



ACCEPT

Establishment of a new Greenlandic Cohort Study
Adaption to Climate Change,
Environmental Pollution and Dietary Transition.

Danish - Greenlandic Research Cooperation
Aarhus University and Ilisimatusarfic, Nuuk

Eva Cecilie Bonefeld-Jørgensen
Professor in Human Toxicology
Centre for Arctic Health (CAH),
Department of Public Health,
Aarhus University





ACCEPT AIMS

- **Overall Aim**

- Establishment of a Greenlandic mother-child cohort being compatible with other circumpolar / international mother-child cohorts to investigate trends in health upon global changes in generally as well as global climate changes
- To establish a prospective research program to explore and identify relations between health outcomes, environmental contaminants, climate changes, lifestyle and genetic in Greenland
- To give a formalised and continued information service on environmental health issues to the local communities in Greenland (health professionals as well as the general population).
- To increase the possibility for a healthy environment for the next generation.

- **The Specific Aims**

- Establishment of a geographical representative mother-child cohort of 800 pregnant Inuit women to explore effects of exposures during fetal development
- Follow-up on the mother and child with 5 years intervals to
 - Register life style parameters for health and development
 - Register time trend of exposure levels
 - To evaluate the possible relation between health and changes in life styles and exposures.
- Comparison with data from birth cohorts in Northern Norway, China and Denmark



ACCEPT – Hypotheses and Objectives

Hypotheses

- Environmental exposures can disrupt fetal development but the health effects may first become manifested later in life such as
 - Impairment of brain development .
 - Reproductive disorders
 - Impaired immune functions
 - Metabolic syndrome e.g. obesity and type 2 diabetes
 - Hormone related cancers

Objectives

- Biomonitoring and documentation of longtime effects of pre- and postnatal exposures to POPs (PCBs, OC pesticides, PFCs, MeHg, Pb, Se & other trace elements)
- Evaluation of epidemiologic mother and child cohort data vs. toxicological mechanistic *ex vivo* effect biomarkers
- Evaluation of whether PUFAs og Selenium can protect against adverse effects of environmental contaminants
- Documentation of adverse effects on child development upon smoking, alcohol, and medicine intake during pregnancy

ACCEPT project steps

- Phase I:
 - 2009 – 2011: Preparation of documents for questionnaire and blood sampling, start of the cohort sampling and start of analyses
 - 2013-2014: Sampling ongoing and expected to be finished December 2014
- Phase II: 2013 – 2015
 - Finalizing chemical and biomarker analyses
 - Data evaluation
 - Final report to The Danish Environmental Protection Agency
 - Writing of manuscripts for international peer-reviewed journals

ACCEPT mother - child indicators

- Based on questionnaires
 - Basic data for anthropometric data, diet and lifestyle factors and general health status
- Analytical standard data for biomarkers of diet and health
 - Blood pressure
 - Fatty Acid profile (n-3, n-6),
 - Cholesterol (total, HDL, LDL); triglycerides,
 - Serum urate,
 - Thyroid Hormone status,
 - Glyco-hemoglobin (metabolic control of glucose level over time),
 - Gamma-glutamyl-transferase (GGT, diabetes index),
 - Endocrine biomarkers such as FSH, LH, estradiol, androgen hormones, SHBG)

Analytical follow-up program

- **Chemicals/POPs:**
 - DDT/DDE, HCH, toxaphenes, HCB, PCBs, BFR, PFCs, toxic metals (Hg, Pb, Cd), trace elements (Cu, Zn, Se)
- **POP effect biomarkers**
 - Total serum dioxin activity
 - Serum POP related xenoestrogen, xenoandrogen og xenothyroid activity
 - Oxidative stress status
 - “Epigenetic biomarkers”
 - “Gene polymorphisms relevant for POP exposure and health risk and Gene-profile analyses”
- **Blood samples and DNA for the biobank at AU**

