

# Recommendations in Prenatal Screening in the World and Connections to Other Diseases Like Thyreopathy



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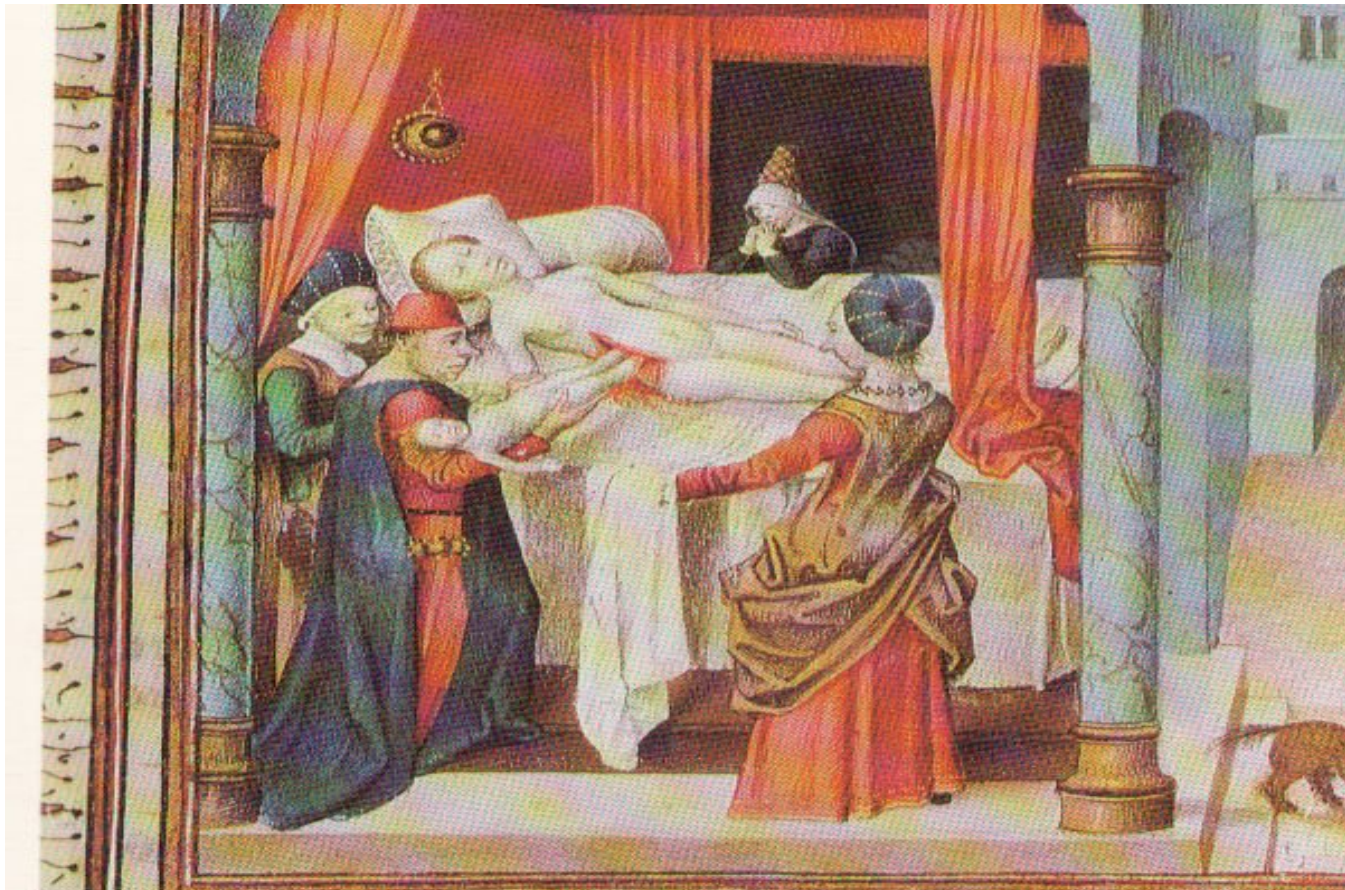


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# Aim of maternal-fetal care

- the uncomplicated birth of a healthy baby to a healthy mother at term



Paulus Orosius, Histoire du monde, 1460 ?

# History of fetal-maternal care

- Diagnose post natal
- Early 20<sup>th</sup> centuries – institutionalize of handicapped person



Germany 1507

# Screening tests in pregnancy

- Risk for mother and fetus – curable
  - gestational diabetes
    - (new standard, screening between 24 and 28 WG)
  - infection (HIV, hepatitis and syphilis)
  - rhesus incompatibility
  - thyroid dysfunction

