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# Frozen Ovarian Tissue Transplant Results:

# Pregnancy and Live Births

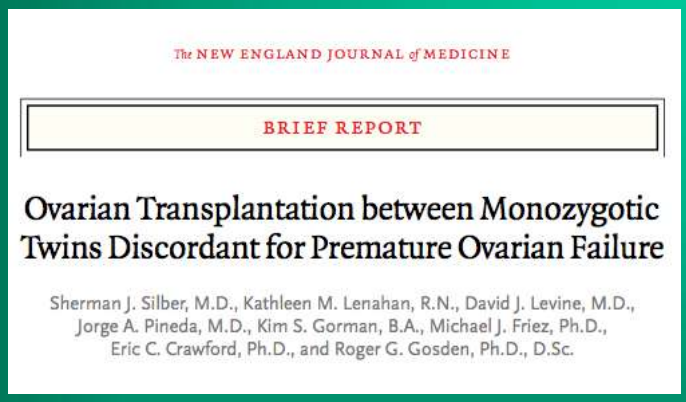
## Frozen Ovarian Tissue Transplant Results:

## Vitrification Vs. Slow Freeze

## Frozen Ovarian Tissue Transplant Results:

## Leukemia

# IDENTICAL TWINS DISCORDANT FOR PREMATURE OVARIAN FAILURE

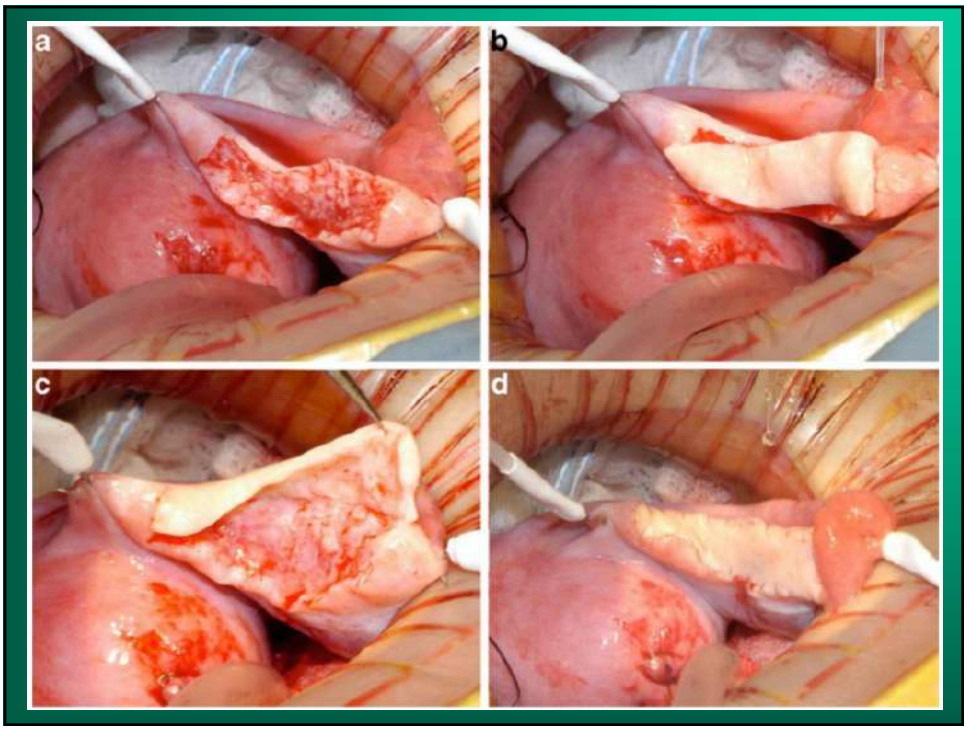


The NEW ENGLAND JOURNAL of MEDICINE

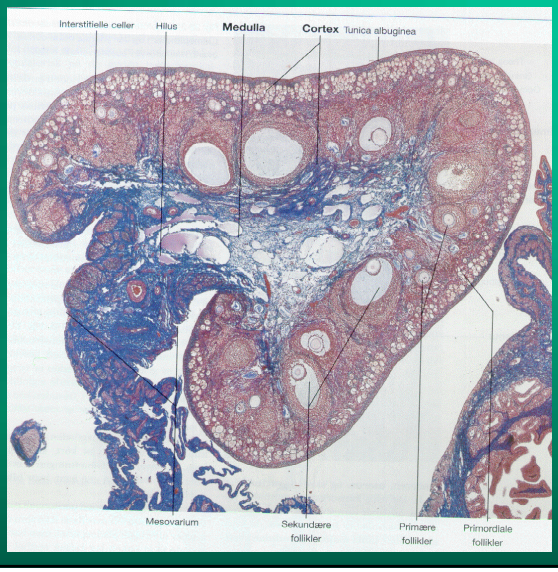
BRIEF REPORT

Ovarian Transplantation between Monozygotic Twins Discordant for Premature Ovarian Failure

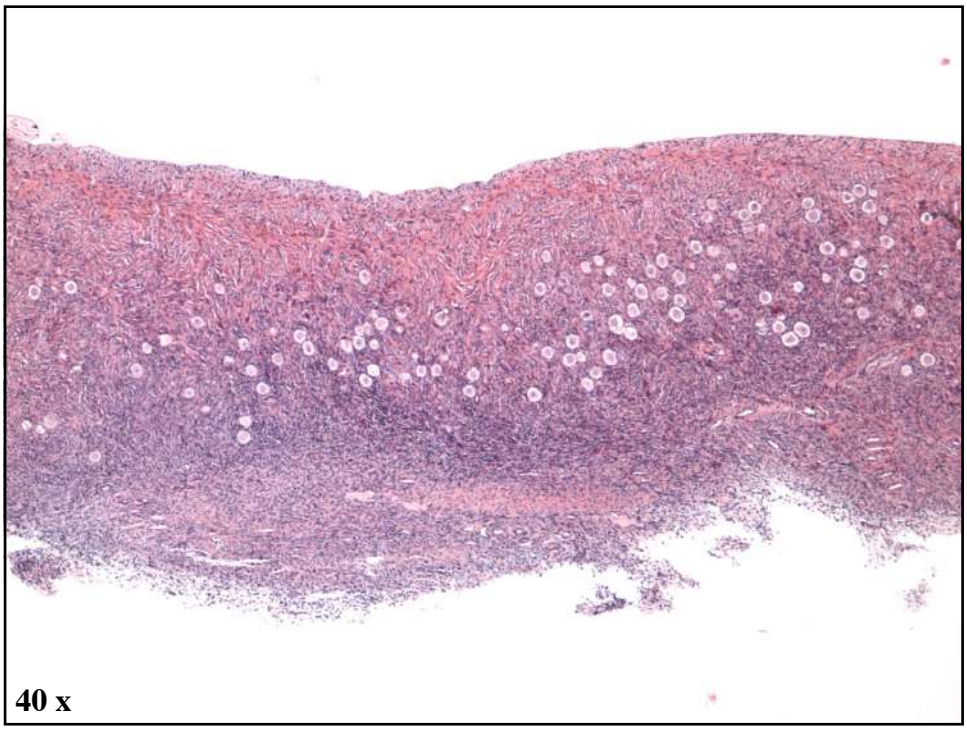
Sherman J. Silber, M.D., Kathleen M. Lenahan, R.N., David J. Levine, M.D., Jorge A. Pineda, M.D., Kim S. Gorman, B.A., Michael J. Friez, Ph.D., Eric C. Crawford, Ph.D., and Roger G. Gosden, Ph.D., D.Sc.