Current Status

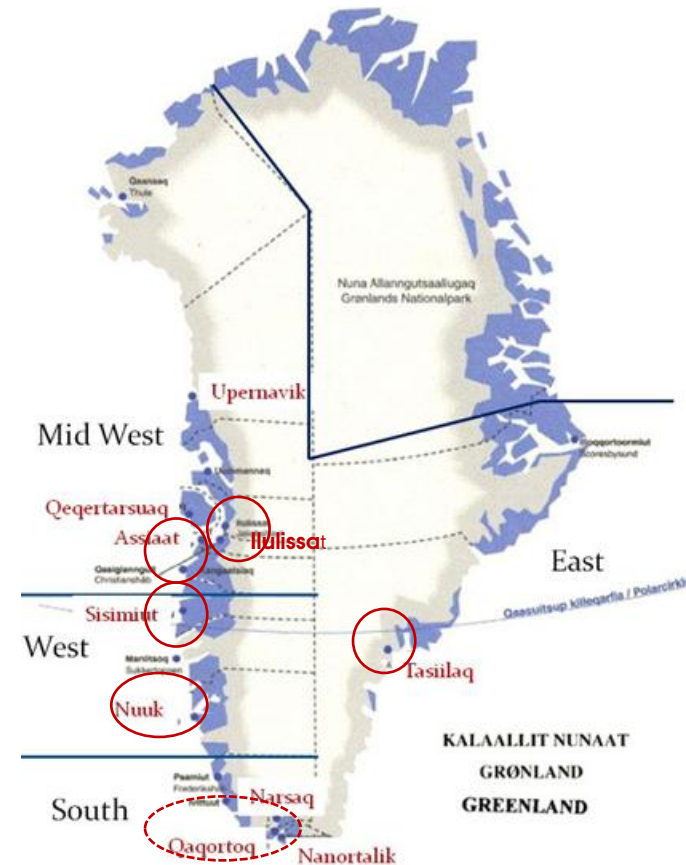
- Blood samples and questionnaires have been sampled 2010-2011 from 192 pregnant women from different districts
 - Nuuk 94, Maniitsoq 40, Aasiaat 18, Ilulissat 33, Paamiut 4, Qaanaaq 3
- Laboratory analyses of some standard parameters are finished for the 192 samples

Continuation of sampling 2013-14

- In cooperation with
 - Head of Institute *Suzanne Møller* & Associated Professor *Lise Hounsgaard*, Institute of Nursing and Health Sciences, Ilisimatusarfik (GRL. University Nuuk), midwife's and the Laboratory at Queen Ingrid's Hospital

- District to be included
 - Nuuk / Sisimiut /Aasiaat(West)
 - Disco Bay / (Mid-West GRL)
 - Tassilaq (East GRL)
 - Qaqortoq(maybe) (South GRL)

- Sampling expected finished Dec. 2014
- Follow up on the children age 3 will be started during 2013



PERFLUORINATED COMPOUNDS ARE RELATED TO BREAST CANCER RISK IN GREENLANDIC INUIT: *A CASE – CONTROL STUDY*

EC. Bonefeld-Jorgensen, M. Long, R. Bossi, P. Ayotte, G.
Asmund, T. Krüger, M. Ghisari, G. Mulvad, P. Kern, P.
Nzulumiki, E. Dewailly. .

Journal of Environmental Health 2011, 10:88



Breast cancer in the Arctic – changes over the past decades

- Breast cancer, the most common cancer in women in the western world, is increasing all over the world, including in the Arctic.
- The enormous transition in health conditions and lifestyle in the Arctic might be contributing to *the known risk factors*.
- In Greenland the *age at menarche* has diminished by three years during the course of 100 years, and the *number of children per women* as well as the duration of *breastfeeding* is decreasing.
- Obesity and intake of saturated fat is increasing and the intake of traditional food rich in unsaturated fat (omega-3) and vitamin D is decreasing. Smoking and alcohol consumption in the Arctic has been relatively high but are now decreasing.
- **However, the known established risk factors cannot alone account for the increasing trends observed.**



