] and grandparents, their family members, peers at school, after-school and sporting events all should be part of the educational discussions. Our own approach developed at NEDEC[8,24,41-44] has been fully documented with hypoglycemia prevention and education every Spring for everyone, specific queries about hypoglycemia severity and frequency as well as downloaded meter or pump/sensor data at each followup encounter with a focus on identifying, decreasing and preventing future hypoglycemia episodes and their severity. At the same time reviewing the importance of MedicAlert identification in wallets, necklaces and/or bracelets has been emphasized at each encounter and specifically at the annual Spring sessions and making sure

that emergency glucagon was up-to-date and available. With the introduction of nasal glucagon kits (see below), less scary and easier to use plus not needing refrigeration storage, this has become an easier chore for healthcare providers as well as patients and their family in recent years in the same way that prefilled glucagon syringes and glucagon pens have added to the treatment options to be considered.

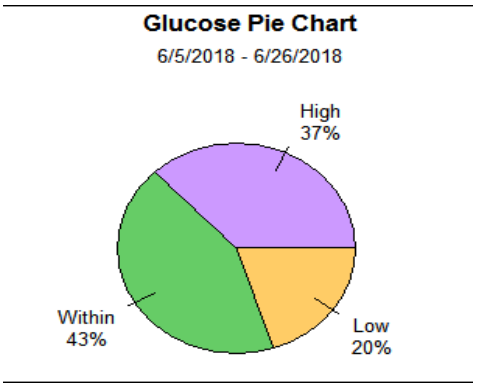
The research team at the University of Virginia has had great success, as have others, producing a system that they have called BGAT, (Blood Glucose Awareness Training) [57] and successfully used in the United States as well as several European centers. BGAT is a psychoeducational programmatic intervention designed to improve the accuracy of the PWD's detection and interpretation of hypoglycemic symptoms and other cues. BGAT has been shown to improve accuracy of recognition of glucose levels as well as specifically to improve detection of both hypoglycemia and hyperglycemia.[58,59] The PWD who is the least accurate in detecting current glucose levels, such as those with documented hypoglycemia unawareness, seems to benefit the most in such studies.[60] BGAT also was specifically studied in those receiving intensified T1DM management programs, ie. pumps and targeted MDI regimens; the same benefits accrued and there was demonstrable improvement in hormonal counterregulation responses as well.[61] Using driving accidents as an outcome marker in these studies showed only a 5% collision occurrence in those who participated in the BGAT program compared to 42% of those who had not.[62] Not only hypoglycemia detection was affected by BGAT as well as whether to drive a motor vehicle, but also detection of hyperglycemia, and how to respond improved with decreased DKA episodes reported by participants. Improved psychological functioning and worry was also reduced especially quality of life and worry subscales as knowledge about diabetes improved from baseline. An on-line internet version of BGAT was also developed and found to be beneficial in a self-directed and cost-effective manner.[63]

Education and awareness are the best ways to help prevent most episodes of hypoglycemia and, more importantly to eliminate the most severe types of such episodes.[64,65] What we still need are reliable predictors of future and imminent hypoglycemia coupled to improved ways of managing glycemia. These are all moving along and improving in recent years, at an amazingly quick pace. Since insulin was first used successfully in PWD in Toronto by Banting and Best one hundred years ago[66], we have come a long way towards these goals by moving from animal-source beef and pork insulin products to human-bioengineered insulin, and then analogs of such bioengineered insulin that work more physiologically compared to endogenous insulin. LisPro insulin (Humalog ®) [67,68] as well as aspart insulin (Novolog ® or Novorapid ®) [69,70] have mostly replaced older fast-acting animal and human insulin with the benefits of bringing down postprandial hyperglycemia more effectively and, at the same time, reducing the tail effect so decreasing hypoglycemia several hours after administration.[71,72] Even newer rapid acting insulin (Viajet ®) has also been produced with a more rapid insulin absorption compared to human regular insulin and insulin lispro.

Replacing ultralente, lente and NPH "basal" insulins also involved bioengineered human insulin modified to be flatter, work more consistently from day-to-day, and help avoid middle-of-the night insulin peaks. This is not always possible in parts of the world where donated NPH is the only affordable option to consider and the newer, beneficial bioengineered basal insulins like glargine (Lantus ®)[73,74] and detemir (Levemir ®)[75,76] or degludec (Ideg ®)[77,78 ,79] are simply not available at all or just too expensive in low resource countries (LRC) as well as middle resource countries (MRC). The costs for such newer analog products remain extremely high in countries like the United States where biopharmaceutical limits on pricing are not rigorous compared to better managed health care systems such as in Canada, Australia, New Zealand and most of Europe. Extra attention to LRC and MRC round the world is needed to safely improve

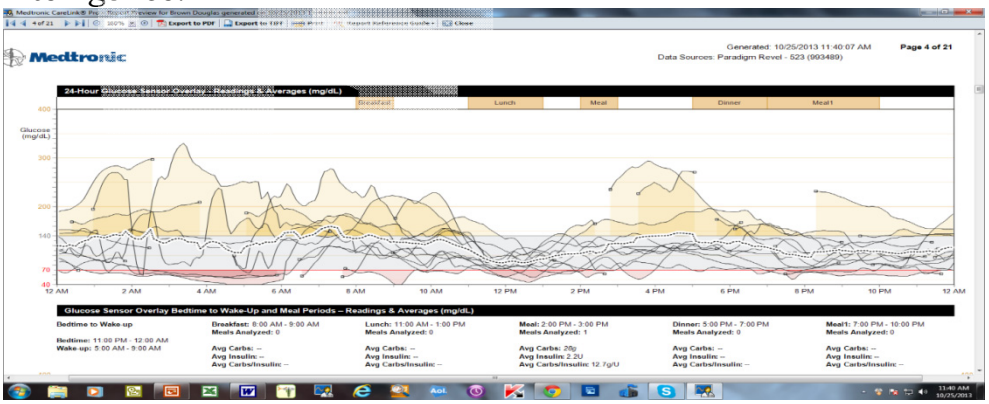
overall glycemia at the same time as minimizing known risks of increased hypoglycemia with older insulin like animal based or human insulins regular and NPH insulins.[80] This has been demonstrated to be done successfully in many parts of the world through the assistance of such programs as LFAC (Life for a Child®) and CDIC (Changing Diabetes in Children®) and with educational tools designed to facilitate and encourage pediatric and adolescent diabetes health care professionals.[81]

Similar progress as noted above include improvements in smaller and more accurate blood glucose meters, ability to download most meters to mobile telephones or home computers as well as office computers at followup visits for computerized rapid analysis of time in range vs percentage of time hyperglycemic as well as percentage of time with hypoglycemia. Many children, teens as well as adults simply learn easier and can recognize patterns in circle graphic displays (see below) as well as other displays in place of logbook style visibility. These tools facilitate self-analysis at home between visits and also facilitate discussions with health care professionals as well as being time-saving. Two examples of such programs are shown: to the left, from a meter with an easy glucose pie chart allowing health care providers easy discussion startups; the second one below from a pump user with enormous amounts of data but again easy for the PWD and their family as well as health care professionals able to focus on the three prolonged overnight hypoglycemic events as well as mid-day hypoglycemia patterns and pre-sleep hypoglycemia also which may be preventable with increased awareness.



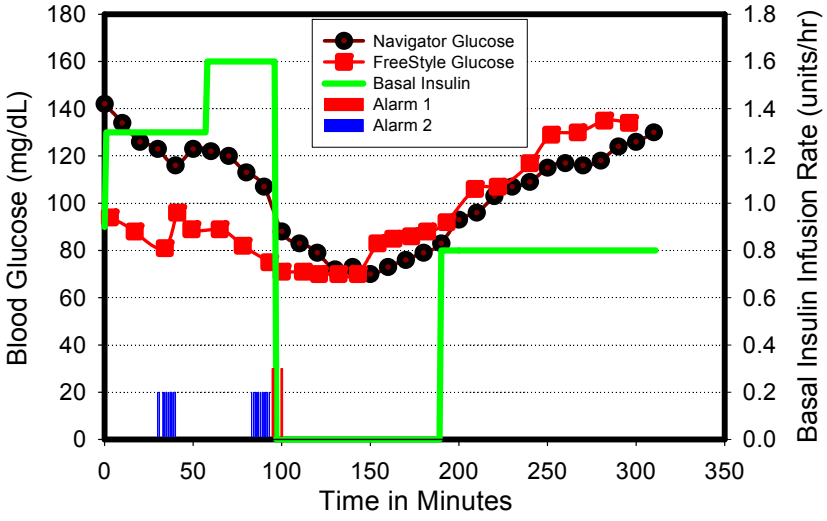
Not only are insulin pumps more available and smaller but they also now have better computer programs with better

alerts and predictive options because of advancing artificial intelligence.



The newest pumps can use the faster and more physiologic insulin analogs and be connected to much improved continuous glucose sensors (CGMS) automatically every 5 minutes.[82,83]

These newest sensors provide much more information but they do so automatically so that if analyzed properly once or twice a week and certainly at followup ambulatory visits



with the health care team, identification of nocturnal

problems (unrecognized hypoglycemia)[84] as well a potential patterns for both hyperglycemia and hypoglycemia in easier fashion.

The latest models have bidirectional connections so that the sensors can allow predictive trend analyses and alerts as well as automatic low glucose suspend (LGS) functions when the BG pattern "predicts" [85] that there will be either worsening hyperglycemia to be expected, therefore increase the insulin being delivered, or, in the case of hypoglycemia predictions, slowing down or temporarily stopping insulin delivery. This can happen safely and reliably in advance of both patterns with AI advances and this can automatically increase time in range, virtually eliminate all nocturnal hypoglycemia episodes and drastically reduce most day-time hypoglycemic episodes while improving and lowering A1c levels progressively. These tools are expensive; there are still too many insurance and government health care benefits that do not automatically cover such prescriptions. Such barriers are decreasing with more and more research studies showing improved glucose metrics with such semi-automatic (hybrid closed loop, HCL) devices as we continue to move toward fully automatic closed loop systems. Medtronic ® was the leader in such semi-automated systems with a Veo ®system and their Guardian ® sensor that connected to its pumps, and then advanced and connected bidirectionally with LGS HCL systems. DexCom ® sensors connected to T-Slim ® as well as Insulet ® pump systems; and all the data collected can be shared on mobile telephone applications as well as home and office computer systems. No DKA risk have been found and pump suspension programs have satisfactorily and significantly reduced severe episodes of hypoglycemia, total episodes of hypoglycemia, and especially, the most dangerous episodes, nocturnal hypoglycemia, were essentially automatically eliminated.

With what has been learned from several decades of research studies about hypoglycemia and hypoglycemia prediction and prevention, about 8% of future severe hypoglycemia episodes are predicted from known variables as reported in the DCCT and DCCT/EDIC studies.[4,36] Current AI

in use with such HCL and predictive suspend features show marked improvements in quality of life as well as specific glucose outcome metrics.

Ongoing hypoglycemia research has also focused on self-behaviors that increase risk of recurring and/or severe hypoglycemia. Skipping meals or snacks, delaying meals or snacks, irregular timing or mis-timing of insulin to food or eating all are obvious behaviors that should be considered with educational efforts to motivate improvement for the PWD. Not always carrying fast acting carbohydrates (glucose tablets, LifeSavers, gummy bears or Swedish fish) and not having glucagon available to treat future hypoglycemia also should be included in this list. Not wearing medic-alert identification also adds risk for the PWD in the midst of hypoglycemia since anyone around to assist would not automatically be aware of the underlying diabetes diagnosis and might easily assume an alcohol or some other kind of substance abuse especially if there is loss of consciousness, rambling speech or odd behavior or convulsions. While emergency medical personnel have been taught to consider hypoglycemic events, this is not always the first thought that occurs when there is no information about the person in distress.

Lack of routine blood glucose monitoring as well as not regularly reviewing BG monitoring data (or sensor data if using CGMS) allows missing some obvious episodes that may reflect a pattern which may be preventable in the future. Any family chaos, particularly substance abuse, anxiety and depression adds to the compliance problems that may appear with any chronic illness, including diabetes. If this is present in the PWD, then there are obvious added burdens in having to deal with more than just the demands of life with diabetes. Most specifically, attention deficit with or without hyperactivity (ADD and ADHD) and all forms of dyslexia and dyscalculia can contribute to compliance issues for the PWD but also to difficulties comprehending the many daily tasks needed to balance insulin, food, activity, life stress etc.