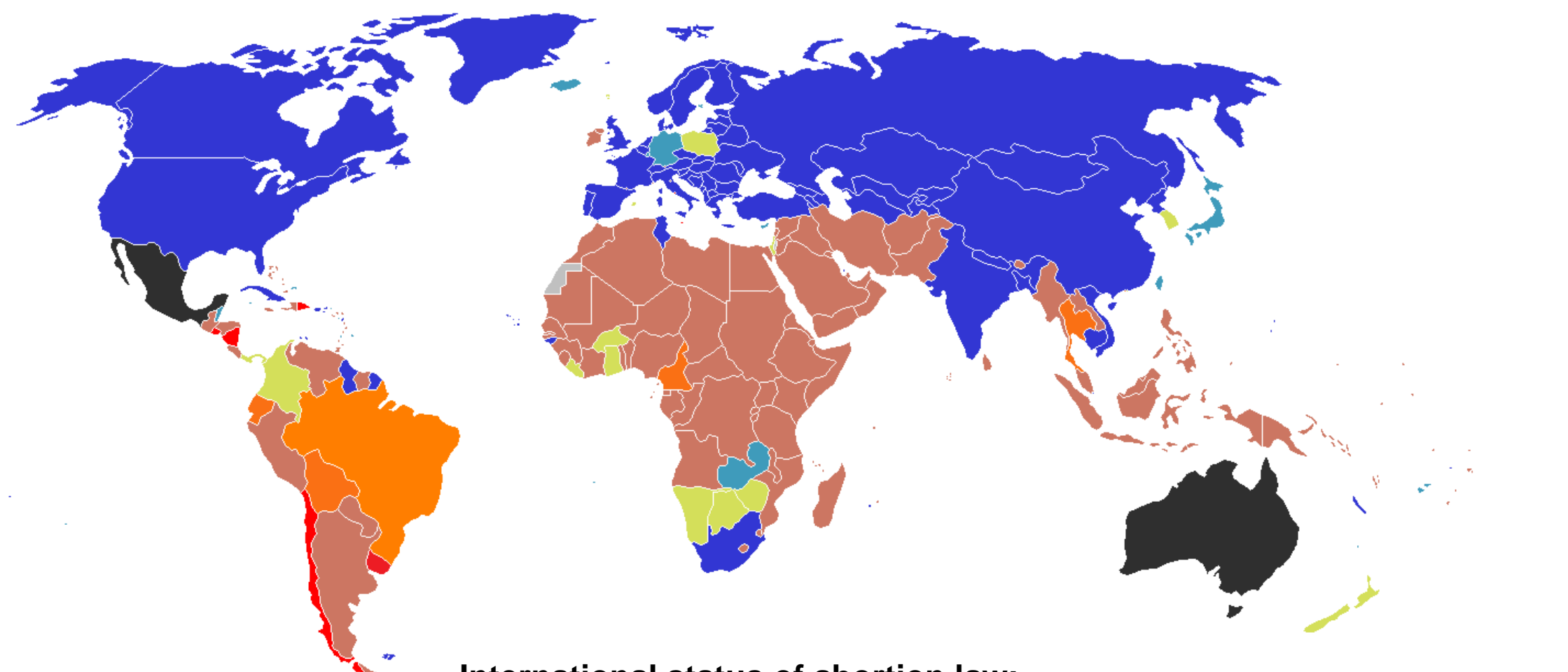
# Screening tests in pregnancy

- Screening of congenital defects
  - Biochemistry in 1<sup>st</sup> and 2<sup>nd</sup> trimester of pregnancy
  - UZ screening
- Diagnostics tests
  - Amniocentesis (AMC) 15<sup>th</sup> – 18<sup>th</sup> week of pregnancy
    - karyotyping
    - molecular biology techniques
  - Chorionic Villus Sampling (CVS) 11<sup>th</sup> - 13<sup>th</sup> week of pregnancy
    - sample removed from developing placenta

# Screening and law

- From 1998, among the 152 most populous countries, 54 either banned abortion entirely or permitted it only to save the life of the pregnant woman.
- In contrast, another 44 of them generally banned late-term abortions after a particular gestation age: 12 weeks (Albania, Cuba, Czech Republic, Denmark, France, Russia, South Africa, Ukraine, Tunisia, Turkey....), 13 weeks (Italy), 14 weeks (Austria, Cambodia, Germany...), 18 weeks (Sweden), viability (Netherlands and to some extent the United States), and 24 weeks (Singapore and the United Kingdom [Northern Ireland excluded]).





### International status of abortion law:

- Legal on request
- Illegal with exception for rape, maternal life, health, mental health, fetal defects, and/or socioeconomic factors
- Illegal with exception for rape, maternal life, health, mental health, and/or fetal defects
- Illegal with exception for rape, maternal life, health, and/or mental health
- Illegal with exception for maternal life, health, and/or mental health
- Illegal with no exceptions
- Varies by region
- No information

# Screening and Ethics

- The Hippocratic Oath



- Principles
- Autonomy – being in control of one's own body
- Beneficence – doing good
- Non-maleficence – doing no harm  
("primum non nocere")
- Justice – being fair and equitable



# WMA International Code of Medical Ethics

A physician shall

- respect a competent patient's right to accept or refuse treatment.
- be dedicated to providing competent medical service in full professional and moral independence, with compassion and respect for human dignity
- strive to use health care resources in the best way to benefit patients and their community
- always bear in mind the obligation to respect human life.
- act in the patient's best interest when providing medical care

*Composed 1949, amended 1968, 1983 and 2006*

# Ethical considerations

- Who is the patient?
- Who benefits?
- Highly stressful - patients and staff
- Stigmatisation of surviving Down's patients
- Can appropriate counselling be provided?



Alexandria - 3rd century BC

# Prenatal screening history

- 1866 : First description of Down Syndrom (John Langdon Down)
- 1930 - Down syndrome – maternal age association
- 1966 : First karyotype on amniotic cells culture
- 1974 : First fetal ultrasound scan in France
- 1980 - 2<sup>nd</sup> trimester AFP (*with maternal age*)
  - 2<sup>nd</sup> trimester Multiple markers (double, triple, quad)
- 1990 - 1<sup>st</sup> trimester nuchal translucency (NT)
  - 1<sup>st</sup> trimester NT + PAPP-A + free  $\beta$ hCG
- 2000 -Integrated 1<sup>st</sup> and 2<sup>nd</sup> trimester
  - Sequential 1<sup>st</sup> and 2<sup>nd</sup> trimester
- 2011 - Fetal nucleic acids in maternal plasma?

# Testing for Downs Syndrome

- indirect fetal testing, by biochemical maternal serum screening, was introduced
- maternal serum screening does not detect specific marker
- multiple biochemical markers used to calculate risk
- software packages available to calculate risk
- can screen in first or second trimester
- Last big studies – FASTER, SURUSS

# Screening of congenital development defects

## Screening of DS and NTD in the 2<sup>nd</sup> trimester of pregnancy

- hCG, AFP, uE3, Inhibin

## Screening of DS in the 1<sup>st</sup> trimester

- Free  $\beta$  hCG, PAPP-A
- Nuchal translucency – NT, other US markers

## Integrated test

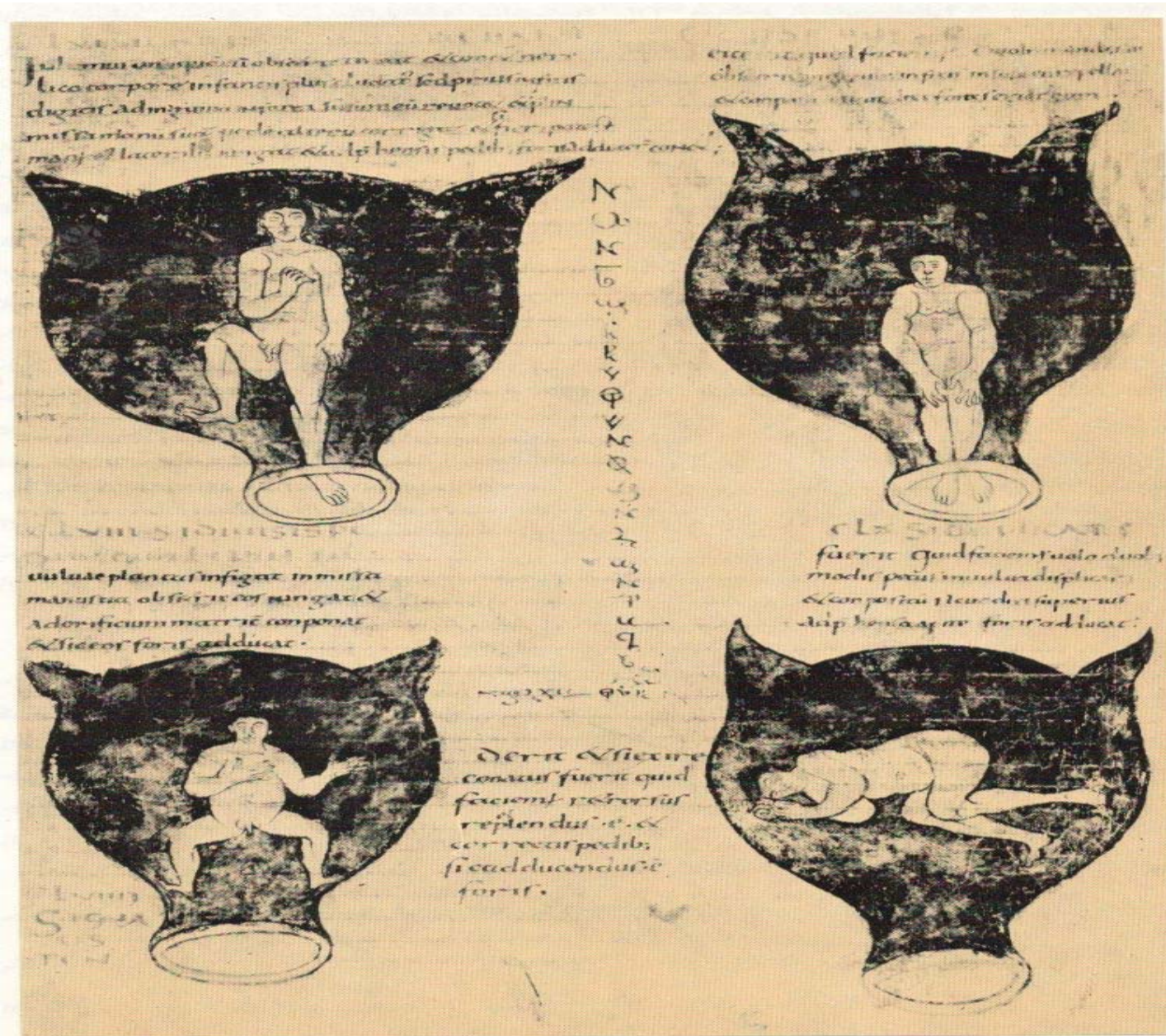
- Common evaluation results from 1<sup>st</sup> and 2<sup>nd</sup> trimester
- **Serum integrated test** – only serum markers, US not needed
  - Free  $\beta$  hCG, PAPP-A
  - hCG, AFP, uE3, Inhibin