## Primordial Follicles All Located in Cortex



## Follicles Develop Inward Toward the Softer Ovarian Medulla

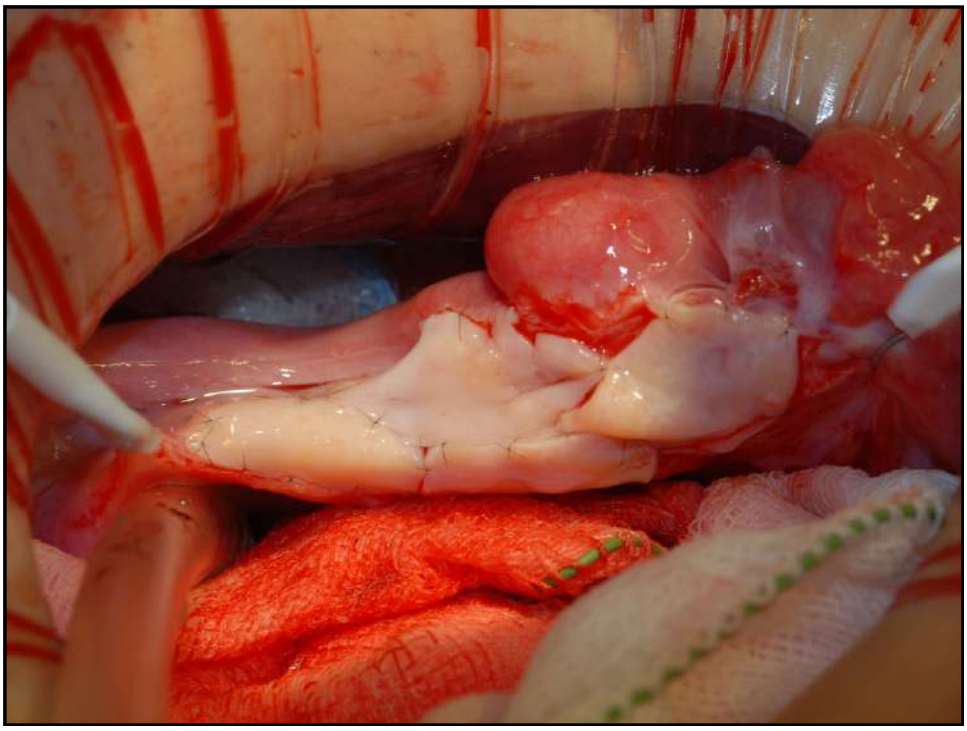



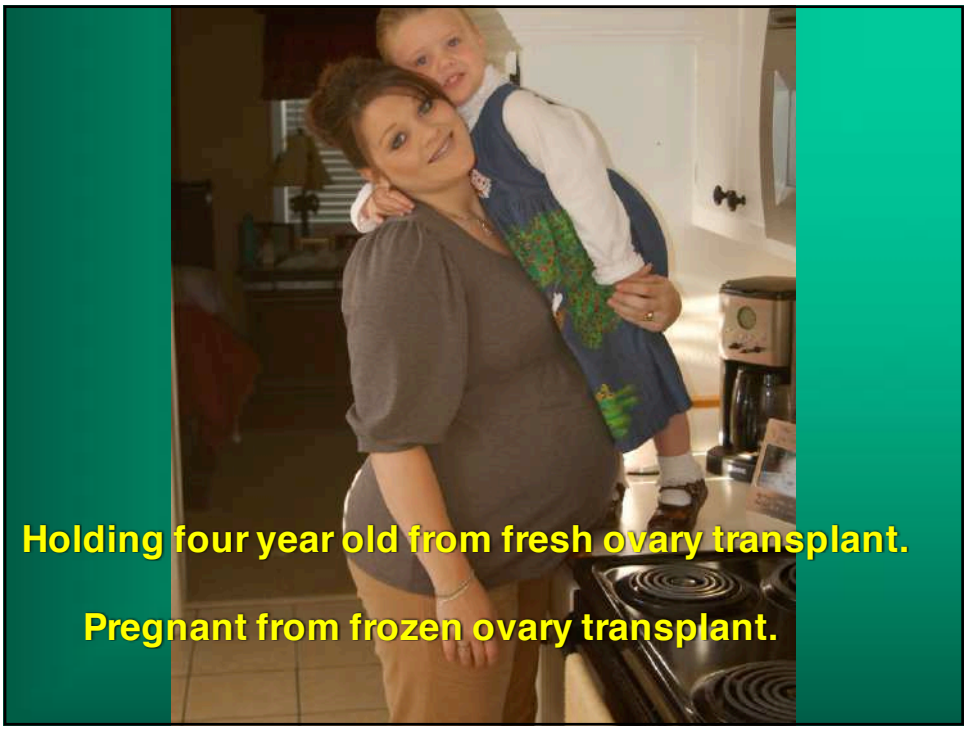
Melanie and Stephanie

case # 1

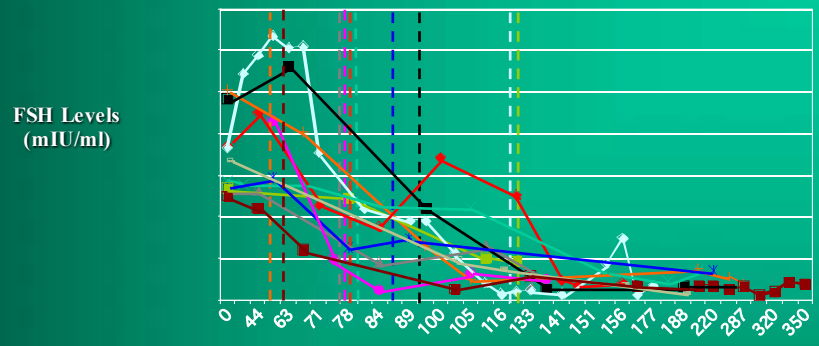

June 7, 2005: normal healthy baby girl
routine vaginal delivery,
3600 grams, apgar 9 and10.


FIRST SUCCESSFUL FRESH
TRANSPLANT DEPLETED
OF EGGS AFTER >4 YEARS




Holding four year old from fresh ovary transplant.
Pregnant from frozen ovary transplant.