Physical activity, obviously, also plays a part in increasing risks of hypoglycemia but can be managed with

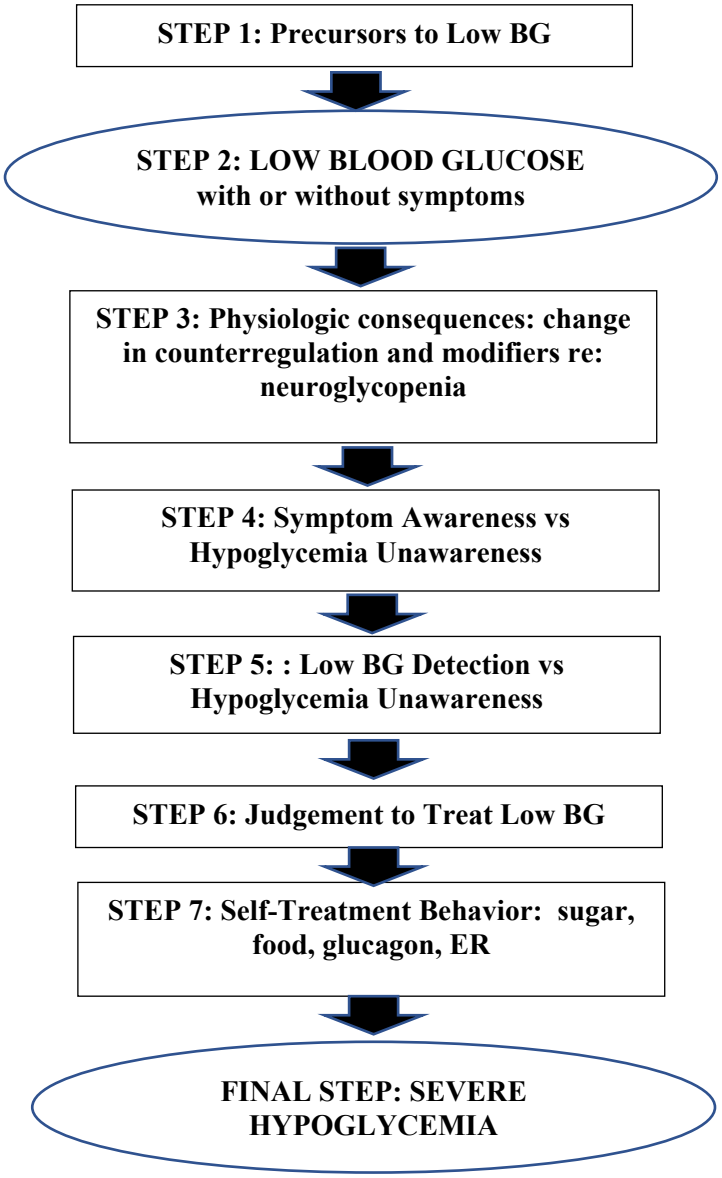
education. But failing to reduce insulin and/or increase food appropriately for activity adds to the complexity of physical activity that occurs randomly and not with appropriate counterbalance of food and insulin or just unscheduled and random activity changes can all produce more frequent and more severe episodes of hypoglycemia in the PWD. Alcohol, marijuana, cocaine, heroin and most other psychoactive drugs when added to diabetes impairs decision making regarding all the usual diabetes self-care tasks but then may increase risks re: driving in addition to making recognition of subtle hypoglycemia symptoms more problematic, more prolonged and even increase severity.

Clarke and his colleagues have proposed a seven-step assessment in order for health care professionals to be able to address various issues related to recurrent hypoglycemia in the PWD.[5]

Step 1 Precursors to low BG include availability and participation in diabetes education including periodic, planned knowledge updates, training manuals such as Type 1 Diabetes Manual,[86] Learning to Live Well with Diabetes,[87] Type 1 Diabetes for Dummies,[88] Understanding Diabetes. A Handbook for People who are Living with Diabetes,[89] American Diabetes Association Wizdom Kit (For Kids Only and For Parents Only versions),[90] Bright Spots and Landmines. The Diabetes Guide I Wish Someone Had Handed Me. 2007, diaTribe Foundation, USA,[91] Think Like a Pancras. A Practical Guide to Managing Diabetes with Insulin.[92] Life for a Child (LFAC) and Changing Diabetes in Children (CDC) as well as Walt Disney and many local, national and international diabetes organizations also have many excellent educational publications that are geared for young children, school-age children, teenagers, young adults and older adults with diabetes to not only help educate but also to help motivate the PWD and his or her family and acquaintances.

There are also excellent on-line support services such as cwd (childrenwithdiabetes) that offer support as well. The LFAC website has a detailed list of such educational tools in multiple languages.

As was mentioned earlier, the older animal-based and human regular and NPH insulins as well as ultralente, PZI, lente and CZI preparations cause more episodes of hyperglycemia as well as more episodes of hypoglycemia because they are not as physiologically absorbed compared to the newer insulin analogs.



Unfortunately, these new analogs are priced significantly higher so that LRC and MRC countries cannot afford them unless they are subsidized or donated. Similarly, costs for blood glucose self-monitoring, meters and batteries fall under the same problematics for affordability and availability. The more monitoring that can be done and sustained, the better is the potential for discovering patterns, asymptomatic situations for both high and low BG results and ways to reduce both extremes of BG levels as well as decrease day-to-day variability. All these are part of the step 1 precursors to low blood glucose construct. Hopefully there will soon be some improvement in affordability and availability in HRC as well as MRC and LRC sites with increasing attention being paid by PWD, their families and health care providers as well as government politicians and general citizenry.

In Step 2, the first appearance of low blood glucose levels, there can be no symptoms so only ongoing monitoring would detect such episodes or there can be a variety of symptoms already discussed. Not everyone has symptoms of hypoglycemia, not everyone has the same symptoms and at what levels such symptoms occur, how long they last and how severe they are is also extremely variable. Any episode of hypoglycemia also doubles the chances of subsequent low BG and decreases the intensity of neurogenic symptoms so that future episodes are more difficult to recognize. More glycemic day-to-day variability also makes identifying hypoglycemia more difficult.

Step 3 reflects neuroglycopenia as a brain/central nervous system response to hypoglycemia physiologically. This has been documented to produce alterations in brain waves on EEG [2,3,93,94] with changes in counterregulation which have been documented in research studies and possible modifiers of maximum or absent response to be considered in the future.[95,96] In general there is a staged response to hypoglycemia at approximately these glycemic thresholds in adult studies: ~80 mg/dl, insulin inhibition (not relevant in PWD without sufficient endogenous insulin), ~68 mg/dl, glucagon responsivity (abnormal in many with PWD), ~ 65

mg/dl, decreased adrenalin and GH responsivity, ~58 mg/dl decreased cortisol/ACTH as well as adrenergic symptoms, ~ 50 mg/dl, neuroglycopenic symptoms, ~30 mg/dl, liver response, ~28 mg/dl, seizures or coma. How quickly are the decreases in BG levels also may be an important factor to consider and only with more CGMS/sensor use have we been able to consider this factor in recent years. Caffeine intake seems to increase hormonal counterregulatory responses while males seem to have a different degree of responsivity than females, recent low BG dampens responsivity, alcohol intake dampens responsivity and overt autonomic neuropathy which can be present subtly or symptomatically with longstanding hyperglycemia all affect counterregulatory responsivity. Neuroglycopenia itself is also best described on a continuum from slowed mental or motor responses which may or may not be self-recognizable, errors, confusion or decreased, and finally loss of consciousness with or without convulsions. The PWD may or may not be aware of these changes but others with him or her may be able to recognize them and help the PWD to respond if taught how to do so.

Step 4 involves symptom awareness or hypoglycemia unawareness and reflects all of the above commentary and research progress over the past three decades with more sophisticated neurologic studies, more sophisticated hormonal and neuroregulatory measurements possible in addition to sensor studies concomitantly. Whether or not an or all of the autonomic/adrenergic (trembling, shaking, sweating, pounding heart, fast pulse, higher BP, body temperature changes, extremity tingling, heavy breathing) or neuroglycopenic (slow thinking, trouble concentrating and paying attention, blurry vision, slurred/absent speech, incoordination, numbness, dizziness, sleepiness/fatigue) symptoms are present, recognized and responded to remains very variable from patient-to-patient and even, sometimes, in the same patient. Hunger, generalized weakness, nausea/vomiting, headaches, anxiety or mood changes and a generalized feeling of “something not right” all are a bit more difficult to classify as neurogenic vs

adrenergic. It's important to stress that not everybody has symptoms when hypoglycemic, symptoms are not always reproducible even with the same actual BG levels and symptoms may worsen with longer duration worsening glycemic control and rising A1c levels. Hypoglycemia awareness, [97] may worsen over time if there is no change in number or severity of hypoglycemic episodes, overall A1c levels and control. If things improve, and especially if there is a real decrease in hypoglycemia, then awareness may return but it may take weeks or months to see this effect.

Step 5 involves low BG detection problems: in the very young or very old where communication or symptom recognition may not be possible, more monitoring and sensors may have a specific benefit even though more costly – but only if there is concomitant awareness and efficacy in home assessment of such results; just doing the BG tests and not responding or wearing sensors without review and response, has no benefit but large costs. Efforts to minimize or avoid beta blockers or other medication which can have a negative effect on hormonal, adrenergic/ANS or brain response consideration to avoid or change such medications would be important for health care providers but this mostly affects adults and the elderly. Similarly, efforts to avoid alcohol, marijuana and other substances becomes ever more important in type 1 diabetes for all the reasons already specified. Family education including grandparents and siblings, peer education through ambulatory group visits as well as hotel or camp stays allow such education to proceed as well and the same for positive programs involving teachers, school nurse and coaches to help become more aware of type 1 diabetes issues and consider urgent or emergency situation responses and preparedness.[55,98,99,100,101,102] School personnel should specifically be trained that the child with diabetes requesting to go to the nurse for a BG test and treatment, should never go alone or else there should be a system in place to call for nursing assistance from the principal or nursing office to come escort the child so that symptoms don't accidentally escalate. This should all be part of the annual

school support form in place which parents should review, as well, annually each start of the school year.[5]

Step 6 Judgement to Treat can also be problematic. Some specific questions to present to the PWD (and family): Am I too slow? Is it suddenly too hard to do? Am I making mistakes? Am I in need of assistance/help right now? How do I get help right now?

All such decisions can be taught and practiced so that, perhaps in an emergency situation, someone may be able to assist or a simple mobile phone emergency call started. All such episodes may be worsened when asleep, by certain medications affecting CNS functioning and by the effects of hypoglycemia per se. The longer the hypoglycemia and the lower the hypoglycemia, the more possibility for impairing judgement. Parent, sibling or other observer's decisions to treat, how severe is the episode and which treatment to consider compared to calling for an emergency medical response all depend on the open and repeat discussions with all those people, education of all those people and involving the PWD and/or family with inviting those folks to be educated and how to assist/respond. This h to be the responsibility of both mothers and fathers. The PWD and his or her significant others may also have some fears of simple sugar substances that may legitimately be used for mild and moderate hypoglycemia vs their typical understanding that "sugar is to be avoided." For young children, the Disney/Lilly series of education booklets is excellent in discussing the role of youngsters and even supervisory adults in situations like parties, sports events, sleepovers and hypoglycemia episodes.

When considering glucagon treatment, which many NEDEC patients call "the spear", concerns are with a relatively large gauge needle attached to a relatively large syringe (compared to a typical insulin syringe) adds an added emotion of "fear" with the older glucagon preparations. The difficulties of correctly mixing and administering glucagon in needle/syringe and vial preparations has been "fixed" by more recent premixed glucagon pens/syringes, smaller or hidden needles and semi-automatic administration of the newest

pens and syringes as well as very effective and safe nasal glucagon. (see below) Expired glucagon preparations, overpriced glucagon preparations and simply omitting to refill the glucagon prescription (because it isn't used on a daily basis) are all issues to help explain how often glucagon is simply not available.[8,41-45]

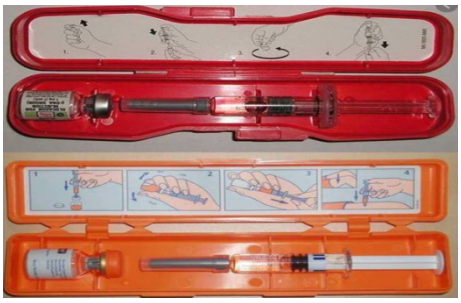
Step 7 Self Treatment behaviors focus on potential compromise as in Step 6 or simply insufficient education and reinforcement vis-à-vis parents, grandparents, siblings, peers, teachers and school nurses, coaches, camp staff. For the PWD it cannot be overemphasized the importance of discussions about hypoglycemia at every ambulatory visit in terms of downloaded BG data to review frequency/severity of hypoglycemia as well as information provided as to how to respond, when to call for assistance etc. Review should include possible sources of liquid, candy or tablet glucose/sucrose vs foods like nuts and chocolate that are loaded with sugar but potentially dangerously slow to be absorbed because of the fat and/or protein effect slowing down such absorption and availability to compensate for the hypoglycemia.[5,8,15,16,24,41-43] A simple pamphlet or slide show as part of the annual Spring education review with ice cream, donuts, meat on sandwiches, peanut butter, pizza, hamburgers and hot dogs, French fries, coffee with milk and sugar or cream, chocolate candies, most candy bars, cakes and pies, bread and bagels and potato or cheese chips are commonly used to treat hypoglycemia when "nothing else is available." While this is better than nothing, it isn't likely to boost the low sugar causing the problems very quickly (or at all for an hour or two) but appropriate options can be taught.