# Markers of screening

	1 <sup>st</sup> trimester	2 <sup>nd</sup> trimester	Integrated test	Serum integrated test
<b>PAPP-A</b>				
<b>free <math>\beta</math> hCG</b>				
<b>Nuchal translucency</b>				
<b>AFP</b>				
<b>hCG</b>				
<b>inhibin</b>				
<b>uE3</b>				
<b>Efficacy</b>	<b>75-80%</b>	<b>65%</b>	<b>90-93%</b>	<b>75-80%</b>

## Position of fetus in uterus According Sorano (2nd century AC) from manuscript - 9th century



# Prenatal diagnosis DS 2004-2008

Region	Total Cases	Prenatally Diagnosed	% of Total Cases
France (Paris)	551	477	87%
Italy (Tuscan)	256	198	77%
Malta	38	1	3%
Spain (Barcelona)	167	144	86%
Switzerland (Vaud)	148	133	90%
UK (Thames Valley)	322	203	63%
Poland	1776	166	9%
Ukraine	147	24	16%

# Prenatal diagnosis DS 2004-2008

Region	Total Cases
France (Paris)	Since 2010 1, st TM screening, > 38 – AMC without biochemistry
Italy (Tuscan)	1, st TM screening or 2nd TM screening, vary by region
Malta	Termination is nonlegal, NT or AMC is not performed
Spain (Barcelona)	1st TM screening or 2nd TM screening , health insurance
Switzerland (Vaud)	1st TM screening and 2nd TM screening , in case of 1st TM screening is recommended AFP in 2nd TM
UK (Thames Valley)	1, st TM screening or 2nd TM screening (Scotland), vary by country
Poland	Illegal with exception for rape, maternal life or health
Ukraine	since 2009 obligatory screening

Eurocat website database: <http://www.eurocat-network.eu>

## *UK NSC Policy recommendations*

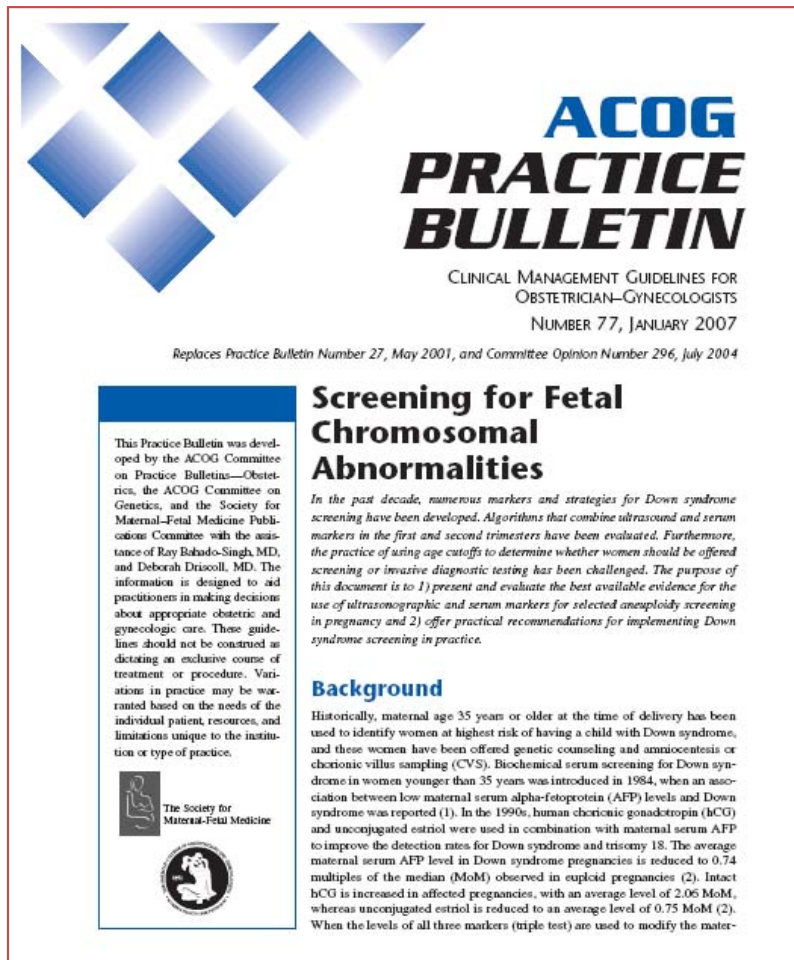
- by April 2010: A detection rate (DR) of greater than 90% of affected pregnancies with a screen positive rate (SPR) of less than 2%.
- Cut-off:
  - 1 in 200 at term for 2nd trimester screening strategies
  - 1 in 150 at term for 1st trimester screening strategies.
- 1st trimester – Combined testing. This is the preferred method to aid early diagnosis
- 1st and 2nd trimester – Integrated testing. This test requires the woman to attend at least twice for screening
- 1st and 2nd trimester – Serum Integrated testing – This test does not include ultrasound NT
- 2nd trimester – Quadruple testing – This test is required for those women who attend later in the pregnancy



# USA

## The National Academy of Clinical Biochemistry (NACB)

- Maternal and Fetal Health Risk Assessment
- Laboratory Medicine Practice Guidelines



<http://www.nacb.org>



Procedure	Detection rate	Reported rate	Uptake	Process time (wk)
<b>1st TM screening</b>				
Maternal age	32%		80%	0
NT	74%	73%	80%	0
1st TM double test (PAPP-A, hCG)	63%	62%	80%	1
NT, PAPP-A, hCG	86%	80–85%	80%	1
<b>2nd TM screening</b>				
Maternal age	32%		80%	0
2nd TM double test (AFP, hCG)	60%	58–59%	80%	1
Triple test (AFP, hCG, uE3)	68%	67–69%	80%	1
Quadruple test (AFP, hCG, uE3, inhibin A)	79%	76–79%	80%	1
Integrated test (1st TM: NT, PAPP-A; ) 2nd TM: quadruple test	95%	94%	80%	1

# US GUIDELINE 9: First trimester testing

- Maternal age-based screening should no longer be accepted as indication for invasive prenatal diagnosis
- Integrated, age-based, nuchal translucency and biochemical screening should be used to detect aneuploidy.
- Integrated first-trimester screen or a two-step first (nuchal translucency, PAPP-A, and  $\beta$ hCG) and second (quadruple biochemistry) trimester screen.
- Nuchal translucency should be introduced only after appropriate training and certification, with access to the integrated computer programs, and continuing quality assurance.