## Recovery of Ovarian Function: Fresh

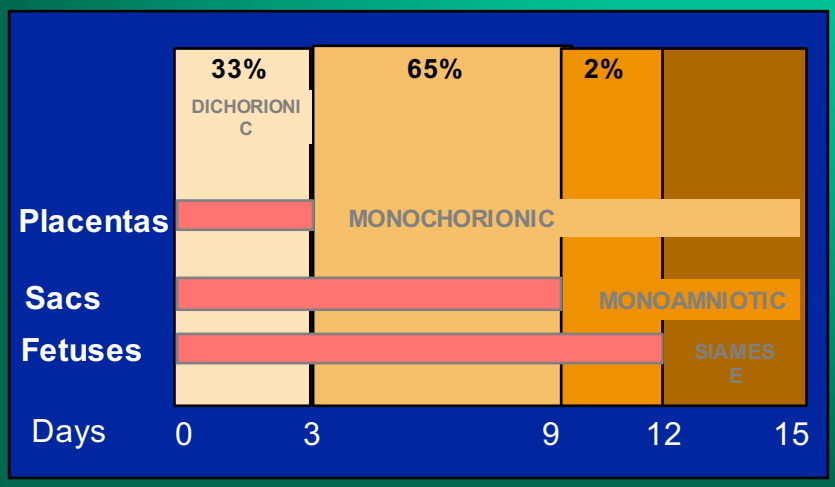



## OVULATION ALWAYS RECOVERS IN 4.5 MONTHS

**THEREFORE IT REQUIRES 4.5 MONTHS FOR RECRUITED RESTING FOLLICLES TO REACH OVULATION STAGE**

## Monozygotic twin pregnancies:

### *IPS Cell Study (Amanda Clark)*




ELSEVIER
www.sciencedirect.com
www.rbmonline.com

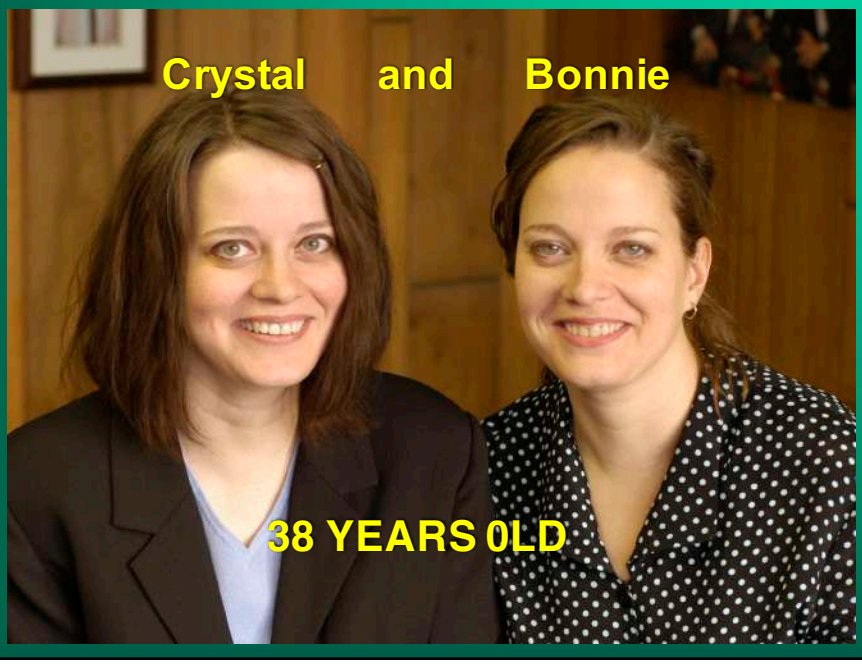
REVIEW

**Long-term duration of function of ovarian tissue transplants: case reports**

Claus Yding Andersen [a], Sherman J Silber [b,*], Stinne Holm Berghold [c], Jan Stener Jorgensen [d], Erik Ernst [e]

Reproductive BioMedicine Online (2012) Vol. 25

## Identical Twin Case # 2




## Frozen Ovaries

- 13 patients over a 10 year period underwent thaw and transplantation of ovarian tissue that had been frozen up to 20 years earlier with detailed follow-up of hormones, menstruation, pregnancies, and birth.
- Three of the thirteen had had leukemia. Nine had undergone slow freeze and 4 had undergone vitrification.

## Materials and Methods

- 108 females between age 6 and 35 were referred for possible ovary tissue cryopreservation, 92 of whom had the procedure performed, over a 20 year period.
- Before 10 years ago only slow freeze was utilized, and after that only vitrification. Nineteen underwent slow freeze, and the rest vitrification.

## Materials and Methods

- 13 cases whose tissue was frozen prior to age 35 returned up to 18 years later to have their tissue transplanted back, and have more than 2 years of post-transplant followup.
- 9 were from slow freeze before 10 years earlier, and 4 were from vitrification in the last 10 years. No IVF or ancillary treatment was administered.

## Results

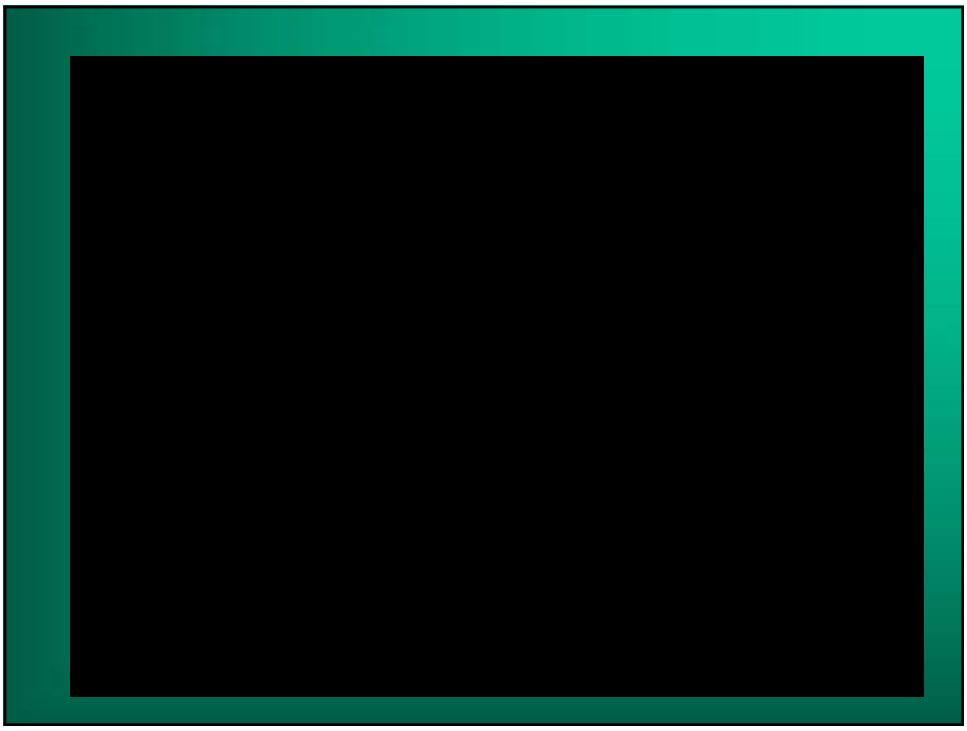
- All 13 cases had return of ovarian function 4 to 5 months post frozen transplant with return of FSH to normal with regular menstrual cycling. AMH rose to high levels as the FSH came down to normal.
- Then AMH 4 months later declined to very low levels, but the grafts remained functional for 5 years or longer, and 8 of the 13 are still functioning.