Glucagon has been available manufactured by Eli Lilly, NovoNordisk and Fresenius Kabi in small, portable red or orange plastic kits with powdered glucagon and a diluting solution in vials. A disposable plastic syringe is also included in the glucagon kits with which to withdraw the appropriate amount of diluting solution, inject it into the powder glucagon vial, thoroughly and quickly mix it up without making excessive air bubbles, then withdraw the correct amount of

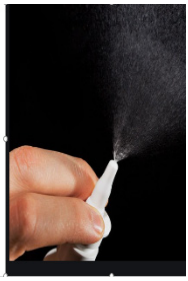
glucagon and inject subcutaneously (or intramuscularly) to provide glucagon for a moderate or severe hypoglycemic episode. Any injection site in any part of the body can be used but most recommendations suggest the buttocks or the anterior thigh. The glucagon is not always available because non-routine use makes it more difficult to remember how to use, especially in the midst of such an emergency, more difficult to remember to request prescription refills and more difficult because of the high costs and high co-payment costs attached to most family health plans. In many of the world, glucagon is simply too expensive unless being donated, and then with the older preparations, the problem of refrigeration and/or lack of reliable electricity is added to the situation.

Glucagon can be used in very young children with a titrated dose.[103] NEDEC emergency glucagon treatment of hypoglycemia, if moderate or severe hypoglycemia occurs or if there is any question of inability to swallow (danger of aspiration), then physicians and nurses can administer intravenous, intramuscular or subcutaneous liquid glucagon. Non-health care professionals can also administer intramuscular or subcutaneous liquid glucagon with the Lilly, NovoNordisk or Fresenius Kabi kits as long as they are available. Typical dose would be 0.5-1 mg glucagon IV, IM or SQ with an overall dose recommendation of ~10-20 ugm/kg body weight. Intravenously some have recommended 0.2 g/kg intravenous glucose @ 4-6 mg/kg/min (very slowly to prevent rapid osmotic shifts) if a large vein is available already or glucagon is not available. Studies comparing intravenous glucagon and dextrose showed both to be effective with glucagon taking just a few minutes longer to raise BG levels when compared.[104,105]

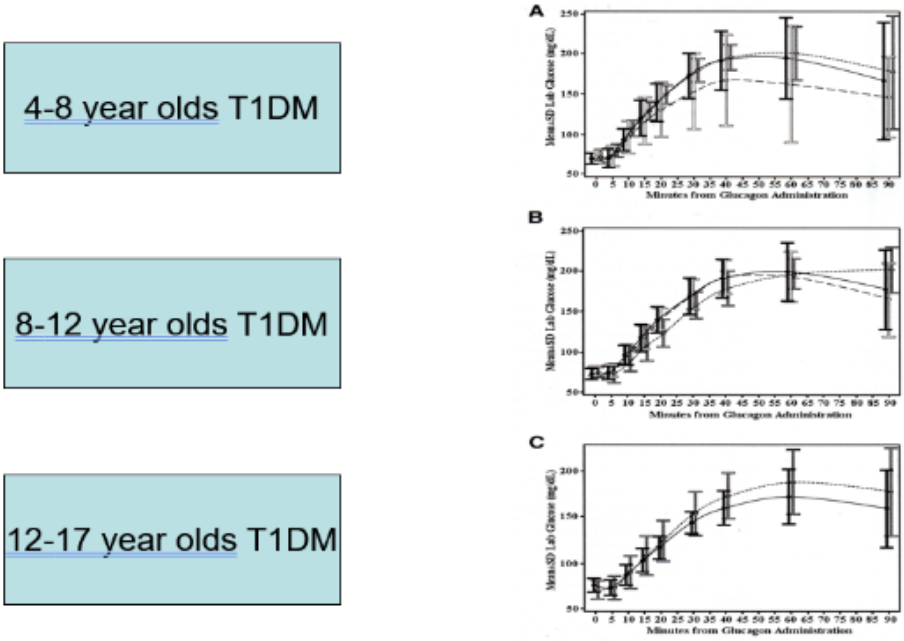
The typical Lilly, NovoNordisk and Kabi glucagon kits are shown on the left. More recently, Gvoke ® by Xeris, middle pictures above, has a prefilled syringe formulation and also a prefilled glucagon pen while G-Pen ® on the right also has a prefilled glucagon pen (shown next to a typical syringe for size comparison).



The benefits of the newer prefilled preparations are that there is no scary, confusing preparation needed in the midst of a severe or moderate hypoglycemic emergency. The same with the prefilled syringe by Gvoke although it still has a relatively large syringe and needle to be used during this type of crisis. Gvoke does not need refrigeration and Gvoke comes in a 0.5 mg dose form for 2-11 year olds and a 1.0 mg dose form for older children, adolescents and adults. The Gvoke and G-Pen also has a hidden needle and simple 2 step administration. How available are the G-voke and G-Pen formulations of injectable glucagon is also yet unanswered and similarly, what kind of insurance coverage and cost barriers will exist for these newer products is yet to be determined. The newest glucagon preparation is a nasal powder that does not require refrigeration or any liquid vehicle. It is sprayed into the nose, comes "already loaded" in a small, single disposable plastic dispenser and does not require any special or scary preparation or injection.



Our patients who participated in the clinical trials named this nasal glucagon "NasaGluc" in an unofficial "contest" we had at the time at NEDEC. Multicentered study data with participation from children, adolescents and adults all showed equivalent, positive and rapid glucagon uptake through the nasal mucosa and blood glucose increased as shown with rising glucose levels as seen in the three pediatric age cohorts studied when compared to minutes from actual nasal glucagon administration; Locemia ® developed the nasal



glucagon spray. This has been approved and brought to market by Eli Lilly as Baqsmi® nasal spray glucagon. No needles, no refrigeration, not scary and "everyone" already knows "how to use nasal spray devices" so the chances of missed doses or partial missed doses with injectable glucagon are decreased with the nasal glucagon working in equivalent fashion. There was minimal nasal irritation and occasional temporary sneezing which did not affect the blood glucose rise. As with any glucagon preparation some mild to moderate headache and nausea but no differences in the comparable trials reported. There was nearly 100% approval from

participants and their family members for nasal spray glucagon over injectable forms of glucagon when asked which was their preferred treatment if glucagon was needed. FDA approval for nasal glucagon includes 4 year old children, school-age children, teenagers and adults with the same 3 mg nasal spray dosage for all ages.

More recently there has also been some interest, especially in the small percentage of PWD who have recurring episodes of severe hypoglycemia, recurring seizures or loss of consciousness, and all strategies seem unable to stop the recurring crises. Specially trained dogs [106] called diabetes assist dogs, who are able to smell something as yet unidentified in the PWD having hypoglycemia, has allowed such dogs to alert the PWD even if hypoglycemia unawareness does not allow this to happen on its own.

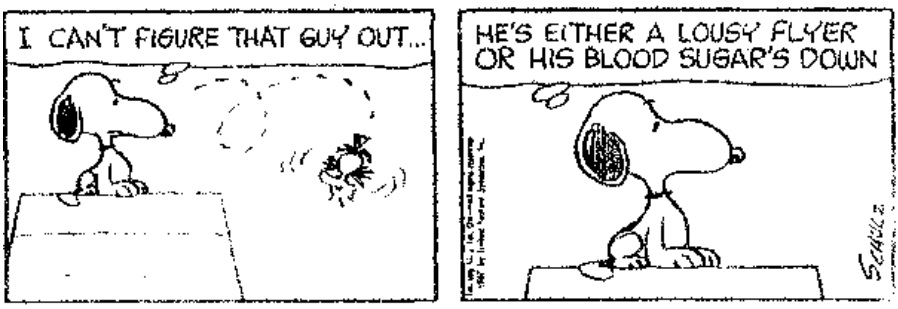
NEDEC strategies to prevent severe hypoglycemia incorporate initial teaching and then followup teaching sessions for all newly diagnosed type 1 diabetes patient and their families. At every ambulatory followup visit afterwards, blood glucose meter or sensor data is reviewed very specifically focusing on average blood glucose for the prior two weeks, time in range (TIR), percentage hyperglycemia and percentage hypoglycemia as well as any specific problems voiced by the PWD or family. Discussions about food and snacks, insulin administration, medic-alert tags, logbooks or computer/mobile phone systems for record keeping and motivational interviewing geared to help with mutual problem solving. At a minimum, every Spring, there is a more in-depth discussion around medic-alert identification, hypoglycemia causes, prevention, glucagon refills needed and who else besides the immediate family needs to get a glucagon review session: peers, friends, other relatives, scoutmasters, coaches,

school staff, school bus drivers etc. Specific questions about refills for glucagon and reminders about free coupons/waivers/programs for high copayments for glucagon are an important part of these annual sessions because we know from a recent survey following a significant episode of hypoglycemia that only 11% had filled the provided prescription following the hypoglycemic episode.[107] Asking people to practice mixing the diluent with the powder vial, withdrawing the correct dose for the PWD and answering any questions helps to emphasize the importance of having glucagon and filling refills. Similarly, finding out if school systems will want and be able to use emergency glucagon kits need specifically to be discussed too.

Hypophobia [41-43] also needs specific discussion to be started by health care professionals since it is quite rare for the PWD or their family to bring up this topic. Appropriate, and not overtreatment of hypoglycemia, is important to try to prevent hyperglycemia that often follows such overtreatment and also for helping to minimize excess weight gain from such increased calories. Purposeful excess insulin underdosing also comes into question under such circumstances. Indifference towards hypoglycemia [30] may also jeopardize physical well-being particularly when such hypoglycemia is related to alcohol or substance abuse, sloppy treatment adherence and/or lack of monitoring (ie. omitted bedtime blood glucose levels).

All members of the diabetes health care team need to have the same approach and message about known causes of hypoglycemia and prevention as well as treatment.[24] Consistency by team members allows different motivational discussions to empower better compliance, better understanding, review subtleties of each PWD and family/peers involved and optimal use of technologies for monitoring and review between visits. The overall goal for type 1 diabetes involves optimizing glucose target ranges possible with more time in range as well as less time hyperglycemic as well as hypoglycemic to be able to minimize long term complications associated with type 1 diabetes mellitus. At the

same time, it must be recognized at present that we do not have the ability to completely avoid all episodes of hypoglycemia, although we are much better at doing so with the newest insulins and the newest technologies utilizing artificial intelligence and predictive algorithms for semi-automatic or automatic responsivity.



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BONE HEALTH
IN CHILDHOOD AND ADOLESCENCE

Oana Alexandra Velea - Barta, Iulian P. Velea

Introduction

The bone is a very "dynamic" and specialized connective tissue which has a certain stiffness and resistance that provides mechanical support for the muscle activity. At the same time it provides physical protection for the internal tissues and organs and it is a deposit for systemic mineral homeostasis. [1]

From a morphological point of view, the skeleton has two components: the cortical (compact) bone with mechanical and protection functions and the trabecular (spongy) bone with mainly metabolic functions (*Figure 1*).

The cortical bone represents almost 80% of the skeletal mass and is situated in the long bone's diaphyses (ex. the femur, the radius). Histologically it is composed of dense collagen fibers concentrically arranged in cylindrical structures (Haversian systems), surrounding a central canal inside which there are blood vessels, lymph vessels and nerves. [2] (*Figure 1*)

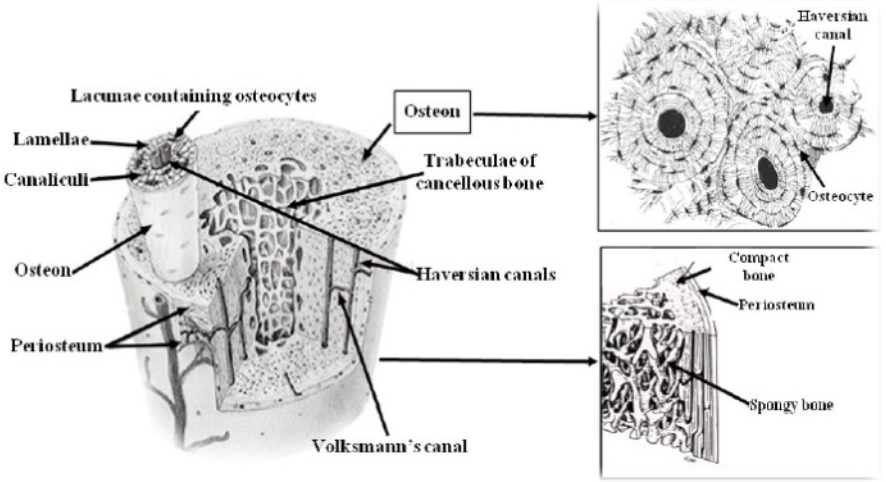

Figure 1 – Bone structure (after 3)

The trabecular bone predominates in the axial skeleton (rib cage, spine, long bone epiphysis) being formed by a network of thin threads called trabeculae, their position being determined by the pressure exerted on the bones during development.[3]

From the histological point of view, the bone has three main components: bone cells, organic matrix and the mineral part (forming 2/3 of the dry matter of the bone). The mineral part is composed of: calcium and phosphate crystals in the form of hydroxiapatite and also from small amounts of magnesium, carbonate and sodium.[4,5]

In both children and adults, the bone is constantly "active" meaning it undergoes a remodelling process that involves the continuous replacement of "old" bone with "new" one. This entire process takes place in the so-called "remodeling units". This remodeling process involves the resorption by osteoclasts of a mineralized surface followed by the recruitment of osteoblasts that secrete a new matrix capable of replacing the resorption cavity.[4]

After reaching the final height, this process is able to maintain the whole bone mass of the individual. After

reaching the final waist this process is able to maintain the bone mass of that individual.[3]

In the growing child, the formation of new bone is predominant leading to neoapposition which ensures the longitudinal growth of the skeleton and also changes in the size and shape of the skeleton.[6]

During childhood until puberty, bone mass development depends on several factors: *main* factors (genetic, ethnicity, sex), hormonal factors (growth hormone, IGF-1, primary hiperparathyroidism) and also a series of *secundary* factors: nutrition (calcium intake, vitamin D, other dietary factors), autoimmune, kidney, gastrointestinal diseases, diabetes mellitus, obesity, medication (glucocorticoids, proton pump inhibitors, anticonvulsants), physical activity.