# US GUIDELINE 10: Screening assays

- **Alpha-Fetoprotein [AFP] Assay and Chorionic Gonadotropin [hCG] Assay** - The coefficient of variation should not exceed 5%. The accuracy should be within 3% from lot to lot.
- **Unconjugated Estriol [uE3]** - The coefficient of variation should not exceed 7%. The accuracy should be within 5% from lot to lot.
- Since uE3 is not stable on storage at room temperature, programs should monitor time from specimen collection to analysis and reject specimens that are old enough to exhibit deterioration.

# Prenatal Screening for Fetal Aneuploidy in Canada

- First trimester screen
- Second trimester quad screen
- Two-step screens
  - Contingent
  - Integrated
  - Serum integrated
  - Sequential

# **A Guideline developed by the Human Genetic Society of Australasia and the Royal Australian and New Zealand College of Obstetricians and Gynaecologists**

- Prenatal screening tests for trisomy 21 (Down syndrome), trisomy 18 (Edwards syndrome) and neural tube defects
  - Combined first trimester screening
  - Maternal serum screening
  - Integrated Screening – **no!**

# **Prenatal genetic screening and testing in Singapore**

- Prenatal screening should be offered universally to all women who desire to know the health status of the child they bear.
- There are many modes of screening



# Jewish community



Anonymous carrier testing done prior to engagement alleviates the above concerns

Within the Jewish community, a system of premarital genetic testing for a number of conditions has been established

This project is called Dor Yesharim, The Committee for Prevention of Jewish Genetic Diseases

The couple is not told what disease they carry, just that the match is likely to lead to a child affected by one of these conditions

As this system takes the halachic concerns into consideration, it has the approval of many leading halachic authorities It is also subsidized

# Summary I

- **Down's Syndrome screening feasible**
- **Biochemical testing still has major role in Down's screening**
- **Current aim is for first trimester screening but many practical problems associated with this**
  - **certification, high level of cooperation (one-day service)**
- **Increased standardisation is imminent**
- **Biochemical testing for NTD is being superseded by US**
- **Ethical issues cannot be ignored**
- **Informed decision making essential**



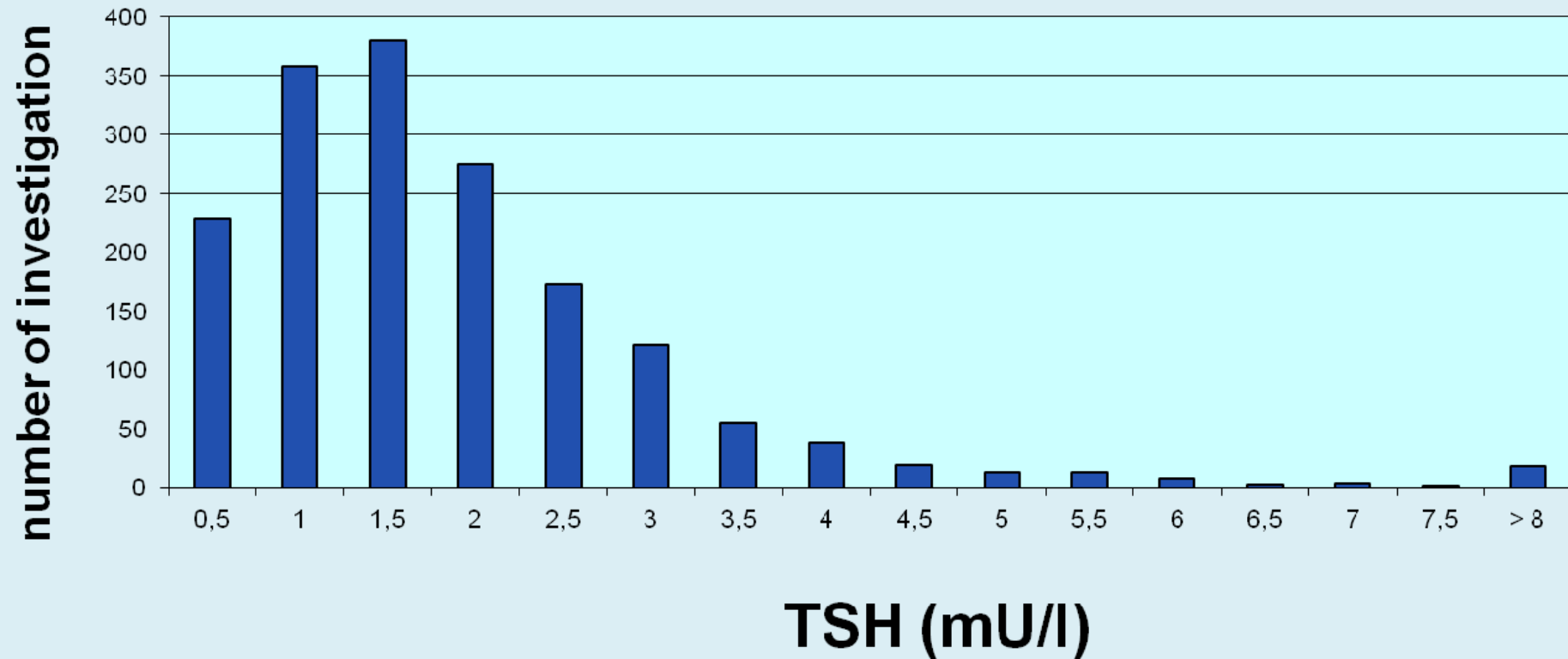
# Thyroid dysfunction

- Some diseases of the thyroid gland can affect both the pregnant woman and the fetus
- Maternal hypothyroxinemia results in the birth of children with decreased mental and psychomotor development
- Iodine status – important and regional differences
- Shift of TSH reference interval
- Incompatible cut-off TPO Ab

# Study group

- **7,530 pregnant women** (9<sup>th</sup> – 11<sup>th</sup> week of pregnancy, 99% Caucasian)
  - undergoing their first trimester prenatal screening
- The average age was 31.3 (+/- 4.6) years.
- Serum were assayed for TSH, FT4 and anti-TPO
- Lower serum TSH in pregnancy is influenced by the thyrotropic activity of elevated hCG concentrations, mainly in the first trimester