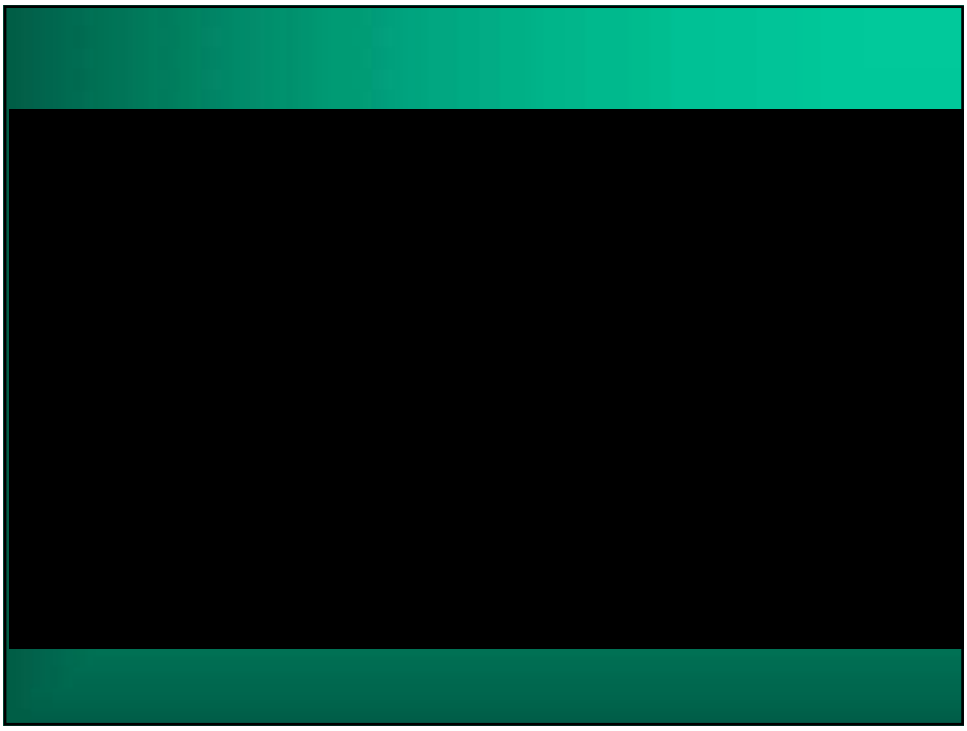
## Conclusion

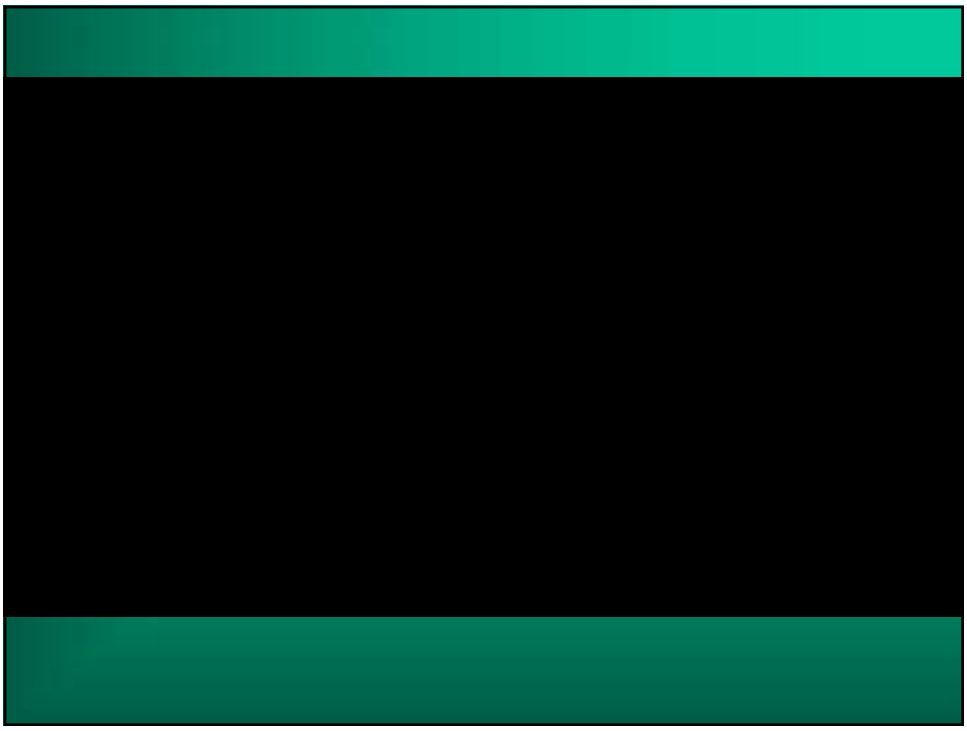
- Ovarian tissue cryopreservation with subsequent transplantation is a robust method of fertility preservation.
- Vitrification is easier, and results in less loss of oocytes (over 50 %).
- But not superior to slow freeze. Why?

## Conclusion

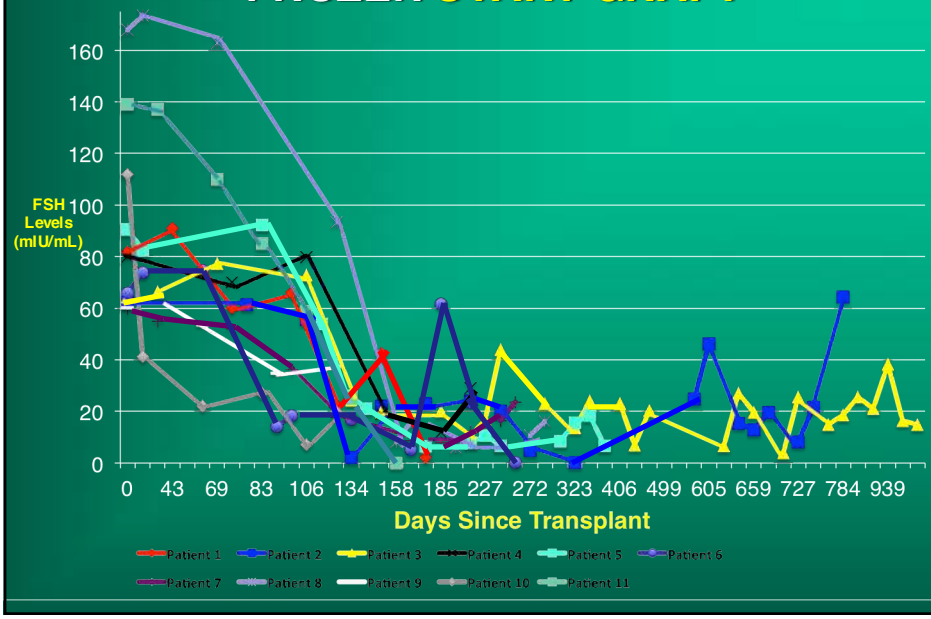
- When you have 200,000 oocytes, losing 100,000 from slow freeze makes no significant biologic difference.
- The lower the ovarian reserve, the lower is the rate of primordial follicle recruitment.
- Transmission of cancer cells has not been a problem, but for leukemia, the tissue must be frozen when the patient is in remission.
- Ovary tissue transplantation, fresh or frozen, should not longer be considered “experimental”.







## RETURN OF FSH TO NORMAL AFTER FROZEN OVARY GRAFT




## Preg Fresh Transplant: 7/9 (78%)
## Preg Frozen Transplant : 10/13 (77%)

13 babies from frozen
11 babies from fresh

TOTAL: 24 HEALTHY BABIES FROM OVARY TRANSPLANT IN ONE CENTER (ST LOUIS)

## Ovary Tissue Freeze Transplants

| Date of Transplant | Age At Transplantation | Age At Freeze | Diagnosis | Pregnant | Live Birth Or Ongoing | Time Until Pregnancy (Days) | Miscarriages | Duration of Ovarian Function (Days) |
|---|---|---|---|---|---|---|---|---|
| 3/6/07 | 26 | 24 | POF | YES | Female | 174 | | 675 (Ended) |
| 1/13/09 | 31 | 20 | Hodgkins | YES | Male | 272 | | 885 (Ended) |
| 6/9/09 | 29 | 24 | POF | YES | | 276 | 1 | 561 (Ended) |
| 6/17/11 | 33 | 20 | Hodgkins | NO | | | | 1155 (Ended) |
| 10/12/12 | 33 | 31 | MS | YES | Female | 481 | | 1846 (Still Functioning) |
| 3/29/13 | 32 | 25 | POF | YES | Female | 243 | | 794 (Ended) |
| 4/5/13 | 33 | 30 | Brain Cancer | YES | Male | 665 | | 1671 (Still Functioning) |
| 4/12/13 | 25 | 18 | Leukemia | YES | Male | 502 | | 1664 (Still Functioning) |
| | | | | YES | Female | 998 | | |
| | | | | YES | Ongoing ? | 1578 | | |
| 10/1/13 | 29 | 28 | Synovial Sarcoma | NO | | | | 1492 (Still Functioning) |
| 10/7/13 | 39 | 24 | Leukemia | YES | Ongoing ? | 1287 | | 1486 (Still Functioning) |
| 7/21/15 | 28 | 25 | Leukemia | NO | | | | 834 (Still Functioning) |
| 8/5/15 | 32 | 21 | Hodgkins | YES | Female | 343 | | 819 (Still Functioning) |
| 9/18/14 | 36 | 20 | Hodgkins | YES | Female | 473 | | 1140 (Still Functioning) |
| | | | | YES | Ongoing (2 Female) | 908 | | |
| Totals | 13 Cases | 9 Babies (+4 Ongoing) | | 10 Pregnant (77%) | | | | 4 Vitrification<br>9 Slow Freeze |