During puberty and adolescents, stages with a fundamental role in the acquisition of bone mass, adequate thyroid hormone, growth hormone and sexual steroids production is required.[3]

It is proven that bone mass accumulation continues even after the complete longitudinal growth of the individual. The exact age at which bone mass is finalized in different areas of the skeleton is not determined precisely. It is estimated that at the spine and femur level, it is between 16-18 years old while at the level of the skull it is around the age of 35. [7] (*Figure 2*)

All these observations support the idea that the higher the bone mass at the age of the young adult, the further the risk of osteoporosis.

Main factors of bone mass accumulation.

The first distinctive phase in bone development is represented by the embryonic period in which the position and form of the skeleton elements are determined by the regulatory genes expression and also by the local growth factors.

The second phase begins with the mineralization whose location is influenced by the mechanical deformation. In this context, the theory of the mechanostat was issued according

to which the bone tissue is regulated in order to maintain its integrity and structural strength.[8]

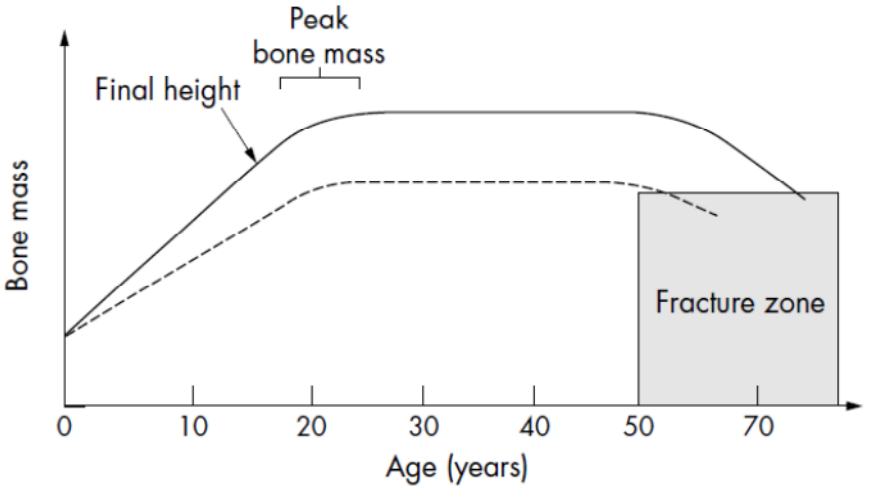

Figure 2 – Schematic representation of changes of bone mass with age as measured by DEXA.
The dotted line shows the theoretical consequence of a reduction in peak bone mass.[8]

Fetal development and bone mass

Several longitudinal studies have shown that there is a correlation between low birth weight and low mineral bone density (BMD) in adult age suggesting that intrauterine scheduling influences the risk of osteoporosis later in adulthood.

Genetic factors of bone mass.

Genetic predisposition determines up to 80% of the peak bone mass, the rest of 20% is influenced and modulated by environmental factors and by the level of sexual hormones.[9,10]

Identification as early as possible of the negative impact factors on the bone health can help children and adolescents to increase their mineral density before reaching the peak bone mass. (PBM) (*Peak bone mass*).

In 2012, the genome-wide association study (GWAS) developed on a cohort of children in Holland[11], established that WNT16 rs917727_T is associated with systemic BMD in 2660 children. This locus is also associated with BMD in adults.

Rare variants next to EN1, identified for the first time in adults, have been confirmed to be associated with a big bone mass in children.[12]

Chesi et al.[13] have reported two loci (rs7797976 in CPED1 in girls and rs7035284 at 9p21.3 in boys) associated with BMD.

The fact that the effect of genetic variants associated with BMD in childhood are associated with BMD in adults suggests that these can act during the entire life.

Hormonal factors with impact upon bone mass.

The Growth Hormone (GH) is a peptide hormone synthesized and secreted in a pulsating manner by the anterior pituitary gland, and has as primary effect, promoting linear growth in children. [14]

It's secretion is controlled mainly by growth hormone releasing hormone (GHRH) which has a stimulating effect and by the somatostatin which has an inhibiting effect. To a small extend: grelin (stimulating), IGF-1, leptin, free fatty acids and the central nervous system that regulates GH secretion through endocrine and paracrine/autocrine mechanisms and then stimulates linear bone growth, are involved.[14]

GH acts at the bone epiphysis level in order to achieve the linear growth by diferentiating precondrocytes and expanding osteoblasts.[15]

GH - IGF-1 axis represents the main endocrine system of regulating linear growth in children;[16] however, the exact cellular targets for the direct effects of GH remain imprecisely defined in complex tissues, such as growth plate. The contribution of direct and indirect actions of GH is still being discussed.[17]

In the liver, activating the GH receptor leads to a high production of IGFBP-3 and the acid-label (ALS) unity links IGF-1 in a ternary complex, thus increasing its half-life. [18]

During childhood, until final growth, the GH is the hormonal factor having the major impact regarding bone mass accumulation.

In the development of each long bone there are two secondary ossification centers- "the growth plates" located at each end of the long bones in the metaphyseal area, centers that have the endochondral ossification area.

From an anatomical point of view, the growth plate has three types of components: a cartilaginous component (the epiphyseal plaque), an ossified component (in the metaphysis) and a fibrous component (consisting of the Ranvier groove and the La Croix perichondral ring) that surrounds the periphery of the plate.

During childhood, the growth plate connects the cartilage thus permitting the long bone to grow both in length and in diameter.

The endogenous and exogenous effects of GH involve four main domains:

- They stimulate the linear bone growth,
- They grow the bone mass,
- They act on adipose tissue to increase lipolysis, inhibit lipoprotein lipase, stimulate hormonal lipase, reduce glucose transport and reduce lipogenesis.
- They act on muscles in order to grow the transport of amino acids and can affect muscular fiber distribution[19]

Thus in patients with GH deficit (GHD), GH therapy not only increases the linear growth speed but also promotes and maintains a healthy body composition, a reduced weight, normal levels of blood glucose and a favorable lipid profile.

If before puberty, bone growth depends mainly on growth hormone, sexual steroids are essential for completing the maturation of the epiphysis and for fixing bone minerals during puberty and adolescence. [20]

During childhood, height increase is relative stable; until the age of 4, girls grow slightly faster than boys; then,

until puberty, the growth process has a medium speed of 5-6 cm and 2.500 kg per year for both sexes. From early childhood to late adolescence there is a constant accumulation of skeletal mass that increases from 70-95 g at birth to 2.400 grams in women and 3.300 in young men, respectively.[1,4]

Actually between early puberty and adulthood, the skeletal mass doubles. This "accumulation" takes place at different speeds depending on the considered skeletal segment. Limb growth is finalized before the axial skeleton growth, after which, under the influence of sexual steroids, a spine growth. Taking into consideration that during puberty bone mass growth is achieved according to the maximum growth rate, the bone is sub-mineralized but this period is transient because bone mass accumulation continues even after length increase of the individual is finalized.[3] The exact age at which bone mass is maximum in different areas of the skeleton is not precisely defined and it varies from 16-18 years (spine) even until 35 years (the skull). [5,20]

Nutritional and environmental factors

All this complex succession of hormonal changes can be influenced by nutritional and environmental factors, which are capable to modify the genetic potential of the individual.

Vitamin D and bone growth

Vitamin D performs the intestinal absorption of calcium, this being essential for the growth plate calcification and osteoid mineralization at the trabecular and cortical bone level.[21]

It is well known that Vitamin D sources are either exogenous (vitamin D2- ergocalciferol that comes from vegetal sources and vitamin D3 - colecalciferol that comes from animal sources), or endogenous (colecalciferol that is synthesized in the skin by transforming 7 dehydrocholesterol under the action of ultraviolet rays). Regardless of their origin, they reach the liver where they suffer a hydroxylation under 25-hydroxylaze action, therefor forming 25(OH)D that reaches the kidneys where it suffers the second hydroxylation under

the action of another enzyme (1-alpha-hydoxylaze), $1,25(HO)_2D$ active metabolit being obtained which increases calcium and phosphorus absorption at the intestinal level, increases tubular re-absorption of Ca by the kidneys and contributes to calcium ions deposition in the bone.

In case of low serum vitamin D concentration, intestinal absorption of calcium and phosphorus is decreased with low levels of ionic calcium. Under these conditions there is an increased secretion of PTH, important regulator of 1-alpha-hydroxylase that helps in maintaining a normal range of 1,25(OH)D in order for an optimum intestinal calcium absorption to be achieved.

Generally, serum level of 25(HO)D or calcidiol is considered to be a good indicator of the nutritional status of vitamin D.[22] Depending on the serum level of 25(HO), vitamin D status can be:[23]

- sufficient > 50 nmol / L (>20ng/ml)
- insufficient: 30 – 50 nmol/L (12-20 ng/ml)
- Deficient < 30 nmol/L (< 12 ng/ml)

Despite the consensus on these vitamin D levels, there is a considerable disagreement regarding the optimum level of 25(OH)D that should be reached and maintained throughout the year.[24] Nevertheless it can be states that 20 ng / ml (50 nmol / L) represents a threshold value that should be reached throughout the year by all individuals regardless their age.

With puberty onset and throughout adolescence, $1,25(OH)_2D$ increases calcium absorption according to the increase of bone mineral accumulation. Several studies regarding calcium absorption in children and adolescents, ages between 4.9 and 16.7 years old with 25(OH)D levels between 28 and 197 nmol / l, have not identified a relationship between circulating levels of 25(OH)D and calcium absorption. [25,26]

Winzberg et al. have reported in an extended meta-analysis,[27] developed on six studies with 541 persons that received vitamin D and 343 that received placebo, aged between 1 month and 20 years that dietary supplementation with vitamin D affects bone mineral density increase in

lumbar spine level but not in the hips and forearm. Individuals with vitamin D deficit can benefit from vitamin D supplementation, the benefit not being found in children and adolescents with vitamin D normal levels. [27]

Vitamin D receptor

In children in comparison with adults, vitamin D receptor polymorphisms have a greater influence on bone mineral density (BMD).

In a study developed on prepubertal girls, dietary calcium intake has been correlated with bone mineral density modification which suggests that there is a variation of the response to calcium supplementation. [8]

On the other hand, estrogen receptor polymorphisms in adolescents, IL-6 and osteocalcine genes are independent predictors of bone mineral density.

Calcium effect on bone growth

Calcium represents 1% –2% of the human body weight in adults, more than 99% from the body total calcium being found in bones and teeth.[28] Calcium trans-epithelial absorption is performed in the epithelial cells from the intestinal lumen under vitamin D action.

In a double-blind randomized study with a duration of 12 months Dibba et al. evaluated in children aged 8,8-11,9 years the effect of calcium supplementation upon bone mineral content (BMC) and established that the group with calcium supplement had BMC and BMD greater at the distal race and medium ax comparing to the control group. [29]

In the randomized study developed by Prentice et al. conducted on a period of 13 months, found that in boys aged 16-18 years, calcium carbonate supplementation (1000 mg calcium / day) led to an increase of bone mineral content in: the lumbar spine, hip and intertrochanteric comparative to the control group that received placebo. [30]

Body exposure to the glucocorticoids.

Bone mass loss after exposure to glucocorticoids (glucocorticoid osteoporosis) is the result of supressing bone formation and improvement of osteoclast activity.

Glucocorticoids lower IGF-1 and sexual steroids expression, and on the other hand inhibit renal reabsorption of tubular calcium.[31]

Type 1 Diabetes Mellitus

Osteoporosis pathogenesys in long-term unbalanced type 1 Diabetes Mellitus is complex and is related to the calcium, vitamin D metabolism and most likely to the subinsulinization of the organism, having as consequence, blood calcium and phosphorus levels decrease. Hyperglycemia seems to stimulate excess formation of IL-1 and IL-6 pro-inflammatory cytokines that can promote osteoclast activity inhibiting osteoblast differentiation and mineralization.[32]

Gastrointestinal diseases

Celiac disease in children is often found in those who have type 1 DM, due to the nutritional deficiency secondary to malabsorption being associated with bone loss.[33] The decrease in calcium absorption and the increased level of inflammatory cytokines including IL-1, IL-6 and TNF-α, seem to be responsible for increase bone resorption. [34]

Ulcerative colitis and Crohn disease are also associated with bone loss. The mechanisms responsible for bone loss include the inflammatory activity linked to the disease and also should be mentioned the secondary effects linked to the treatment including glucocorticoid therapy and nutritional deficiencies.