Breast Cancer incidences and suspected risk factors in the Arctic

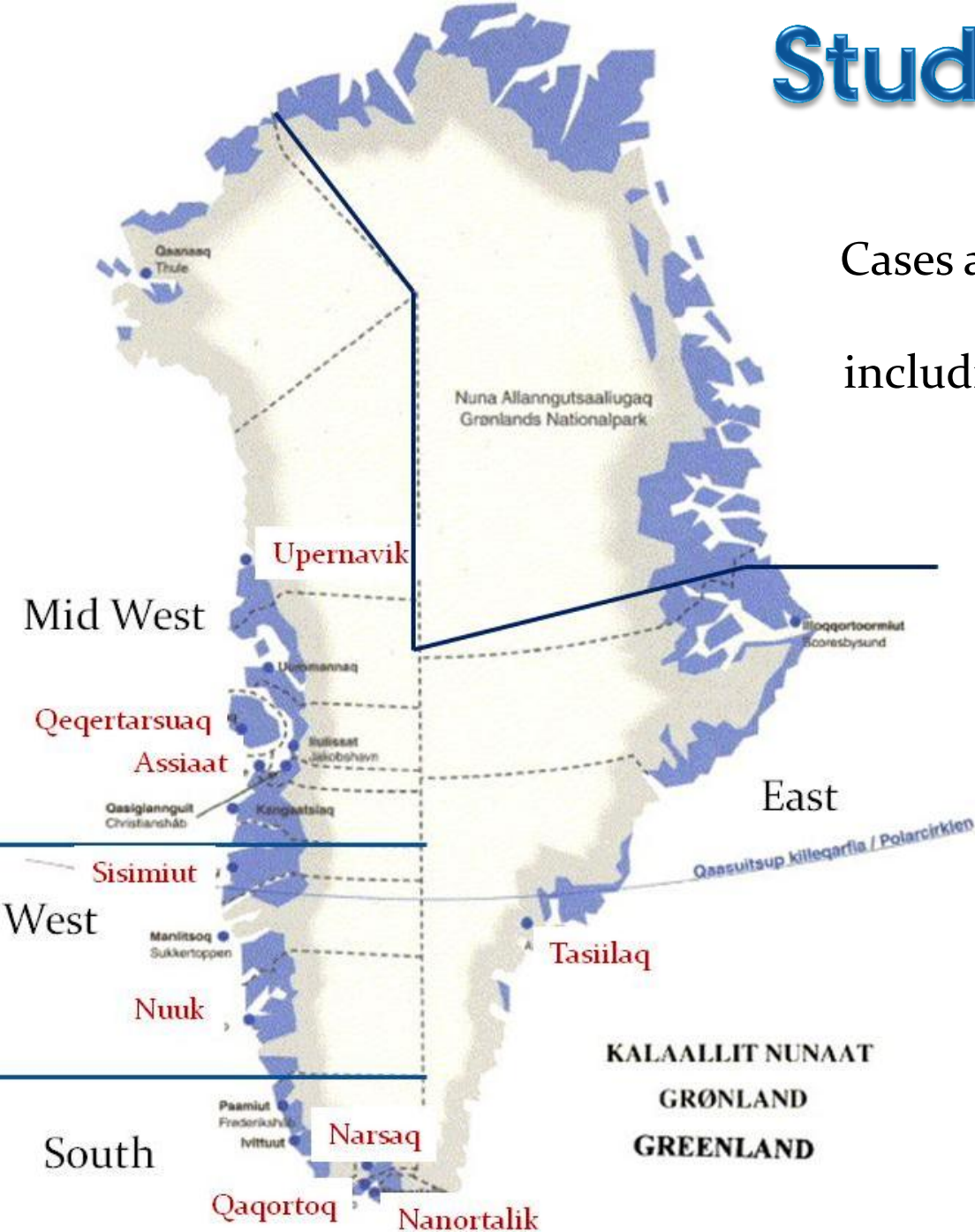
- From very few cases in 1960's an increase in BC has been observed in the Inuit population of Greenland and Canada
 - Greenland, 1950-74: 57 Breast cancer cases were diagnosed ($\sim 2.3/\text{year}$)
 - Greenland, 1988-97: the age-adjusted incidence rate was **46.4 per 100.000 women** (world-standard)
 - Denmark, 2010: age adjusted incidence rate was **100 per 100.000 women**(world-standard)
 - USA, 2001 to 2008: the age-adjusted incidence was **124 per 100.000 women** (world-standard)
- Previous studies suggest that exposure to persistent organic pollutants (POPs) e.g. PCBs might contribute to the risk of BC
- Rat studies showed that perfluorinated compounds (PFCs) cause significantly increase in mammary fibroadenomas

Aim of the study

- To elucidate factors involved in the increasing incidence of Breast cancer in Greenlandic Inuit by means of
 - Evaluation of the association between serum levels of POPs such as Perfluorinated Compounds (PFCs) and lipophilic POPs (lip-POPs) in breast cancer cases and their frequency matched controls
 - PFCs: omnipresent water and oil repellent, water, dust, food, food packing etc.
 - Lip-POPs: fatty tissues of (marine) animals and humans
 - Evaluation of hormone disruption by the combined serum POP mixture bio-effects on nuclear and hormone receptor functions
 - Evaluation of the genetic aspect in the risk of breast cancer