## Ovary Tissue Freeze Transplants

| Date of Transplant | Age At Transplantation | Age At Freeze | Diagnosis | Pregnant | Live Birth Or Ongoing | Time Until Pregnancy (Days) | Miscarriages | Duration of Ovarian Function (Days) |
|---|---|---|---|---|---|---|---|---|
| 3/6/07 | 26 | 24 | POF | YES | YES | 174 | | 675 (Ended) |
| 1/13/09 | 31 | 20 | Hodgkins | YES | YES | 272 | | 885 (Ended) |
| 6/9/09 | 29 | 24 | POF | YES | | 276 | 1 | 561 (Ended) |
| 6/17/11 | 33 | 20 | Hodgkins | NO | | | | 1155 (Ended) |
| 10/12/12 | 33 | 31 | MS | YES | YES | 481 | | 1846 (Still Functioning) |
| 3/29/13 | 32 | 25 | POF | YES | YES | 243 | | 794 (Ended) |
| 4/5/13 | 33 | 30 | Brain Cancer | YES | YES | 665 | | 1671 (Still Functioning) |
| 4/12/13 | 25 | 18 | Leukemia | YES | YES | 502 | | 1664 (Still Functioning) |
| | | | | YES | YES | 998 | | |
| | | | | YES | YES | 1578 | | |
| 10/1/13 | 29 | 28 | Synovial Sarcoma | NO | | | | 1492 (Still Functioning) |
| 10/7/13 | 39 | 24 | Leukemia | YES | YES | 1287 | | 1486 (Still Functioning) |
| 7/21/15 | 28 | 25 | Leukemia | NO | | | | 834 (Still Functioning) |
| 8/5/15 | 32 | 21 | Hodgkins | YES | YES | 343 | | 819 (Still Functioning) |
| 9/18/14 | 36 | 20 | Hodgkins | YES | YES | 473 | | 1140 (Still Functioning) |
| | | | | YES | YES YES | 908 | | |
| Totals | 13 Cases | 13 Babies | | 10 Pregnant (77%) | | | | 4 Vitrification<br>9 Slow Freeze |

# Pregnancy After Frozen Autografts

| Vitrified Slow Freeze | Duration of Ovarian Function (Days) | Date of Transplant | Date of First Menstruation post OT | Diagnosis | Miscarriage | Baby Born | Ongoing | Girl | Boy |
|---|---|---|---|---|---|---|---|---|---|
| Slow freeze | 675 (Ended) | 3/6/07 | 9/19/08 | POF | | 1 | | 1 | |
| Slow freeze | 885 (Ended) | 1/13/09 | 6/7/09 | Hodgkins | | 1 | | | 1 |
| Slow freeze | 561 (Ended) | 6/9/09 | 11/28/09 | POF | 1 | 0 | | | |
| Slow freeze | 1155 (Ended) | 6/17/11 | 11/5/11 | Hodgkins | | 0 | | | |
| Vitrified | 1846 (Still Functioning) | 10/12/12 | 3/2/13 | MS | | 1 | | 1 | |
| Slow freeze | 794 (Ended) | 3/29/13 | 4/5/13 | POF | | 1 | | 1 | |
| Vitrified | 1671 (Still Functioning) | 4/5/13 | 12/27/13 | Brain Cancer | | 1 | | | 1 |
| Slow freeze | 1664 (Still Functioning) | 4/12/13 | 1/1/14 | Leukemia | | 2 | 1 | 1 | 1 |
| Vitrified | 1492 (Still Functioning) | 10/1/13 | 12/19/13 | Synovial Sarcoma | | 0 | | | |
| Slow Freeze | 1486 (Still Functioning) | 10/7/13 | 3/6/14 | Leukemia | | 0 | 1 | | |
| Vitrified | 834 (Still Functioning) | 7/21/15 | 11/15/15 | Leukemia | | 0 | | | |
| Slow freeze | 819 (Still Functioning) | 8/5/15 | 10/28/15 | Hodgkins | | 1 | | 1 | |
| Slow freeze | 1140 (Still Functioning) | 9/18/14 | 2/2/15 | Hodgkins | | 1 | 2 | 1 | |
| Totals | 13 Cases | 9 Babies (+4 Ongoing) | 10 Patients Pregnant (77%) | | | | | 4 Vitrification 9 Slow Freeze | |

## Ovary Tissue Freeze Transplants

| Name | Date of Transplant | Age At Transplantation | Age At Freeze | Diagnosis | Pregnant | Live Birth Or Ongoing | Time Until Pregnancy (Days) | Miscarriages | Duration of Ovarian Function (Days) |
|---|---|---|---|---|---|---|---|---|---|
| Yarber | 3/6/07 | 26 | 24 | POF | YES | Female | 174 | | 675 (Ended) |
| Tucker | 1/13/09 | 31 | 20 | Hodgkins | YES | Male | 272 | | 885 (Ended) |
| Yarber | 6/9/09 | 29 | 24 | POF | YES | | 276 | 1 | 561 (Ended) |
| Tucker | 6/17/11 | 33 | 20 | Hodgkins | NO | | | | 1155 (Ended) |
| Remington-Hobbs | 10/12/12 | 33 | 31 | MS | YES | Female | 481 | | 1846 (Still Functioning) |
| Madsen | 3/29/13 | 32 | 25 | POF | YES | Female | 243 | | 794 (Ended) |
| Farrow | 4/5/13 | 33 | 30 | Brain Cancer | YES | Male | 665 | | 1671 (Still Functioning) |
| Schaikewitz | 4/12/13 | 25 | 18 | Leukemia | YES | Male | 502 | | 1664 (Still Functioning) |
| | | | | | YES | Female | 998 | | |
| | | | | | YES | Ongoing ? | 1578 | | |
| Sahr/Phillips | 10/1/13 | 29 | 28 | Synovial Sarcoma | NO | | | | 1492 (Still Functioning) |
| Shipp/Obrien | 10/7/13 | 39 | 24 | Leukemia | YES | Ongoing ? | 1287 | | 1486 (Still Functioning) |
| Dailey | 7/21/15 | 28 | 25 | Leukemia | NO | | | | 834 (Still Functioning) |
| Bernstein | 8/5/15 | 32 | 21 | Hodgkins | YES | Female | 343 | | 819 (Still Functioning) |
| Tucker | 9/18/14 | 36 | 20 | Hodgkins | YES | Female | 473 | | 1140 (Still Functioning) |
| | | | | | YES | Ongoing (2 Female) | 908 | | |

# Ovary Tissue Freeze Transplants SLOW FREEZE

| Name | Date of Transplant | Age At Transplantation | Age At Freeze | Diagnosis | Pregnant | Live Birth Or Ongoing | Time Until Pregnancy (Days) | Miscarriages | Duration of Ovarian Function (Days) |
|---|---|---|---|---|---|---|---|---|---|
| Yarber | 3/6/07 | 26 | 24 | POF | YES | Female | 174 | | 675 (Ended) |
| Tucker | 1/13/09 | 31 | 20 | Hodgkins | YES | Male | 272 | | 885 (Ended) |
| Yarber | 6/9/09 | 29 | 24 | POF | YES | | 276 | 1 | 561 (Ended) |
| Tucker | 6/17/11 | 33 | 20 | Hodgkins | NO | | | | 1155 (Ended) |
| Madsen | 3/29/13 | 32 | 25 | POF | YES | Female | 243 | | 794 (Ended) |
| Schaikewitz | 4/12/13 | 25 | 18 | Leukemia | YES | Male | 502 | | 1664 (Still Functioning) |
| | | | | | YES | Female | 998 | | |
| | | | | | YES | Ongoing ? | 1578 | | |
| Bernstein | 8/5/15 | 32 | 21 | Hodgkins | YES | Female | 343 | | 819 (Still Functioning) |
| Tucker | 9/18/14 | 36 | 20 | Hodgkins | YES | Female | 473 | | 1140 (Still Functioning) |
| | | | | | YES | Ongoing (2 Female) | 908 | | |
| Shipp/Obrien | 10/7/13 | 39 | 24 | Leukemia | YES | Ongoing ? | 1287 | | 1486 (Still Functioning) |
| Totals | 9 Cases | | 7 Babies (+4 ongoing) | | 7 Patients Pregnant (78%) | | | | |