Autoimmune diseases

The underlying mechanisms of bone loss from autoimmune diseases (eg. Rheumatoid arthritis, ankylosing spondylitis, lupus erythematosus, multiple sclerosis) are multiple and complex. On one hand, the severity of the underlying disease comes into question and on the other hand the use og glucocorticoids in long-term schemes. TNF-α, IL-1, IL-6 pro-inflammatory cytokines levels, increased in patients with autoimmune diseases stimulate the osteoclast activity

that secrete proteolytic enzymes and HCl that induce bone mass decrease and even osteoporosis.[35]

Kidney diseases

Renal tubular acidosis is characterized by a hyperchloremic metabolic acidosis.

Increasing the concentration of hydrogen ions when prolonged causes the activation of the buffer system in the bone with the activation of osteoclasts and the mobilization of calcium ions in the bone leading to metabolic bone disorders that can range from osteomalacia to osteoporosis and even fractures. [35]

In children with CKD, abnormal mineralization begins early during CKD [12], but, unlike adult patients, the risk of fracture does not exceed 10%. [35]

CKD is a systemic inflammation, triggered by the accumulation of uremic toxins, immune cell defects, intestinal dysbiosis, dialysis factors and other associated comorbidities.[36] All these factors lead to osteoclast differentiation and bone resorption, inhibiting osteoblast differentiation by mobilizing a large amount of calcium from the bone, resulting in a deficit of bone mineralization.[37]

Anorexia in CKD is related to an imbalance between orexigenic and anorexigenic circulating hormones. There are studies that suggest that leptin (anorexigenic hormone), which controls bone mass, associated with high levels of proinflammatory cytokines, promotes bone resorption and inhibits osteoblast proliferation through a hypothalamic relay.[38]

Vitamin D deficiency and secondary hyperparathyroidism

In CKD, vitamin D deficiency induces mineral deficiency and secondary chronic hyperparathyroidism, which eventually induce altered bone metabolism.

In children with CKD, both low levels of 25(HO)D and high levels of parathyroid hormone (PTH) were correlated with low bone mineral density (BMD), lower cortical volume, and ultimately a higher risk greater fracture.[35]

Secondary hyperparathyroidism also affects skeletal muscle structure, proteins, amino acids and energy metabolism by stimulating muscle proteolysis, increasing the release of alanine and glutamine and affecting energy production, transfer and utilization[39] which explains the reduced muscle strength of these patients.[40]

Chronic metabolic acidosis leads to the dissolution of bone calcium carbonate, activation of bone resorption and inhibition of bone formation mediated by osteoblasts.

May contribute to the suppression of renal synthesis of $1,25(HO)_2D$. It appears to disrupt the GH-IGF-1 axis at the growth plate, disrupting longitudinal growth.

Disruption of the G-IGF1 axis

Systemically, IGF-1 secreted by the liver, as well as IGF-1 produced locally by osteocytes / osteoblasts and myocytes / myoblasts, has pleiotropic effects on the musculoskeletal system. In CKD, there was an insensitivity to GH and resistance to IGF1.

In a recent study[41] IGF-1 was positively correlated with lean mass and negatively associated with the occurrence of bone fragility in children with CKD.

Insulin resistance (IR)

Insulin is also an anabolic factor for bone tissue, promoting the proliferation and differentiation of osteoblasts and inhibiting osteoclastic activity, while stimulating collagen synthesis, alkaline phosphatase production and glucose absorption.[42]

Data in pediatric patients are limited. A recent study showed that IR was negatively correlated with poor relative mass. [43]

Therapeutic strategies in CKD

Given the results of the studies, the therapeutic targets for improving musculoskeletal integrity in CKD are:

- Regulation of homeostasis of calcium, phosphorus, vitamin D and PTH,
- Correction of metabolic acidosis,
- Recombinant growth hormone (rGH) therapy
- Physical activity,

- Optimizing the nutritional status of the patient,
- Dialysis with low inflammatory impact.

Medication

A form of secondary osteoporosis is also drug-induced. In addition to glucocorticoids, a number of other drugs with an impact on bone metabolism are implicated in bone loss.

Proton Pump inhibitors by increasing gastric pH reduce calcium absorption thus having a negative impact on bone metabolism.

Anticonvulsivants commonly used in pediatrics can lead to bone loss. Although the mechanism is incompletely elicated, they increase vitamin D metabolism, so they lead to a low level of 25(OH)D and implicitly induce a secondary hyperparathyroidism, thus increasing bone loss.

Obesity

In early childhood, body weight can be an important indicator of a child's bone health.

Current research investigating the impact of fat on BMD is contradictory. Previous studies have concluded that adipose tissue has not been beneficial to bone structure,[44] while another study suggested that fat mass would have a positive relationship with bone mass in children.[45]

Some studies have found positive correlations between bone mass / density and overweight / obesity,[46] while others have shown opposite conclusions.

In most of these studies, bone mineral density was measured in several places by DXA which may better reflect the bone mineral density of adolescents.

It was found that obese girls had both a higher concentration of serum leptin and a higher BMD.

On the other hand, overweight / obese children have several risk factors (vitamin D deficiency, inadequate calcium intake, sedentary behavior) that could lead to accelerated bone loss. [47]

Physical activity

A number of studies have shown that physical activity can increase bone metabolism, increase bone mineral density, form a good bone structure and thus strengthen bone mass.[48]

In the US Report of the Advisory Committee on Physical Activity Guidelines, it is recommended that children between the ages of 6 and 17 engage in moderate to vigorous physical activity for 60 minutes or more each day.[49]

In addition, it is recommended to promote regular physical activity associated with adequate calcium intake in children and adolescents to improve bone health. This combined effect was more pronounced when the initial level of calcium intake was low and in the early stage of puberty.

For weight-bearing bones, the effect of high-calcium physical activity on bone mass was greater than that of exercise or calcium supplementation alone.

For non-bearing bones, calcium supplementation, whether or not accompanied by an increased level of physical activity, was more effective.

Prospective studies are needed to establish the dose-effect relationship on bone mass of both physical activity and calcium intake to determine calcium supplementation and the level of physical activity that leads to improved bone health.[50]

Conclusions

Since bone mass ends at the end of the individual's longitudinal growth, bone accumulation during childhood and adolescence is an essential factor in determining the risk of adult osteoporosis.

Consequently, more attention should be paid to prevention strategies, from the earliest age of children and the promotion of at least moderate physical activity lasting 60 minutes daily.

Ensuring a normal bone density should be a priority when dealing with children and adolescents. To achieve this goal it is necessary to ensure adequate nutrition that brings a sufficient intake of calcium, vitamin D, with the removal of

secondary (pathological) factors with possible negative impact on bone mass accumulation.

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UPDATE IN HYPOPHOSPHATEMIC RICKETS

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Introduction

Rickets is a condition that occurs commonly in small children as a result of vitamin D, calcium, and phosphorus deficiency.[1] Bone mineralization is thus affected through abnormalities of the growth plate cartilages, most evident in long bones[2]. In children, rickets may often associate osteomalacia, while in adults, osteomalacia is referred to as the equivalent of rickets.

Based on its primary pathologic mechanism, rickets is a disease that ca be classified into two categories: calcipenic and phosphopenic. Common calcipenic rickets is basically a Vitamin D deficiency or defect in efficiency of various etiologies, which translates into low Calcium and Phosphorus intestinal absorption associated with low efficiency of bone mineralization, these two roles being played by Vitamin D. Moreover, low Calcium sets off parathormone (PTH) secretion which has the main role of elevating Calcium serum levels by mobilizing bone Calcium, increasing intestinal absorption and renal reabsorption. High levels of PTH also lead to renal loss of phosphates, leading to hypophosphatemia.[3]

All in all, calcipenic rickets is a deficiency disorder, manifesting in low serum Calcium and Phosphorus, low 25(OH) Vitamin D (and/or low 1.25(OH)2 Vitamin D) and high PTH levels. Most importantly, all these imbalances are easily corrected with Vitamin D supplements, correct exposure to sunlight or management of the underlying disease which

prevents Vitamin D absorption or activation. Should intervention occur under the age of 1, calcipenic rickets resolves without massive sequelae, however, with no treatment, it resolves itself on its own, but with evident sequelae.[4]

On the other hand, in hypophosphatemic rickets, a disease with multiple inherited or acquired etiologies, the primary defect is phosphate deficiency, which occurs by increased renal phosphate loss.[2] Associated to this is a deficit of Vitamin D renal activation which translates in a reduced intestinal absorption of Calcium and Phosphorus[5]. Hypophosphatemia affects the growth plate by preventing apoptosis and thus, causing accumulation of hypertrophic cells which form the rachitic bone.[3] The usual laboratory findings would be hypophosphatemia, normal 25(OH) Vitamin D, but low 1.25(OH)2 Vitamin D, normal/slightly low serum Calcium and normal/slightly elevated PTH levels. Another clue for a general practitioner would be the lack of blood parameters and clinical correction after administration of 25(OH) Vitamin D supplements.

Hypophosphatemic rickets is considered either a disease on its own, either a complication of other hypophosphatemic disorders caused by proximal tubular dysfunction. Clinical setting and correct identification are extremely important for detecting the cause and the most appropriate course of therapy. Most phosphate renal wasting disorders are inherited as isolated defects or associated with generalized proximal tubular dysfunction (Fanconi syndrome), but some may be acquired (tumor-induced osteomalacia, Fancony syndrome induced by drugs).

Phosphate loss

The pathophysiology of phosphate renal loss is intricate, involving many key players and key pathways. The Fibroblast growth factor 23 (FGF23), PTH and 1.25(OH)2 Vitamin D (calcitriol) are the most important players in keeping the phosphate homeostasis.

- FGF23 is a phosphatonin secreted by osteocytes, normally as a consequence of increased calcitriol. This phosphatonin is involved in phosphate excretion and down-regulating the renal activation of vitamin D, reducing thus the 1.25(OH)2 Vitamin D levels.[6,7]
- FGF23 binds to tyrosine kinase receptors (FGFRs 1-4), of which FGFR1c is the most proeminant. These receptors need full-length klotho proteins to convert into full FGF receptors with specific affinity, in order to determine downstream signaling molecules.[8,9,10] Klotho has a co-receptor function because it binds to FGF23 and FGFR1c simultaneously and thus, confers stability to the entire complex.[11]
- In the proximal renal tubules, FGF23 binds to FGF receptor-klotho complexes and activates two kinases – "extracellular signal-regulated kinase-1/2" (ERK) and "serum/glucocorticoid-regulated kinase-1" (SGK). SGK-1 determines the "Na^+/H^+ exchange regulatory cofactor-1" to down-regulate the membrane expression of the "sodium-phosphate co-transporter" in charge of phosphate reabsorption. Therefore, phosphate urinary excretion will increase.[12,13,14]
- On the other hand, suppression of 1α-hydroxylase expression which is responsible for 1,25(OH)2 Vitamin D production, is done also by FGF23 binding with klotho and signaling downregulating mechanisms in the proximal tubules. Less active serum Vitamin D means a decrease in intestinal phosphate absorption, an addition factor to low serum phosphorus levels.[15,16]

HYPOPHOSPHATEMIC RICKETS EVALUATION REGARDLESS OF CAUSE

Hypophosphatemic rickets can be diagnosed and managed by blood, urine and imagistic analysis.
In the Table 1, showed below, the panel of useful parameters.