# TSH in 1<sup>th</sup> trimester of pregnancy

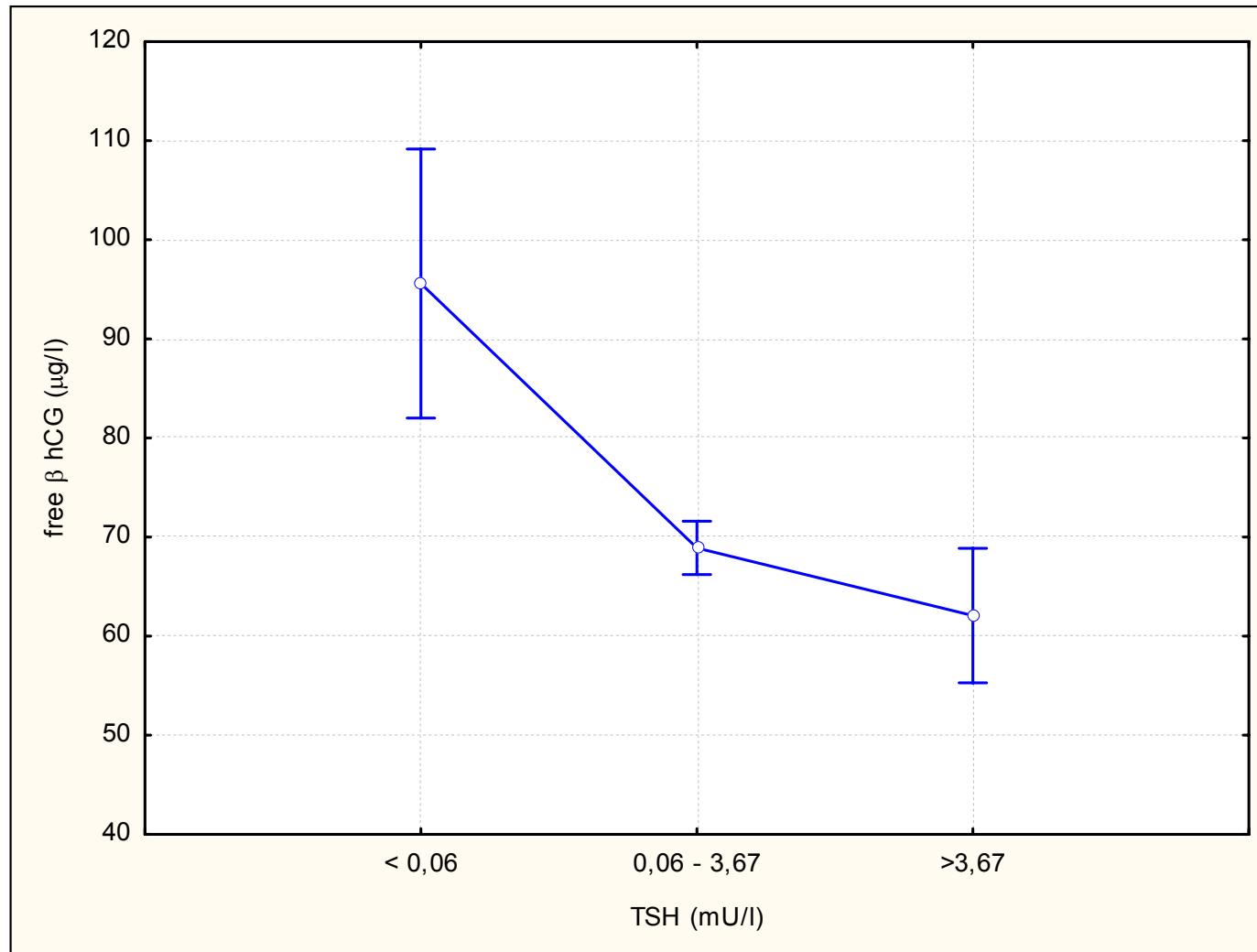




# TSH versus hCG

- In group of pregnant women with suppressed TSH the average level of free  $\beta$  hCG was almost double (M=95.6 mg/ml) in comparison with group with TSH in reference range (M=68.9 mg/ml) or with TSH >3.67mU/l (M=62.1 mg/ml).
- Differences between the normal and raised TSH groups in free  $\beta$  hCG levels were not significant at  $p<0.05$ .

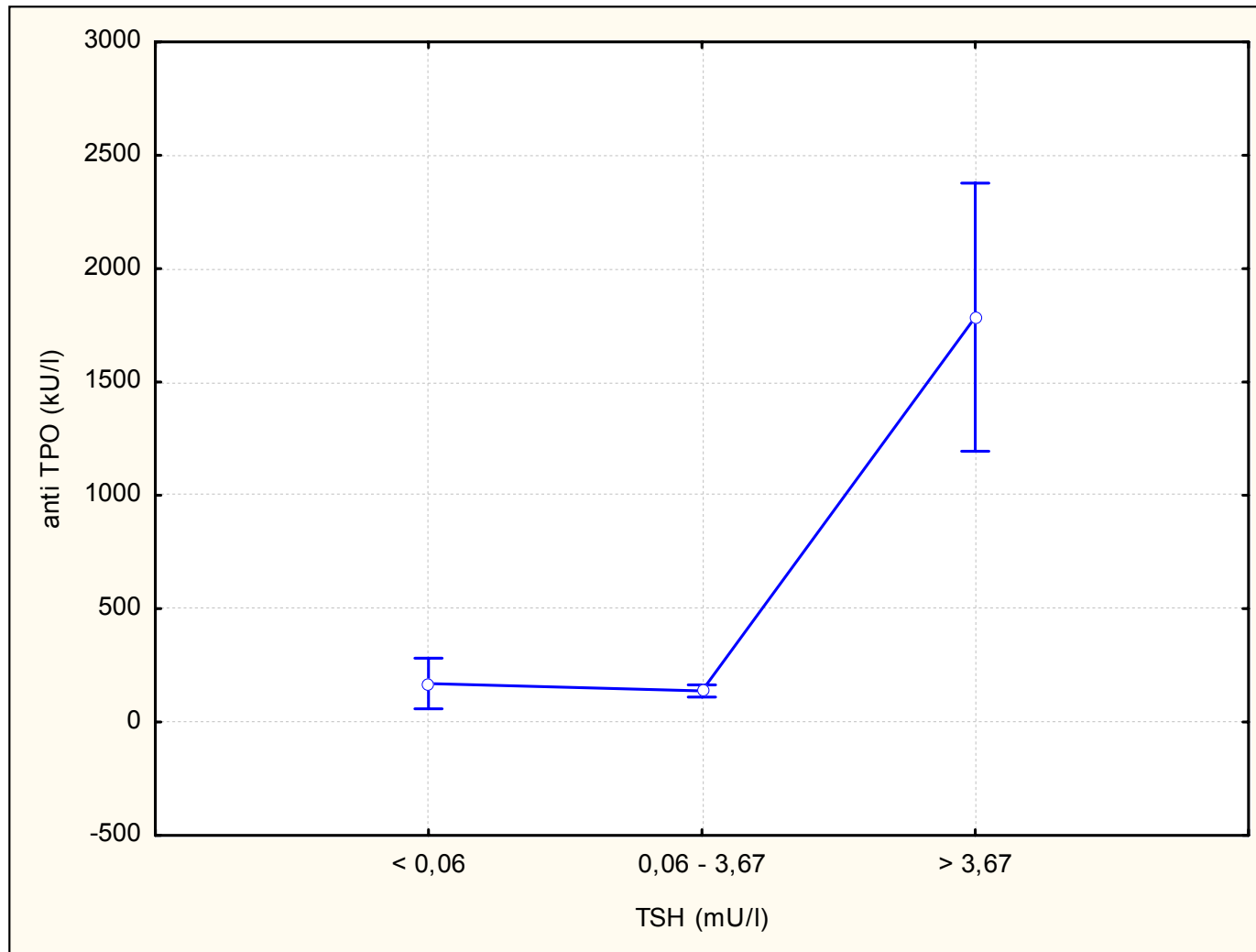
# Free $\beta$ hCG in groups with different TSH level