# Ovary Tissue Freeze Transplants VITRIFIED

| Name | Date of Transplant | Age At Transplantation | Age At Freeze | Diagnosis | Pregnant | Live Birth Or Ongoing | Time Until Pregnancy (Days) | Miscarriages | Duration of Ovarian Function (Days) |
|---|---|---|---|---|---|---|---|---|---|
| Remington-Hobbs | 10/12/12 | 33 | 31 | MS | YES | Female | 481 | | 1846 (Still Functioning) |
| Farrow | 4/5/13 | 33 | 30 | Brain Cancer | YES | Male | 665 | | 1671 (Still Functioning) |
| Sahr/Phillips | 10/1/13 | 29 | 28 | Synovial Sarcoma | NO | | | | 1492 (Still Functioning) |
| Dailey | 7/21/15 | 28 | 25 | Leukemia | NO | | | | 834 (Still Functioning) |
| Totals | 4 Cases | | 2 Babies | | 2 Patients Pregnant | | 0 Miscarriage | | |

# Ovary Tissue Freeze Transplants LEUKEMIA

| Name | Date of Transplant | Age At Transplantation | Age At Freeze | Diagnosis | Pregnant | Live Birth Or Ongoing | Time Until Pregnancy (Days) | Miscarriages | Duration of Ovarian Function (Days) |
|---|---|---|---|---|---|---|---|---|---|
| Schaikewitz | 4/12/13 | 25 | 18 | Leukemia | YES | Male | 502 | | 1664 (Still Functioning) |
| | | | | | YES | Female | 998 | | |
| | | | | | YES | Ongoing ? | 1578 | | |
| Shipp/Obrien | 10/7/13 | 39 | 24 | Leukemia | YES | Ongoing ? | 1287 | | 1486 (Still Functioning) |
| Dailey | 7/21/15 | 28 | 25 | Leukemia | NO | | | | 834 (Still Functioning) |
| Totals | 3 Cases | | 2 Babies + 2 Ongoing | | 4 Pregnancies 2 Became Pregnant (67%) | | 0 Miscarriage | | Average Age (30 yrs old) |

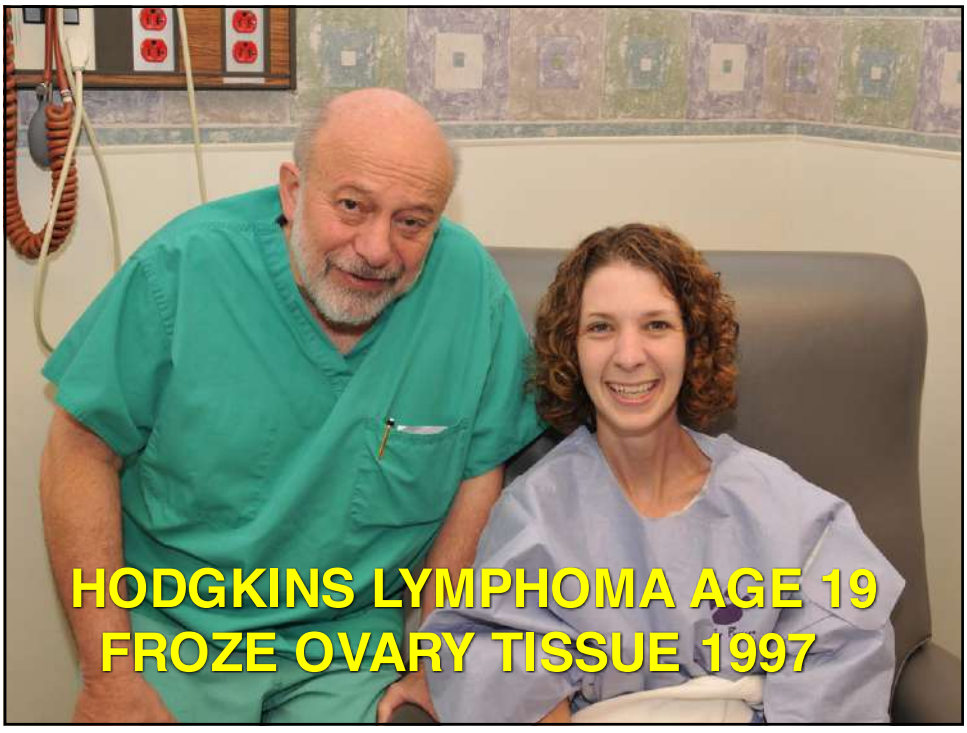
# Ovary Tissue Freeze Transplants NEW

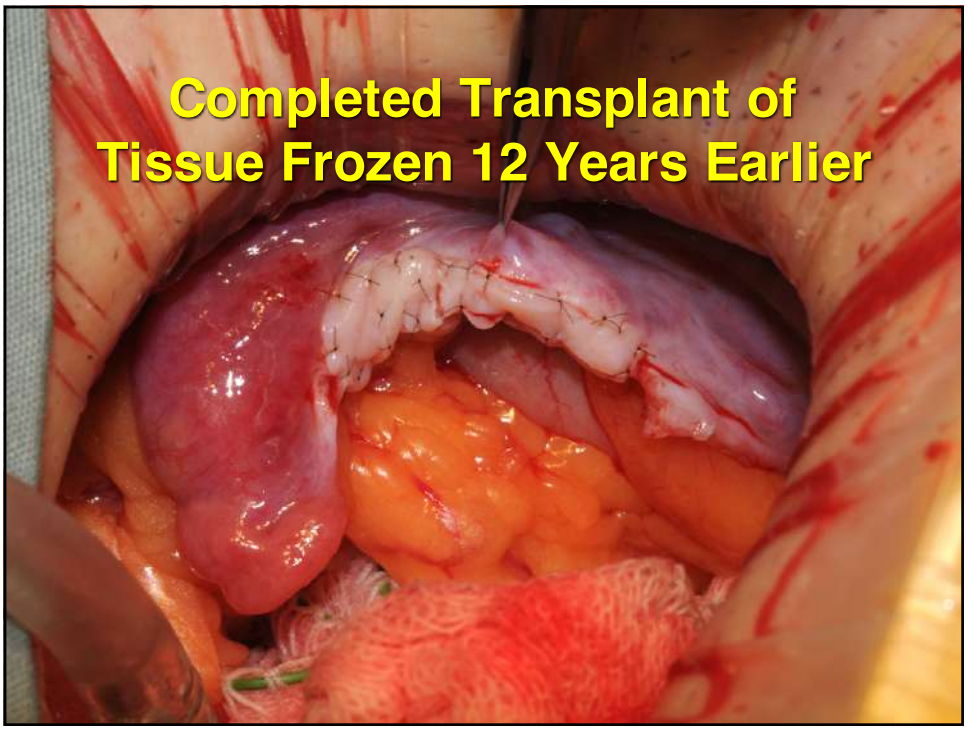
| Name | Date of Transplant | Age At Transplantation | Age At Freeze | Diagnosis | Pregnant | Live Birth Or Ongoing | Time Until Pregnancy (Days) | Miscarriages | Duration of Ovarian Function (Days) |
|---|---|---|---|---|---|---|---|---|---|
| Amthor | 2/1/17 | 35 | 24 | Non-Hodgkins | NO | | | | 273 (Still Functioning)<br>No Period |
| Prosser | 6/1/17 | 36 | 33 | Large B-Cell Lymphoma | NO | | | | 153 (Still Functioning)<br>Period 8/23/17 (203 days) |
| Copeland | 10/2/17 | 25 | 24 | Hodgkins Lymphoma | NO | | | | 30 (Still Functioning)<br>No Period |
| Totals | 2 Cases | | 0 Babies | | 0 Pregnancies | | 0 Miscarriage | | |