Table 1. Necessary parameters in evaluating XLH

Type of analysis	**Specific parameter**	**Usual findings**
Serum	Phosphorus	low
	Calcium	slightly low/normal
	PTH	normal/slightly elevated
	Alkaline phosphatase	very elevated
	FGF23	elevated
	1,25(OH)2 Vitamin D	low/inappropriately normal
	25 (OH) Vitamin D	normal
	TMP/GFR	low
Urine exam /24 h Needed for calculation of tubular phosphate reabsorbtion rate	Urine phosphate	normal/low
	Serum creatinine	normal
	Urinary creatinine	normal
Radiology	Bone age	retarded
	Bilateral anteroposterior Wrist Rx and Knee Rx	hypertransparent bones enlarged epiphysis ill defined zone of provisional calcification
	Rickets severity score (Figure 2, Table 4)	0-10, 10 being the worst modifications

Renal phosphate transport is evaluated by the **ratio of tubular maximum reabsorbtion of phosphate to glomerular filtration rate (TmP/GFR)**. This ratio is influenced by dietary phosphate, PTH, Vitamin D and FGF23 levels, and metabolic disorders like acid base disorders or extracellular fluid volume disturbance. Calculation of TmP/GFR assumes that serum phosphate concentration is equal to its concentration in the glomerular filtrate, and that creatinine clearance is a close approximation to GFR.[17,18,19]

Another useful tool in assessing tubular health is the **fractional tubular reabsorbtial of phosphate (TRP).** A low TRP in the context of hypophosphatemia points to a renal tubular defect.[17]

- Calculate the ratio of phosphate clearance to creatinine clearance (CP/CCr):

$$\textbf{CP/CCr} = \frac{\textbf{serum creatinine x urine phosphate}}{\textbf{urine creatinine x serum phosphate}}$$

This ratio is normally less than 0.15 and is often elevated in primary hyperparathyroidism.[17]

- The fractional tubular reabsorption of phosphate (TRP) is calculated by this formula[17]:

$$\textbf{TRP} = 1 - \frac{\textbf{serum creatinine x urine phosphate}}{\textbf{urine creatinine x serum phosphate}}$$

- If TRP is ≤ 0.86, phosphate reabsorption is maximal and there is a linear relationship between plasma phosphate concentration and excretion and TmP/GFR which is calculated by the fomula:

$$\textbf{TmP/GFR} = \textbf{TRP x serum phosphate}$$[17]

- If TRP is > 0.86, relationship between plasma phosphate concentration and excretion is curvilinear and TmP/GFR is defined as follows:

$$\textbf{TmP/GFR} = \alpha \textbf{ x serum phosphate,}$$

where $\alpha = \frac{\textbf{0.3 x TRP}}{\textbf{1- (0.8 x TRP)}}$ [17]

- TmP/GFR can also be determined using a nomogram after measuring fasting plasma and urine concentrations of phosphate and creatinine.

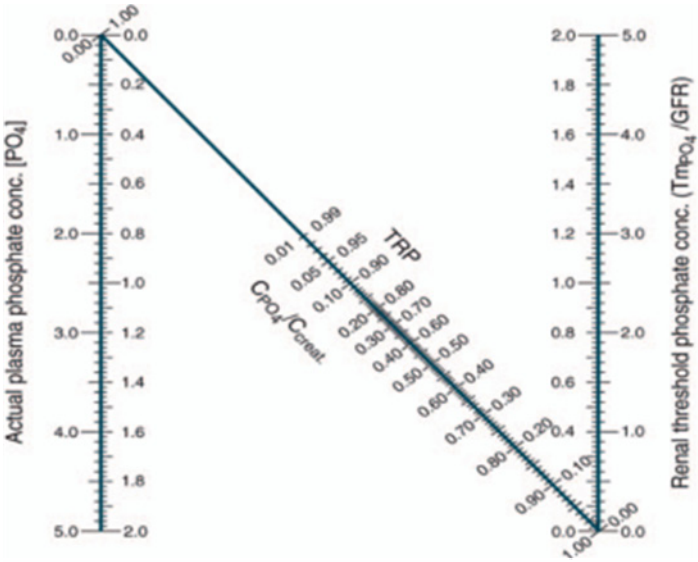

Figure 1. Nomogram to determine tubular maximum reabsorption rate TmP/GFR given plasma phosphate (PO4) concentration and tubular reabsorption of phosphate (TRP). On the vertical axes, the inner scale is in mmol/L, whereas the outer scale is in mg/100 mL. TRP is calculated as detailed in text. A straight line joining plasma phosphate concentration, TRP and the right vertical axis gives the TmP/GFR.[17]

Interpretation: Low levels suggest renal phosphate wasting.[17]

Age-related reference ranges for TmP/GFR are given below (taken from reference.[17]

Table 2. Adult ranges [17]

Age	**Male Range (mmol/L)**	**n**	**Female range (mmol/L)**	**n**
25-35 years	1.00-1.35	15	0.96-1.44	15
45-55 years	0.90-1.35	15	0.88-1.42*	15
65-75 years	0.80-1.35	15	0.80-1.35	15

**Premenopausal*

Table 3. Paediatric Ranges [17]

Age	**Range (mmol/L)**	**n**
Birth	1.43-3.43	20
3 months	1.48-3.30	20
6 months	1.15-2.60	20
2-15 years	1.15-2.44	101

The Rickets Severity Score (RSS) was developed by Thatcher et al. and is useful in the therapy efficiency follow-ups.[20]

The schematic picture below is taken from his published work and so is Table 4.[20]

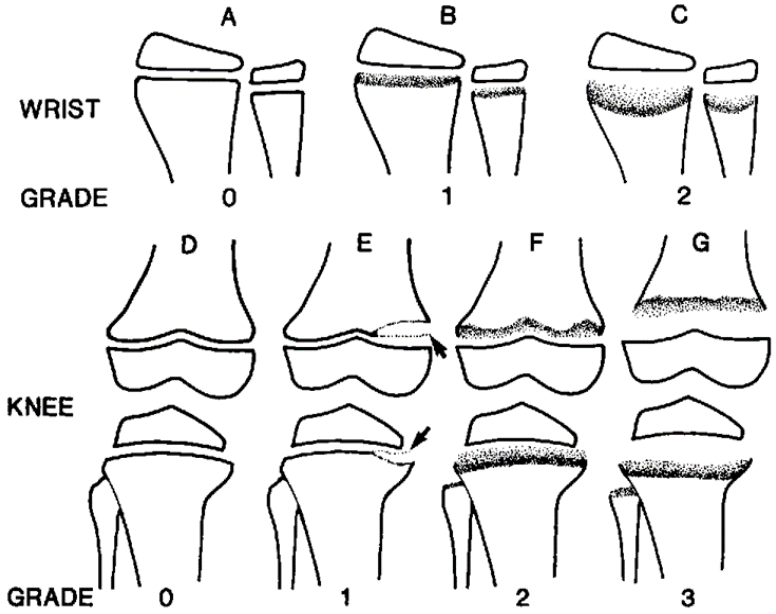

Figure 2. Schematic representation of wrist and knee destruction of bone, by Thatcher et al. [20]

Table 4. Calculation of RSS according to Thatcher et al [20]

Grade definitions for radius & ulna

0	Normal growth plate without changes of rickets				
0.5	Lucency of metaphyseal margin without fraying or irregularity				
1	Widened growth plate, irregularity of metaphyseal margin, but without concave cupping				
1.5	Partial metaphyseal concavity or incomplete fraying of metaphyseal margin				
2	Metaphyseal concavity with fraying of margins				
Grade radius	0	0.5	1	1.5	2
Grade ulna	0	0.5	1	1.5	2
SCORE:	Radius grade ___ + Ulna grade___ = Total wrist___				

Grade definitions for femur & tibia

0	Normal growth plate without changes of rickets			
1	Partial lucency, smooth margin of metaphysis visible			
2	Partial lucency, smooth margin of metaphysis NOT visible			
3	Complete lucency, epiphysis appears widely separated from distal metaphysis			
Grade femur	0	1	2	3
Grade tibia	0	1	2	3
Femur & tibia multiplier	0.5 if ≤ one condyle or plateau affected		1 if two condyles or plateaus affected	
SCORE: (Femur grade ___ × multiplier ____) + (Tibia grade___ × multiplier ____) = Total knee___				

X-LINKED PHOSPHATEMIC RICKETS (XLH)

Firstly described by Albright in 1939, the most common cause of inherited rickets with an incidence of 1:20.000 live births, this type of hypophosphatemic disorder is caused by an X-linked dominant mutation of the PHEX gene.[21]

Mutated PHEX (Phosphate regulating gene with Homologies to Endopeptidase on X chromosome) genes present inactivation mutations. PHEX contains information for a metalloprotease that cleaves small peptide proteins with expression in bone, teeth and parathyroid glands. Although this metalloprotease does not cleave FGF23 directly, it is involved in the downregulating of FGF23 secretion and activity by a yet unknown mechanism.[22,23] Should the gene containing information for this metalloprotease be inactivated, levels of FGF23 and of other small peptide hormones become higher than normal. As mentioned before, FGF23 induces hypophosphatemia by increased urinary phosphate excretion and low phosphate intestinal absorbtion via defective Vitamin D activation. Moreover, an intinsic osteoblast defect contributes to the disastrous effect of hypophosphatemia on bone mineralization, without response to conventional therapy. Mutations of PHEX can pe found in 50-70% of affected patients and up to 300 mutations of PHEX can occur.[24,25] This is why severity of symptoms can vary even among families. There is a very important place for research in the area of which PHEX mutations produce which XLH phenotype.[26]

Clinical aspects of XLH rickets

The only evident finding in the first year of life is the frontal bossing, which can appear as early as 6 months of age. Craniotabes or rachitic rosary are absent. After the child starts walking, progressive lower limb deformities leading to bilateral coxa vara, genu varum, and very rarely, genu

valgum, become more and more apparent. Many children present dental abnormalities as well (abscessed noncarious teeth, enamel defects, enlarged pulp chambers or taurodontium), but this is not a rule.[27] In adulthood, patients have short stature due to very evident genu varum and may present bone pains and even pseudofractures.[28]

Paraclinical aspects of XLH rickets

Blood analysis shows low phosphorus, normal calcium, normal PTH (or slightly elevated), low 1,25(OH)2 Vitamin D, decreased TmP/GFR. If possible, FGF23 should be done as well, with elevated values.

Genetic testing may detect a PHEX mutation, but quite often, one can get a negative result. This does not infirm the presence of hypophosphatemic rickets, it postpones the etiologic diagnosis. When clinical and paraclinical signs are clear, genetic testing should be repeated.

Radiological investigations show signs of severe carential rickets, and, without treatment, the rickets severity score aggravates progressively as the child ages. In the presence of correct treatment, radiological cure happens slowly, over the course of at least 2 years.[20]

Treatment

Treatment is based on phosphate and 1.25(OH)2 Vitamin D replacement. In Romania, phosphate supplements are not available on the market, but phosphate solutions can be prepared in some pharmacies. Phosphate tablets can be procured from abroad (EU), quite easily, through certain pharmacies (Phosphate Sandoz, effervescent tablets 500 mg) and we highly recommend that patients procure them because compliance to the treatment is much better.

Doses of phosphate should be 20-40 mg/kg/day, divided in 3-5 doses (maximum of 2-3 g/day). Doses of

1,25(OH)2 Vitamin D should be 1-3 ug/day [2]. Both products should be titrated progressively, by monthly monitoring the normalizing of serum phosphorus levels and alkaline phosphatase, and keeping calcium, and PTH in normal ranges. To achieve this, monthly blood and 24 h urine samples must be given until normal targets are achieved, and afterwards, check-ups every 3 months are recommended (serum phosphorus, calcium, alkaline phosphatase, creatinine and urine phosphorus and creatinine). PTH levels should be checked once a year. Adverse effects of phosphorus supplement is diarrhea, while excess vitamin D causes asthenia, nausea, headache, diarrhea/constipation, muscle pain or excess sweating. Potential complications of therapy are nephrocalcinosis and tertiary hyperparathyroidism, therefore, renal ultrasounds are necessary before starting treatment and every yearly afterwards.[29,30]

Due to the fact that this replacement treatment is complicated, with low compliance and many unpleasant adverse effects, a biological, etiological treatment in the form of monoclonal antibodies anti-FGF23 (Burosumab, Crysvita) was developed. Burosumab is taken into consideration when conservative replacement therapy does not work. These antibodies bind the circulating excess FGF23 and blocking thus their activity, blocking excess renal phoshpate loss and improving activation of Vitamin D.[31]

Burosumab

- *Dosage forms:* subcutaneous injectable solution, single-dose vials: 10mg/mL, 20mg/mL, 30mg/mL
- *Dosage:* 1 mg/kg SC every 4 weeks; round dose to nearest 10 mg. Not to exceed 90 mg/dose.
- After treatment initiation, assess fasting serum phosphorus monthly, measured 2 weeks post dose, for

the first 3 months of treatment, and thereafter as appropriate.

Discontinue oral phosphate and active vitamin D analogs 1 week before initiating treatment. Do not administer active vitamin D analogs during treatment.

Monitor 25-hydroxy vitamin D levels. Supplement with cholecalciferol or ergocalciferol to maintain 25-hydroxy vitamin D levels in the normal range for age.

Concerns: Being a new therapy, there is little information on long-term effects.

- Tissue calcifications? [31]
- Dental abscess? (31% Burosumab vs. 6% placebo, according to Imel EA et al. [32]

OTHER HYPOPHOSPHATEMIC TYPES OF RICKETS

AUTOSOMAL DOMINANT HYPOPHOSPHATEMIC RICKETS (ADHR)

ADH rickets is the cause of a mutated FGF23 gene, dysplaing incomplete penetrance and different individual age onset. The cleavage of intact FGF23 molecule is impaired, leading to its prolonged activity, and thus, renal phosphate loss.[33]

There are two groups of ADHR patients, classified by age of onset. One group presents hypophosphatemic rickets signs since childhood and, thus, have many similarities to XLH rickets. The other group presents signs in their adolescence or young adulthood. They do not present bone deformities, but bone pains and even pseudofractures.[33]

Biochemical and radiological findings, as well as treatment, are the same as in the case of XLH rickets.