# TSH versus anti TPO

- The relationship between anti-TPO and TSH is not definite, despite it being known that women with high level of TSH more frequently have positive anti-TPO antibodies
- In the group with TSH  $>3.67$  mU/l were 44.1% anti-TPO positive women
- In the group with TSH  $< 0.06$  mU/l resp. TSH in the reference interval were positivity in 9.15%

# Anti TPO level with different TSH level

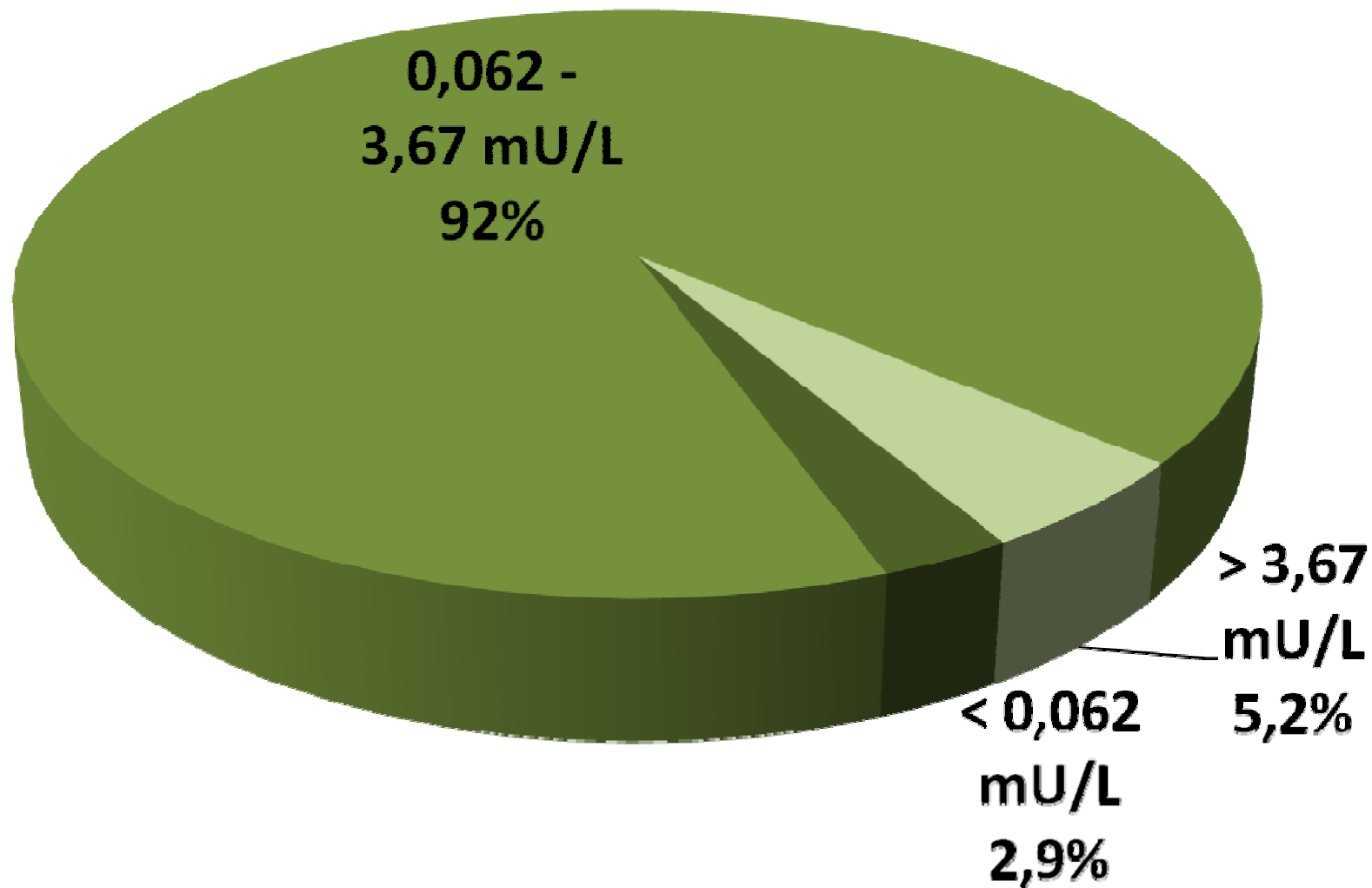


# TSH and FT4 reference interval

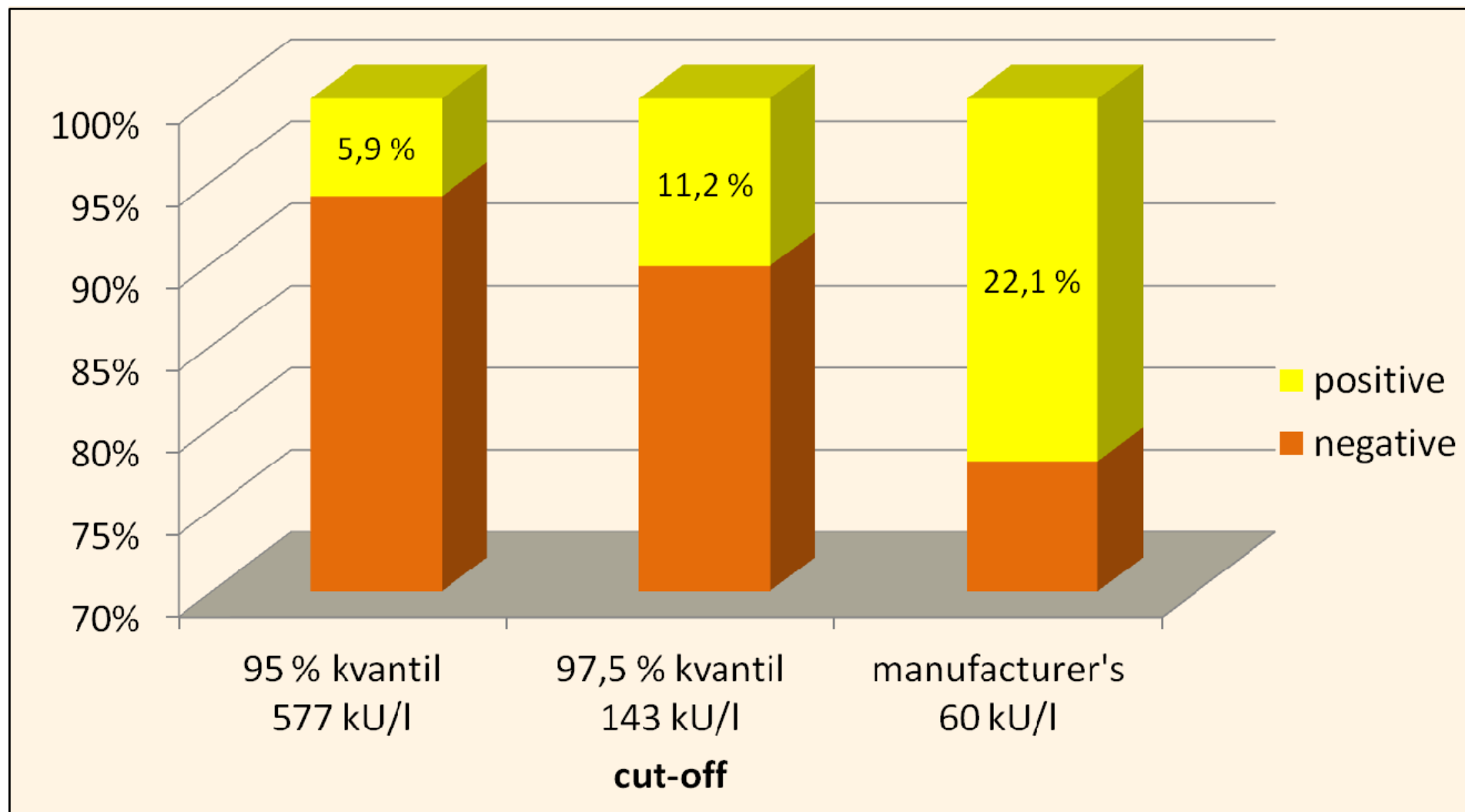
The 1<sup>st</sup> trimester, the specific reference range was used the 2.5<sup>th</sup> and 97.5<sup>th</sup> percentile of log transformed data of the pregnant women group without anti-TPO positive and woman with hCG > triple of the median.

FT4 reference interval were derived using nonparametric analyses such as the 95<sup>th</sup> percentile. The calculated reference interval 9.8 - 23.43 pmol/l was very similar to the manufacturer's interval of 9.8 - 23.0 pmol/l.

# TSH reference interval and positivity in pregnancy



# Selection cut off for anti TPO antibodies



# Anti –TPO Ab cut off

For anti TPO was calculated the cut off at the 90<sup>th</sup> percentile from the group of pregnant women with TSH level in new established pregnancy specific range of 0.06 – 3.67 mU/l.