Study population

Cases and controls were sampled during
2000-2003
including 80% of all cases in this period



DISTRICT	CASE	CONTROL
Mid West		
Upernavik,	1	
Qeqertarsuaq	1	9
Assiaat	4	
%	19.4	7.8
West		
Sisimiut	4	
Nuuk	11	89
%	48.8	77.4
South		
Narsaq	3	10
Qaqortoq	2	
Nanortalik	2	
%	22.6	8.7
East		
Tasiilaq	2	7
%	6.5	6.1
Total	30 (31)*	115

*1 case misses the district information



Data for known Breast Cancer risk factors

Cases vs. Control

- The number of full term pregnancies were significant lower for cases ($p < 0.0001$)
- The premenopausal and postmenopausal status for cases were higher and lower for cases, respectively ($p = 0.001$)
- Lower level of serum cotinine was found for cases ($p < 0.05$)
- Non-significant higher level of serum estradiol and lower level of the ratio n-3/ n-6 and selenium for cases.
- No differences for age, BMI and breastfeeding was found



Exposure Biomarkers and biomarkers of effect

Cases vs. controls

POP Exposure

- Cases had significantly higher serum PFC levels
 - PFOS $p < 0.0001$; PFOA $p < 0.04$
 - Σ PFSA and Σ PFCA $p < 0.0001$ and $p = 0.001$, respectively
- Cases had also significantly higher level of Σ PCBs in the highest quartile ($p = 0.02$)

Hormone disruption

- Cases had higher POP induced hormone-like activity being significant for the androgen system ($p = 0.02$), and lower dioxin-like activity (AhR-TEQ) ($P < 0.009$)



Odds Ratios of BC cases vs. controls

Variables	n (controls/cases)	95% CI	p
Raw			
PFOS (ng/ml)	98/31	1.01(1.00; 1.02)	0.02
ΣPFSA (ng/ml)	98/31	1.01(1.00; 1.02)	0.02
ΣPCB+OC+PFCs	115/31	1.01(1.00;1.01)	0.03
X-AR t-activity (RLU/ug prt)	58/27	8.52(1.55; 46.8)	0.01
Adjusted			
PFOS (ng/ml)	69/9	1.03(1.00; 1.07)	0.05
ΣPFSA (ng/ml)	69/15	1.03(1.00; 1.05)	0.02
ΣPCB+OC+PFCs	85/11	1.02(1.01; 1.04)	0.01
X-AR t-activity* (RLU/ug prt)	49/11	44.1(1.99; 975.7)	0.02

Adjustment: age, BMI, pregnancy, cotinine, breastfeeding, menopausal status

*X-AR-t-act: xeno-androgen receptor transactivity



Gene polymorphism differences between cases and controls

- Cases had a higher frequency (10% vs. 3%) of the variant BRACA 123-118 genotype being in accordance to earlier studies in Greenland
- Cases had a higher frequency of the variant genotype for the P450 phase I *CYP1A1* (30% vs. 24%) and *CYP1B1* (3.2% vs. 1%) genes involved in hormone synthesis and metabolism and detoxification of xenobiotics e.g. POPs
- Cases had also a higher frequency of the variant genotype for the phase II *COMT* (Catechol-O-methyltransferase) gene involved in catabolism of catecholamines and inactivation of catechol estrogens (2-OH-E₂ and 4-OH-E₂)



Conclusions and Perspectives

- PFC and POPs are BC risk factors: High serum POP concentrations, particularly PFCs, are risk factors in the development of Breast Cancer in Greenlandic Inuit
- POPs are hormone disruptive: The combined serum lipo-POP related xeno-estrogenic and xeno-androgenic agonistic activities may contribute to the risk of developing breast cancer in Inuit
- Genetic polymorphisms might cause differences in susceptibility to exposure to contaminants and the risk of breast cancer.

Perspectives

To improve the statistical power and design the sampling of data and blood from new BC and ovarian cancers are currently ongoing in cooperation with *Managing Consultant Peder Kern, Nurse Sofiannnguaq Jakobsen, Secretary Marie S. Jakobsen, Head Nurse Linda Seelk, Quen Ingrid's Hospital Nuuk, Greenland.*



Thank you for the attention