AUTOSOMAL RECESSIVE HYPOPHOSPHATEMIC RICKETS (ARHR)

ARH rickets occurs by mutations of DMP1 and ENPP1 genes. When dentin matrix protein 1 (DMP1) is mutated, the disease is called ARHR type 1. DMP1 is a bone matrix protein present in osteoblasts and osteocytes,[34] responsible for downregulating FGF23 and proliferation of osteocytes. Its mutation impairs leads to loss of it function.

ARHR type 2 is a consequence of mutated ectonucleotide pyrophosphatase/phosphodiesterase 1 (ENPP1).[35] This phosphodiesterase has the role of generating pyrophosphate, the essential inhibitor of calcification. Therefore, its loss of function leads to abberant ectopic calcifications.[30] Both DMP1 and ENPP1 mutations can associate high levels of FGF23. Biochemical and radiological findings, as well as treatment, are the same as in the case of XLH rickets.

HEREDITARY HYPOPHOSPHATEMIC RICKETS WITH HYPERCALCIURIA (HHRH)

HHRH is the cause of mutated SLC34A3 gene, the gene responsible with encoding NaPi2c.[36,37]

The manifestations of this type of hypophosphatemic rickets include bone pains, weakness and pseudofractures, but no dental abcesses or other abdnormalities.

This type of rickets has the most increased risk of nephrolitiasis.[38] Apart from hyperphosphaturia, these patients present hypercalciuria as well. FGF23 levels are normal, as are 1,25(OH)2 Vitamin D kevels. The management and treatment differs from XLHR: phosphate replacement is done, but no analog of active Vitamin D is needed because it only worsens hypercalciuria and increases the risk of nephrocalcinosis.

FANCONI SYNDROME

When hypophosphatemic rickets is associated with generalized proximal tubular dysfunction, the diagnosis may be Fancony syndrome of various etiologies (cystinosis, Lowe's syndrome, proximal renal tubular acidosis type II, tyrosinosis type 1, drugs, Fanconi Bickel syndrome, etc.).[39]

Manifestations include glycosuria, hypokalemia, proximal renal tubular acidosis, hyperuricosuria and generalized aminoaciduria. Treatment is etiological.

TUMOR-INDUCED OSTEOMALACIA OR ONCOGENOUS OSTEOMALACIA

Unlike the disorders described above, this acquired paraneoplastic disorder occurs as a result of phosphatonins produced by mesenchymal tumors of long bones, distal extremity, sinuses, nasopharynx, groin, etc.

These are benign tumors that may present at any age and the patients usually have a long history of bone pains and muscular weakness. The younger the onset, the more similar manifestations are to XLH rickets.[40]

Biochemical findings are similar to XLHR.

Curative treatment is possible through surgery, but when tumors are not located, replacement therapy similar to XLHR can provide symptom relief.

Localization of these type of tumors is done most often through whole-body MRI, but octreotide scintigraphy, 18F-fluorodeoxyglucose positron emission tomography or whole-body venous sampling of FGF23 are efficient as well.[41]

Case report

We present the case of a 3 years and 8 months old girl, with XLH rickets. She has been in foster care from the age of 2 due to severe negligence from her parents. Signs of rickets were present at first consult, with bowing of her legs after the age of one, according to her family doctor.

Family history: Both parents intellectualy disabled, an older sister with lower limbs deformations (affirmatively).

Clinically: Mild facial dysmorphia: narrow forehead, low anterior hairline, bilateral epicanthus, no frontal bossing. Short neck. Oral and dental status according to her age, no abnormalities. Flaring thorax at the basis. Rachitic rosary. Thickened wrists and ankles. Short stature (<5th%tile, SD=-3.17) due to marked bowing of her lower limbs. Bilateral genu varum and coxa vara.

Other diagnosis: myopia forte, mild mental retardation and speech development disorder, probably due to low stimulation.

Paraclinical findings:

BLOOD TESTS		**24 h urine exams**
Calcium (ion & total) N/↓		Urine Phosphate: N
Alkaline phosphatase	↑↑↑	Urine Calcium: N
Phosphorus	↓	Phosphate Reabsorbtion Rate: N
25 (OH)- D3 Vit	↓	
1.25 (OH)2 D3 Vit	↓	
PTH	N	
FGF-23	↑	

BONE AGE	**Lower limbs + basin RX**	**Orthopedic consult**	**GENETIC TESTS**
Bone age corresponding to an age of 2. (Bone age - chronological age = 1 year and 8 months)	Specific modifications for sequelae of rickets.	Excluded Blount's disease due to the Rx aspect. Recommended bilateral orthoses.	***1st testing*** -> Excluded X-linked (PHEX), autosomal dominant (FGF23) and autosomal recessive (ENPP1) mutations, as well as hypophosphatasia (ALP ↑↑↑). ***2nd testing -> CONFIRMED PHEX MUTATION.***

Recommended treatment:

- 1.25-dihydroxycholecalciferol 1-1.5 ng/day
- Phosphate Sandoz effervescent tb, 2 x 500 mg/day
- Calcium carbonate 80mg/kg/day
- Magnesium 75mg/day

Currently in the works for BUROSUMAB initiation.

- Orthopedic treatment: bilateral orthoses; recovery osteotomies only after radiological recovery and ALP normalization.
- Re-evaluations at intervals of 3 months.

Renal phosphate wasting disorders are important causes of rickets to be thought of in appropriate clinical setting. Correct identification of these disorders is important for determining therapy.

In the last few years, our knowledge of a number of new inherited phosphate wasting disorders has expanded. This has contributed to the identification of previously unidentified regulators of renal phosphate reabsorption.

Yet, many more mechanisms are to be unraveled and the world of phosphate homeostasis is getting fascinating day by day.

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september 2021

THYROID NODULES AND CANCER IN CHILDREN

Hilary Seeley

Outline:

- Thyroid nodules
- Thyroid vs non-thyroid
- Initial evaluation
- When to biopsy
- Malignant vs benign
- Thyroid cancer
- Types
- Risk factors
- Pediatric guidelines
- Diagnosis and staging
- Treatment
- Surgery
- Radioactive iodine ablation
- Molecular markers
- Tumor markers

Neck mass – thyroid vs non-thyroid:

Congenital	Inflammatory	Neoplastic
Branchial cleft cyst (anterior to SCM)	Infectious	**Thyroid**
Thyroglossal duct cyst (midline mass in anterior neck)	Non-infectious (sarcoid, Kawasaki)	**Lymphoma**
Vascular anomaly		Salivary gland (parotid)
Laryngocele		Paragangliomas
Thymic cyst / Ectopic thymus		Schwannoma
		Metastatic head/neck carcinoma

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Neck mass – initial evaluation:

Examination:	Imaging:	Laboratory studies:
Location of neck mass	Neck ultrasound: thyroid and all neck regions	TSH, freeT4, thyroglobulin (Tg) level, Tg Ab, TPO Ab
Presence of lymph nodes: cervical, submandibular	+/- chest x-ray	Complete blood count with differential
	+/- CT: neck with contrast, chest without contrast	+/- calcitonin
		+/- comprehensive metabolic panel, uric acid, lactate dehydrogenase

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When to biopsy:

- Large nodule (greater than 3 cm)

OR

- Any concerning characteristics:
  - Micro-calcifications
  - Hypoechoic
  - Solid
  - Absent/incomplete halo
  - Internal vascular flow
  - Irregular margins
  - Evidence of invasion

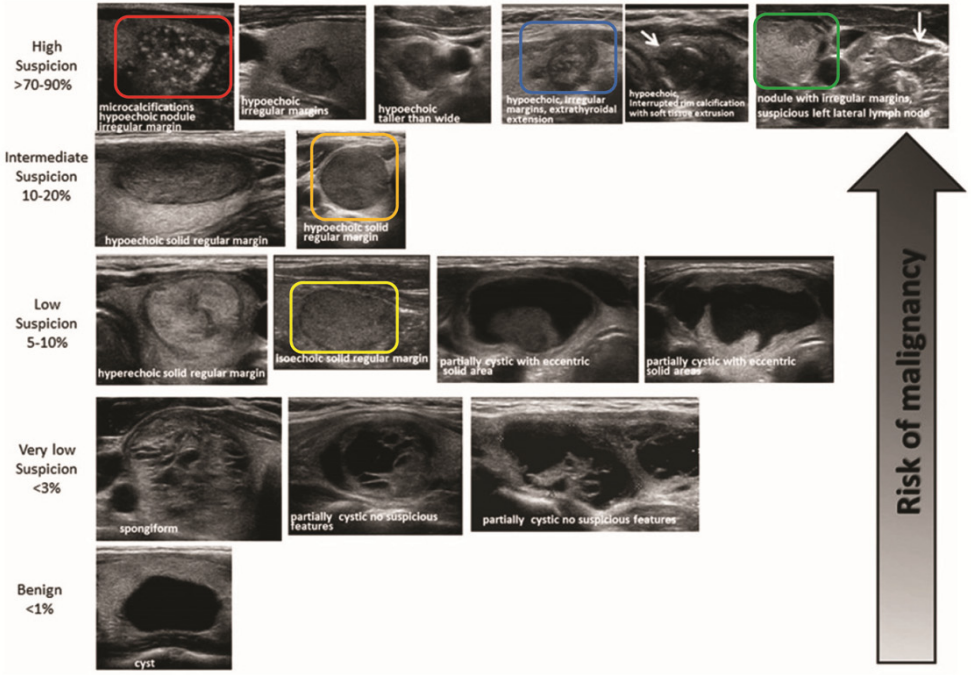

FIG. 2. ATA nodule sonographic patterns and risk of malignancy.

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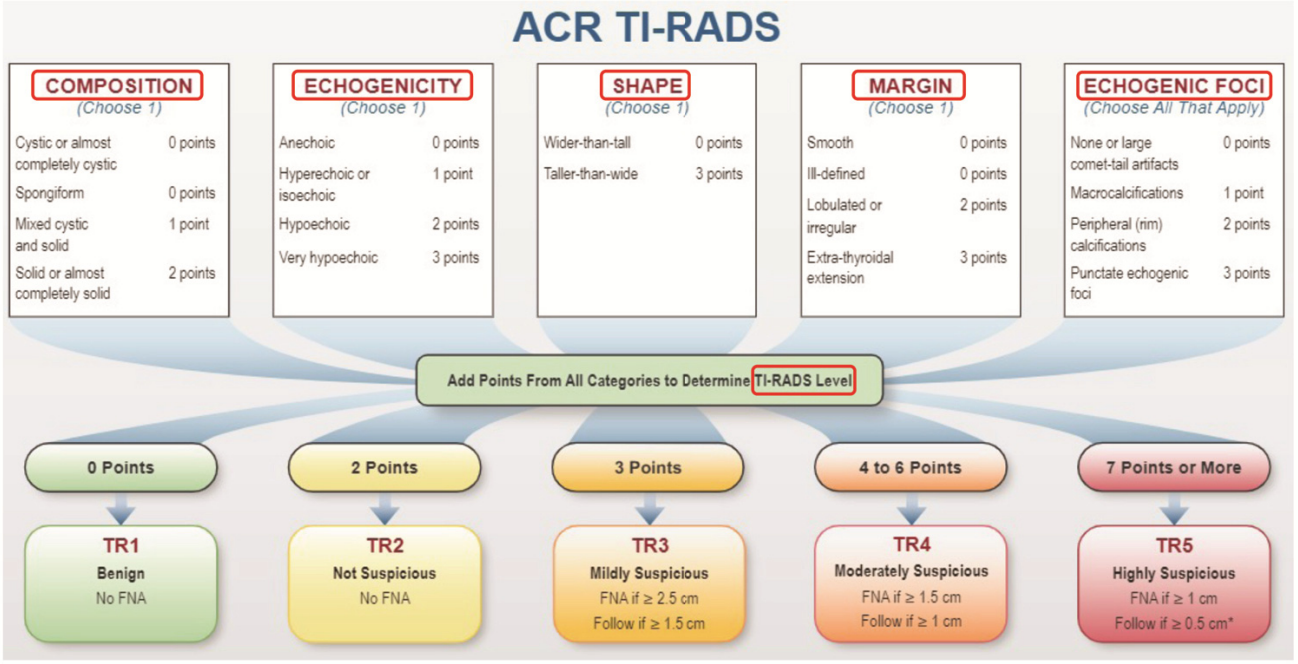
American College of Radiology: Thyroid Imaging Reporting And Data System



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Proposed PED TI-RADS:

Comparison of the ACR and PED TI-RADS Recommendations for FNA per TI-RADS Level [9].

Recommendations for FNA per TI-RADS Level		ACR TI-RADS	PED TI-RADS
TR1	Benign	*No FNA*	*No FNA*
TR2	Not Suspicious	*No FNA*	*No FNA*
TR3	Mildly Suspicious	*FNA if ≥ 2.5 cm* *Follow if ≥ 1.5 cm*	*FNA if ≥ 1.5 cm*
TR4	Moderately Suspicious	*FNA if ≥ 1.5 cm* *Follow if ≥ 1.0 cm*	*FNA if ≥ 1.0 cm*
TR5	Highly Suspicious	*FNA if ≥ 1.0 cm* *Follow if ≥ 0.5 cm*	*FNA for any technically feasible size*

Richman, 2020.