The anti-TPO positivity cut-off was established to 143 kU/l.

The positivity of anti-TPO in nonpregnant individuals is about 11%; in the pregnant population it is very similar.

When was used the reference interval recommended by the producer of reagents ( $> 60$  kU/l), there was 22.1% positivity.

If was used the 90<sup>th</sup> percentile (143 kU/l) as the cut-off for our group of pregnant women, there was 11.5% positivity



# Reference intervals and cut-off

Analyte	Reference interval Producer	1 <sup>st</sup> trimester of pregnancy
TSH	0,37 – 5,0 mIU/l	0,06 – 3,67 mU/l
FT4	9,8 – 23,1 pmol/l	9,80 – 23,43 pmol/l
anti-TPO	< 60 kU/l	< 143 kU/l

Analytical system - ADVIA® Centaur™ Siemens automated random-access immunoassay

**Now, we done the reference interval for 7 analytical systems – in press**

Springer D, Zima T, Limanová Z: *European Journal of Endocrinology* 2009 **160** 791-7.

# Summary II

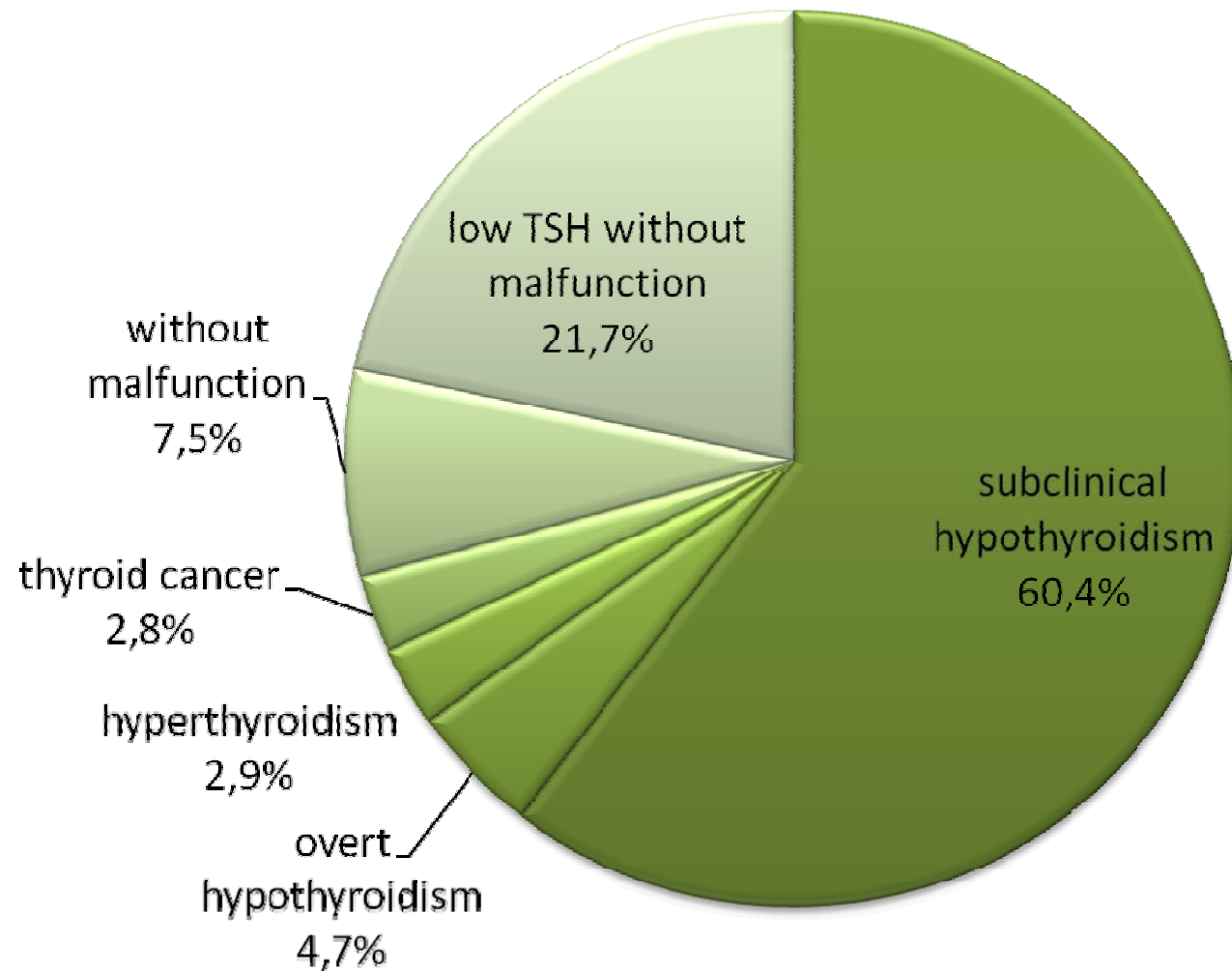
In the group of 7, 350 women, 213 (2.90%) had their TSH under the reference interval ( $<0.06$  mU/l).