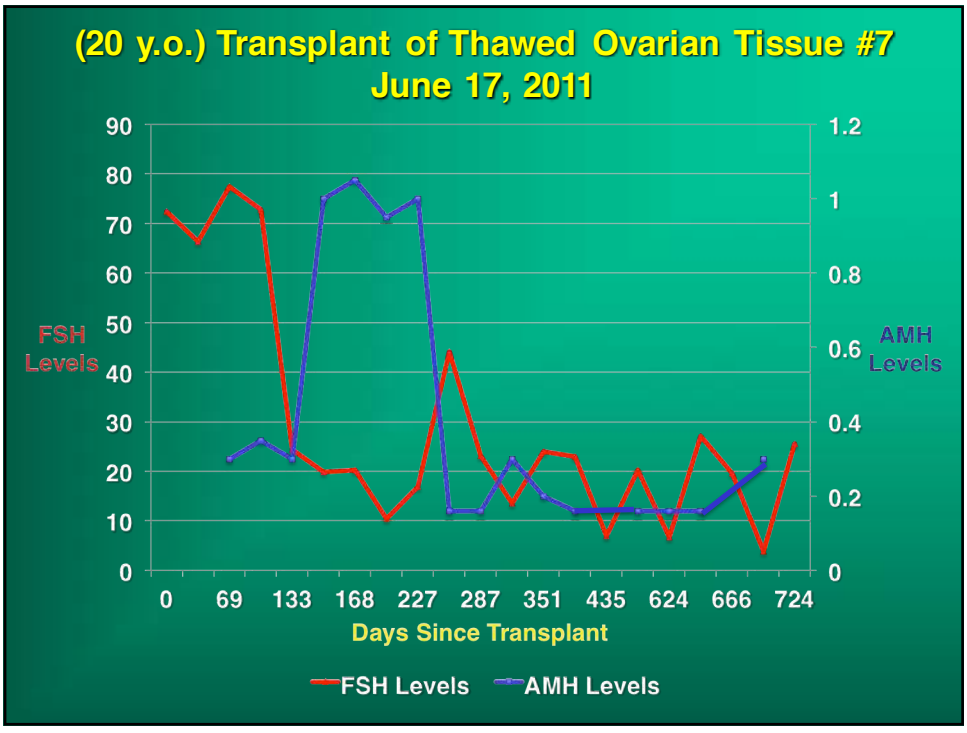
## Ovary Tissue Freeze Transplants

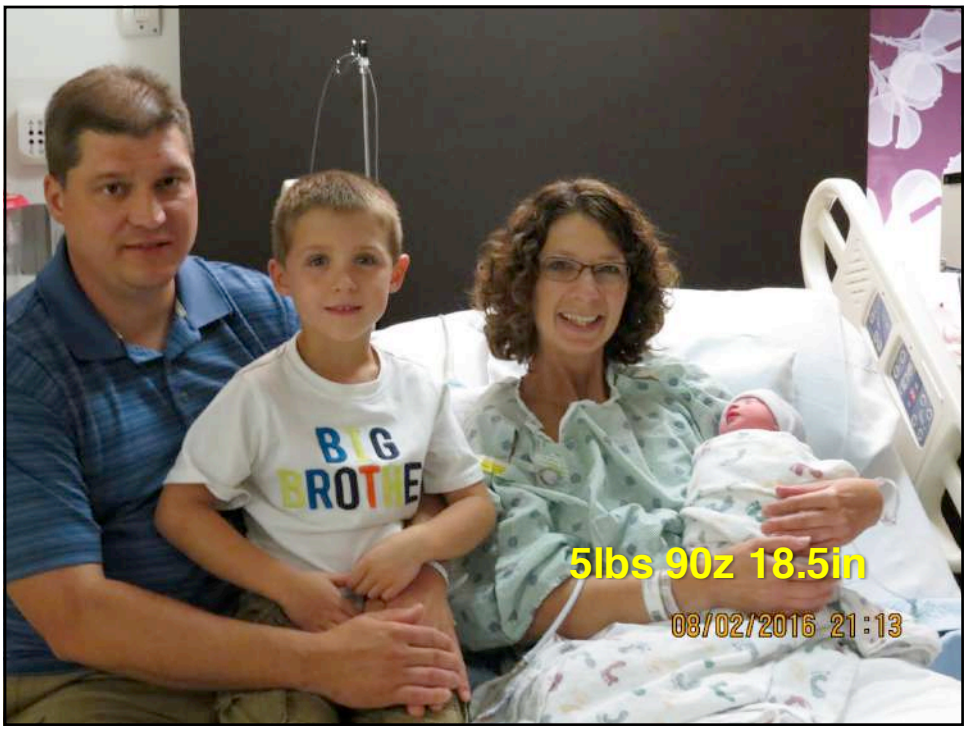
### VITRIFIED VS SLOW FREEZE

| Slow Freeze | Vitrified |
|---|---|
| 9 Cases | 4 Cases |
| 7 Babies (+4 Ongoing) | 2 Babies |
| 11 Pregnancies | 2 Pregnancies |
| 1 Miscarriage | 0 Miscarriage |




Completed Transplant of
Tissue Frozen 12 Years Earlier


(20 y.o.) Transplant of Thawed Ovarian Tissue #7
June 17, 2011
FSH Levels
AMH Levels
90
80
70
60
50
40
30
20
10
0
1.2
1
0.8
0.6
0.4
0.2
0
0 69 133 168 227 287 351 435 624 666 724
Days Since Transplant
FSH Levels
AMH Levels


5lbs 90z 18.5in
08/02/2016 21:13


Reflecting 14 Years Later Why She Feels Fortunate She Had Cancer: A Longer Reproductive Lifespan.

# Techniqiue of the Cryopreservation,

## And of

## The Microsurgery

# DETERMINING THE CONCENTRATION OF CRYOPROTECTANT REQUIRED FOR VITRIFICATION OF OVARIAN TISSUE


T ; Translucent
= Vitrified

I ; Intermediate
= Partially vitrified.

M; Milky
= Not vitrified (Ice formation)

Table 1. Visual aspect of ovarian tissue during cooling and warming.

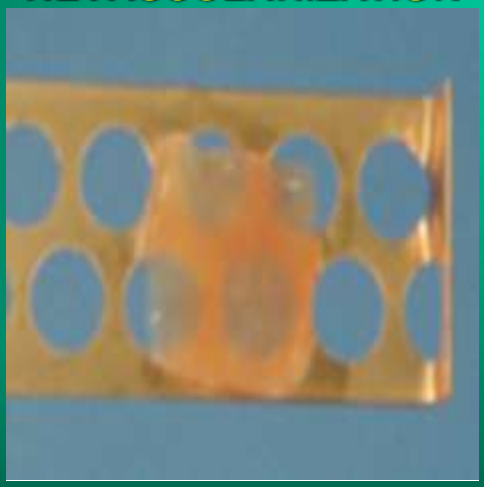
| | Cooling −196°C | Warming 37℃ |
|---|---|---|
| 25% | M | M |
| 30% | M | M |
| 35% | I-T | M-I |
| 40% | T | T |
| 45% | T | T |
| 50% | T | T |
| 55% | T | T |

CPA: EG+ DMSO+ 0.5M Sucrose,
M; Milky and I; Intermediate = Not vitrified. T; Translucent= Vitrified