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Comparison of systems:

Comparison of criteria in identifying malignant surgical pathology. 95% Confidence Interval in parenthesis. (all $p > 0.05$).

	ATA	TI-RADS	PEDTI-RADS	TI-RADS ≥ 3	TI-RADS ≥ 4
Sensitivity	84.62% (54.55–98.08)	76.92% (46.19–94.96)	84.62% (54.55–98.08)	100.00% (75.29–100.00)	69.23% (38.57–90.91)
Specificity	9.52% (1.17–30.38)	71.43% (47.82–88.72)	52.38% (29.78–74.29)	42.86% (21.82–65.98)	76.19% (52.83–91.78)
Accuracy	38.24% (22.17–56.44)	73.53% (55.64–87.12)	64.71% (46.49–80.25)	64.71% (46.49–80.25)	73.53% (55.64–87.12)
Positive Predictive Value	36.67% (30.65–43.13)	62.50% (44.32–77.73)	52.38% (39.90–64.57)	52.00% (42.79–61.07)	64.29% (43.57–80.76)
Negative Predictive Value	50.00% (13.78–86.22)	83.33% (64.12–93.33)	84.62% (59.06–95.45)	100.00%	83">80.00% (63.10–90.34)
# FNA recommended ($n = 138$)	114	32	64	94	43
Missed Suspicious or Malignant Cytology ($n = 10$)	1	3	1	0	1
Missed Malignant Pathology ($n = 13$)	2	3	2	0	4

Ahmad H, 2021.

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Bethesda Criteria for Thyroid Cytopathology:

Category	Meaning
I	Non-diagnostic or inadequate
II	Benign
III	Atypia/follicular lesion of undetermined significance
IV	Follicular neoplasm or suspicious for follicular neoplasm
V	Suspicious for malignancy
VI	Malignant

Source: Cibas et al.[11]

Differentiated thyroid cancer – risk factors:

- Childhood cancer survivors treated with **radiation therapy**
  - Hodgkin lymphoma, leukemia, and CNS tumors
  - Thyroid nodules develop at a rate ~2 % annually with peak incidence 15-25 years after exposure
  - Risk is greatest among those who received **radiation therapy at a younger age** (before age 5) and with **doses up to 20-29 Gy**
- Genetic disorders
  - APC-Associated Polyposis (PTC)
  - Carney complex (PTC, FTC)
  - DICER1 mutation (PTC, FTC)
  - PTEN Hamartoma Tumor Syndrome (PTC, FTC)

Huang, 2018.

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Thyroid cancer guidelines – adults:

- American Thyroid Association (ATA) – 2006, 2016
- European Society for Medical Oncology (ESMO) – 2019

AMERICAN THYROID ASSOCIATION
Optimal Thyroid Health for All

ESMO
GOOD SCIENCE
BETTER MEDICINE
BEST PRACTICE

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Management Guidelines for Children with Thyroid Nodules and Differentiated Thyroid Cancer

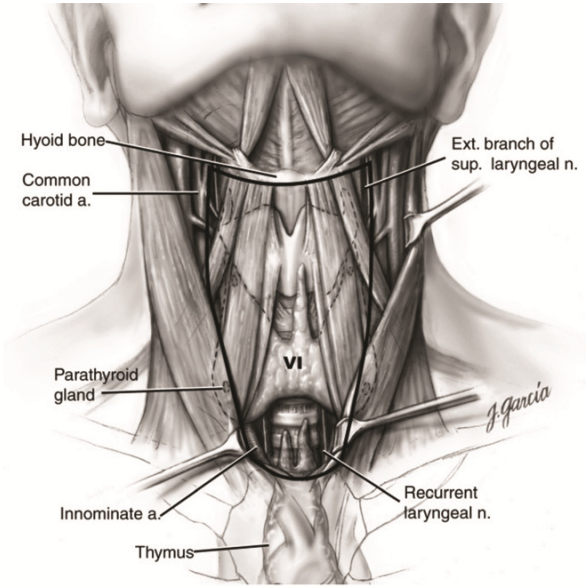
The American Thyroid Association Guidelines Task Force on Pediatric Thyroid Cancer

Gary L. Francis, M.D., Ph.D.*[1], Steven G. Waguespack, M.D.*[2], Andrew J. Bauer, M.D.*[3], Peter Angelos, M.D., Ph.D.[4], Salvatore Benvenga, M.D.[5], Janete M. Cerutti, Ph.D.[6], Catherine A Dinauer, M.D.[7], Jill Hamilton, M.D.[8], Ian D. Hay, M.D., Ph.D.[9], Markus Luster, M.D.[10], Marguerite T. Parisi, M.D., M.S. Ed.[11], Marianna Rachmiel, M.D.[12], Geoffrey B. Thompson, M.D.[13], and Shunichi Yamashita, M.D., Ph.D.[14]

2015: Created in response to good prognosis seen in children with thyroid cancer who were developing significant morbidity due to over-treatment.

Treatment - surgery:

- **Total thyroidectomy (TT) is recommended as initial treatment.**
  - If small unilateral tumor within thyroid gland, consider near-TT
  - Some indications for lobectomy.
    - Unifocal tumors without positive lymph nodes have lower likelihood of contralateral disease (16%).
  - May be some use of systemic agents prior to surgery in some cases of advanced disease



Cherella C, 2021.

Carty S, 2009.

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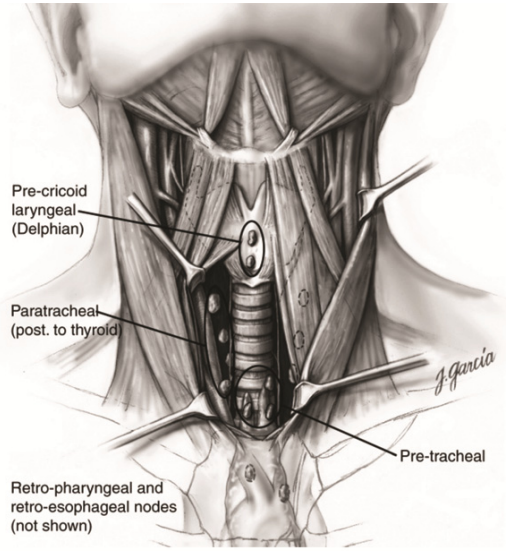
Treatment - surgery:

- +/- **central neck dissection** - prelaryngeal, pretracheal, and paratracheal lymph nodes
  - If there is extrathyroidal invasion and/or loco-regional metastasis
  - Comprehensive and compartment-based lymph node dissection
  - Ipsilateral vs bilateral
- +/- **lateral neck dissection** – levels II, III, IV, anterior V
  - If there is cytologic evidence of metastases
  - Tg in FNA washout of lymph node biopsy



Carty S, 2009.

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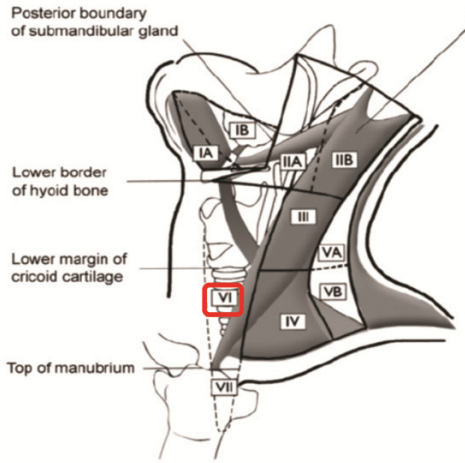
TABLE 5. AMERICAN JOINT COMMITTEE ON CANCER TNM CLASSIFICATION SYSTEM FOR DIFFERENTIATED THYROID CARCINOMA[a]

Primary tumor (T)		
TX		Size not assessed, limited to the thyroid
T1	T1a	≤ 1 cm, limited to the thyroid
	T1b	> 1 cm but ≤ 2 cm, limited to the thyroid
T2		> 2 cm but ≤ 4 cm, limited to the thyroid
T3		> 4 cm, limited to the thyroid, or any tumor with minimal extrathyroid extension
T4	T4a	Tumor extends beyond the thyroid capsule to invade subcutaneous soft tissues, larynx, trachea, esophagus, or recurrent laryngeal nerve
	T4b	Tumor invades prevertebral fascia or encases carotid artery or mediastinal vessels
Lymph nodes (N)		
NX		Regional lymph nodes not assessed
N0		No regional lymph node metastasis
N1	N1a	Metastasis to level VI (pretracheal, paratracheal, and prelaryngeal/ Delphian lymph nodes)
	N1b	Metastasis to unilateral, bilateral, or contralateral cervical levels I, II, III, IV, or V) or retropharyngeal or superior mediastinal lymph nodes (level VII)
Distant metastasis (M)		
MX		Distant metastasis not assessed
M0		No distant metastasis
M1		Distant metastasis

[a]Pediatric patients are considered to have stage II disease if distant metastases are identified (M
considered to have stage I disease.

TNM staging:

PRIMARY TUMOR	LYMPH NODES	DISTANT METASTASIS
T 1-4	N 0-1 N1a: level VI N1b: levels I-V, VII	M 0-1



Sternocleidomastoid muscle

I. Level
submental & submandibular
II. Level - upper jugular
(skull base - hyoid)
III. Level - middle jugular
(Hyoid - cricoid)
IV. Level - lower jugular
(cricoid - clavicle)
V. Level - posterior triangle
above cricoid
cricoid - clavicle
VI. Level - upper visceral
praelaryngeal, prae-, paratracheal
VII. Level - upper mediastinal

Other N groups:
suboccipital, retropharyngeal, parapharyngeal, buccinator, preauricular, peri-, intraparotid

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TABLE 6 ATA Pediatric Thyroid Cancer Risk Levels and Postoperative Management in Children with Papillary Thyroid Carcinoma

ATA Risk Level[1]	Definition	Initial Postoperative Staging[2]	TSH Goal[3]	Surveillance of Patients With No Evidence of Disease[4]
ATA Pediatric Low-Risk	Disease grossly confined to the thyroid with N0/Nx disease or patients with incidental N1a disease (microscopic metastasis to a small number of central neck lymph nodes)	Tg^5	0.5 – 1.0 mIU/L	US at 6 months postoperatively and then annually x 5 years Tg^5 on LT4 every 3-6 months for two years and then annually
ATA Pediatric Intermediate-Risk	Extensive N1a or minimal N1b disease	TSH-Stimulated Tg^5 and diagnostic ^{123}I scan in most patients (See Figure 2)	0.1 – 0.5 mIU/L	US at 6 months postoperatively, every 6-12 months for 5 years, and then less frequently Tg^5 on LT4 every 3-6 months for 3 years and then annually Consider TSH-stimulated Tg^5 ± diagnostic ^{123}I scan in 1-2 years in patients treated with ^{131}I
ATA Pediatric High-Risk	Regionally extensive disease (extensive N1b) or locally invasive disease (T4 tumors), with or without distant metastasis	TSH-Stimulated Tg^5 and diagnostic ^{123}I scan in all patients (See Figure 2)	< 0.1 mIU/L	US at 6 months postoperatively, every 6-12 months for 5 years, and then less frequently Tg^5 on LT4 every 3-6 months for 3 years and then annually TSH-stimulated Tg^5 ± diagnostic ^{123}I scan in 1-2 years in patients treated with ^{131}I

Please refer to Table 5 for AJCC TNM classification system.
FTC=follicular thyroid carcinoma; Tg=thyroglobulin; TSH=thyroid stimulating hormone; US=ultrasound

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Post-operative staging:

- Within 12 weeks after surgery
- ATA Pediatric Low-Risk patients: non-stimulated thyroglobulin
- ATA Pediatric Intermediate- and High-Risk patients:
  - Stimulated thyroglobulin
  - Diagnostic whole body scan
    - Hybrid imaging with single photon emission computed tomography (SPECT/CT)
      - Can help differentiate remnant thyroid tissue from lymph node metastasis

Treatment – radioactive iodine (RAI):

- I-123 iodine uptake scan
- Preparation
  - Goal TSH >30 mIU/L
  - Most use withdrawal: ≧14 days off levothyroxine
  - Some use of rhTSH (thyrogen)
  - Low iodine diet for 2 weeks prior
- +/- I-131 radioactive iodine ablation
  - For persistent locoregional or nodal disease that cannot be resected
  - For distant metastases (pulmonary, etc)
  - Dosing: empiric vs whole body dosimetry
  - Post-treatment scan 4-7 days later

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Post-operative surveillance:

- Monitor post-operatively:
  - Tumor markers: thyroglobulin (Tg) and Tg Ab
  - Neck ultrasound
  - +/- I-123 iodine uptake scan

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Summary:

- When to biopsy
  - Large nodule (>3 cm) OR
  - Any size with concerning ultrasound characteristics
- Papillary thyroid cancer: risk of recurrent disease is related to degree of lymph node metastases
- 2015 ATA Guidelines for Children with Thyroid Nodules and Cancer – update coming soon

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