The prevalence of hyperthyreosis in pregnant women is about 1.7%, and 0.4% of these women had an elevated serum FT4 level. This is similar to that reported for non-pregnant individuals.

Many authors have determined the prevalence of hypothyreodism (overt and subclinical) in pregnancy and it is estimated to be 0.3 - 0.5% for overt hypothyroidism and 2 - 3% for subclinical hypothyroidism.

In this study there were 4.5% of pregnant women with TSH over reference interval ( $>3.67$  mU/l)

# Structure of diagnosis in group of positive women



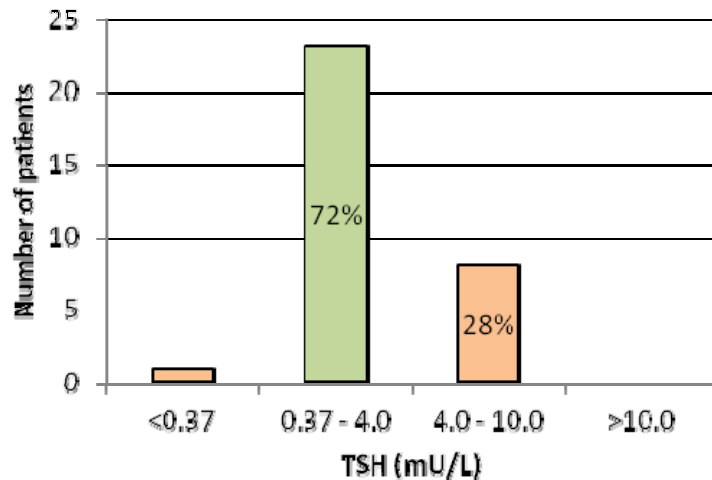
## Thyroid function after delivery in positive screened pregnant women

We invited 821 women who were positively screened for thyroid disorders (TSH, FT4 and TPOAb) in 9th – 11th week of pregnancy (in years 2006-2009, mean age 31 years) for follow-up one to three years after delivery (median 15 months; min. 2, max. 38) months.

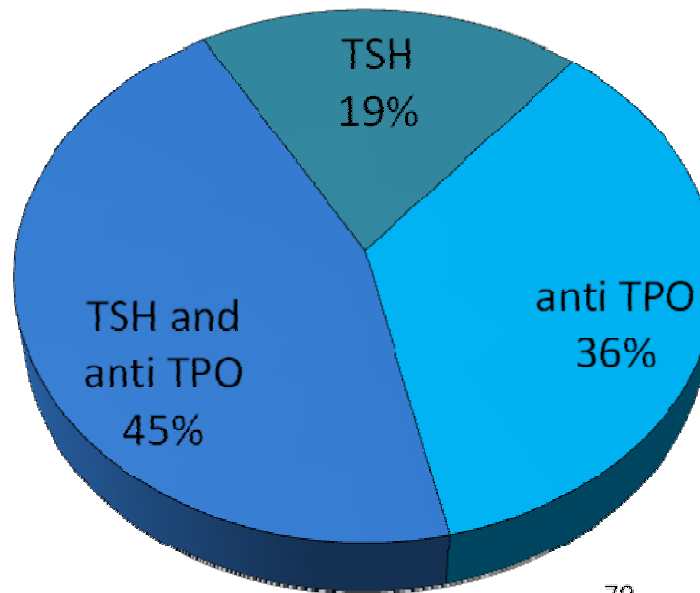
Family history of thyroid disease, personal history of diabetes or previous treatment for thyroid disease were present only in 58 % of the positively screened pregnant women.

Only 67,5 % of all women had normal delivery

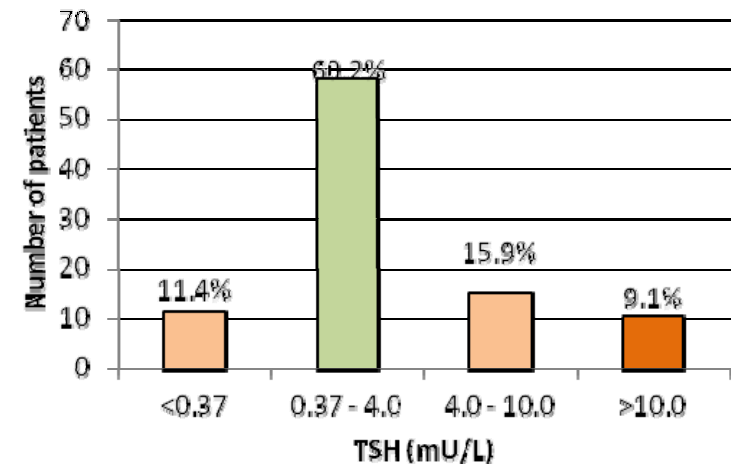
# Positivity of TSH and anti TPO



TSH values of the initially hypothyroid and subclinically hypothyroid anti TPO positive women at follow up



TSH values of the initially euthyroid anti TPO positive women at follow up



# Thyroid dysfunction

- World guidelines for management of thyroid dysfunction during pregnancy and postpartum recommend not universal but only case finding screening

Abalovich M, et al. An Endocrine Society Clinical Practice Guideline. *Journal of Clinical Endocrinology & Metabolism* 2007 **92** S1 – S47

- Vaidya (2007) study shows that targeted thyroid function testing of only high-risk pregnant women would miss nearly one-third of women with overt/subclinical hypothyroidism during early pregnancy

Vaidya et al. *Journal of Clinical Endocrinology & Metabolism* 2007 **92** 203–207

# Thyroid dysfunction – Summary III

- 40% of the initially euthyroid pregnant women positive for TPOAb had a thyroid dysfunction more than one year after delivery
- Around 50 % of the positively screened pregnant women had a high-risk profile according to the medical history
- Our results support the implementation of not only general screening for ATD in pregnant women, but also a close follow-up for prolonged time period after the delivery
- **Screening of thyroid dysfunction during pregnancy is recommended**

# Thanks our collaborators and partners

- 3rd Dept. Of Internal Medicine
  - Prof. Límanová, dr. Potluková
- Dept. Of Gyneacology and Obstetric
  - Prof. Calda, dr. Belosovicova
- Team of lab
- Colleagues of working group – prenatal screening in Czech Republic
- Supports of grant of Ministry of Health, General Health Insurance Company Czech Republic, research project of Ministry of Education
- Pregnant women and mothers which coopereate on our project





# Delivery

Germany approx. 1530