## DETERMINING THE CONCENTRATION OF CRYOPROTECTANT REQUIRED FOR VITRIFICATION OF OVARIAN TISSUE


**Ovary Transplant**
***Vitrification Clip 4***

## ULTRA-THIN SLICE ON METAL GRID: GOOD FOR VITRIFICATION AND RAPID REVASCULARIZATION

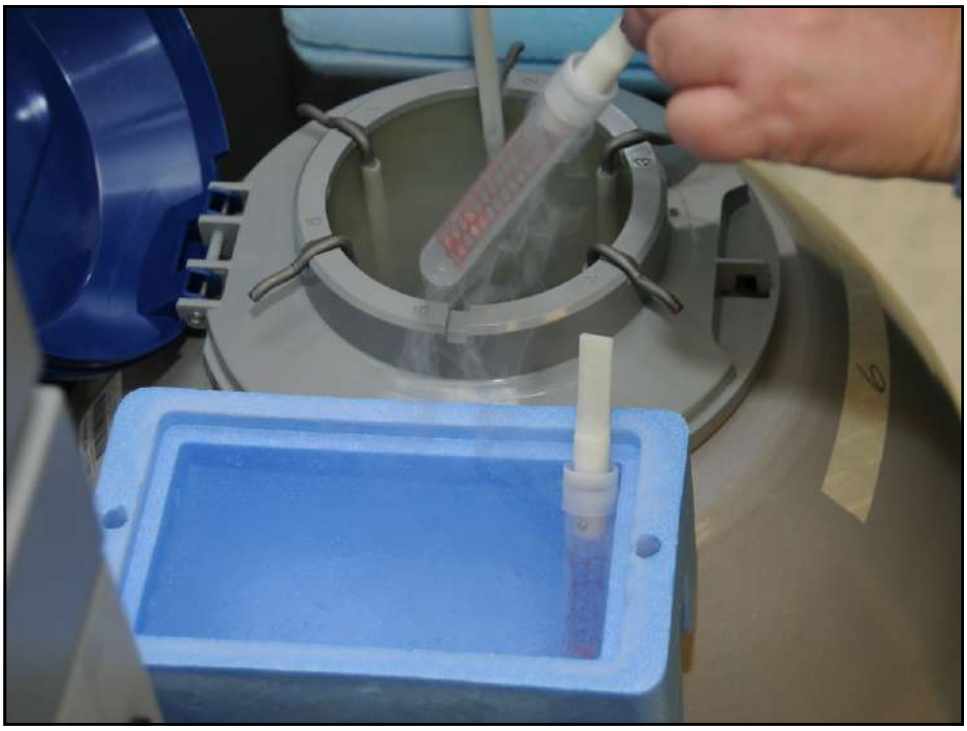


## Vitrification of Ovarian Tissue

- ES Solution is the same as for Embryo or Oocyte vitirfication, 15 % cryoprotectant (7.5 % DMSO and 7.5 % EG with 20 % Protein in Hepes.
- VS Solution however is 40 % cryoprotectant (20 % DMSO and 20 % EG, instead of 15 % DMSO and 15 % EG).
- Leave tissue slices in ES Solution for 25 minutes.
- Then place in VS solution until the thin tissue slices fall to the bottom of the centrifuge tube (usually < 15 minutes).

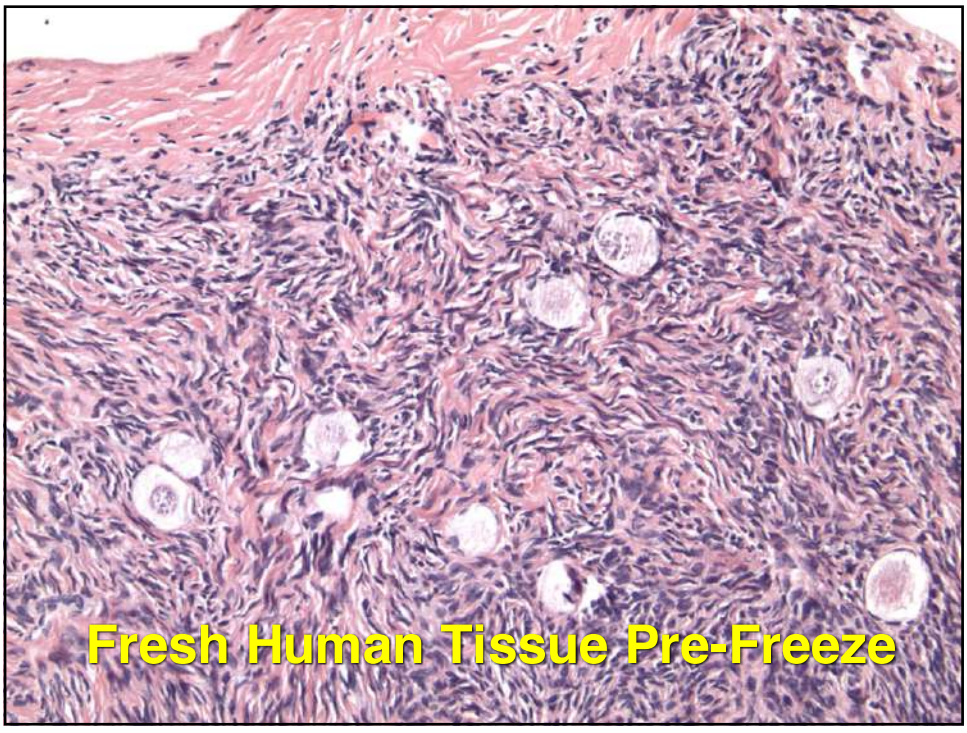
## Ovary Transplant
*Tissue Slices in Test Tube*


Meirow & Silber - July 2009

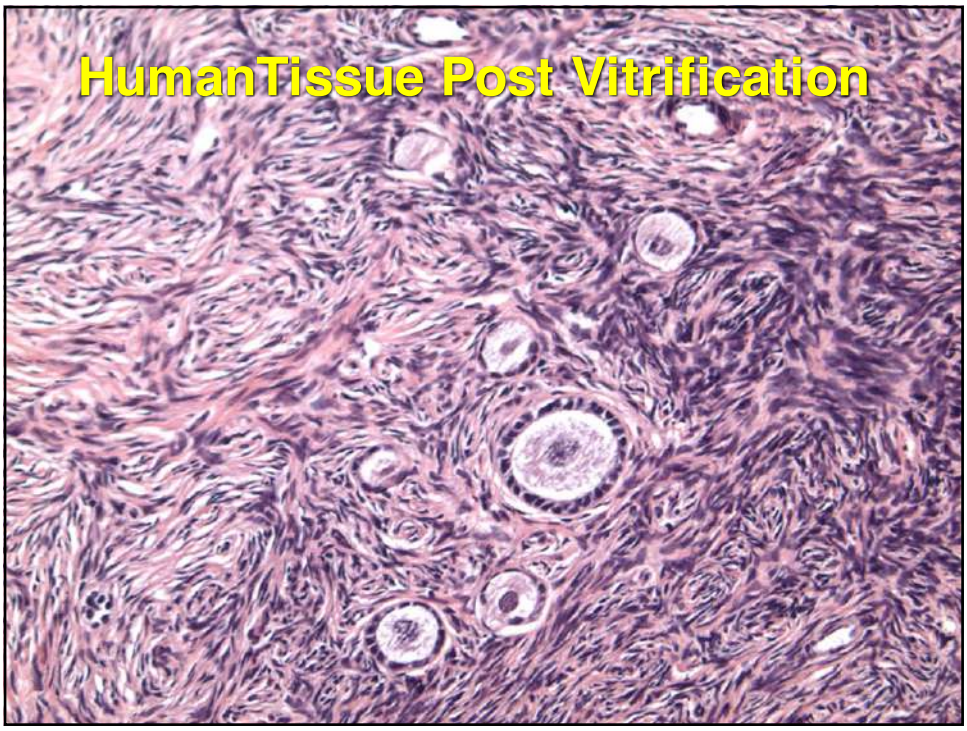


## Vitrification Procedure (Thaw)

- For thaw, metal strip is immersed directly into 40 ml of 1.0 molar sucrose at 37 degrees C for at least 1 minute.
- Then transferred into 0.5 molar sucrose for 5 minutes at room temp.
- Then washed twice. No ice crystal formation observed.



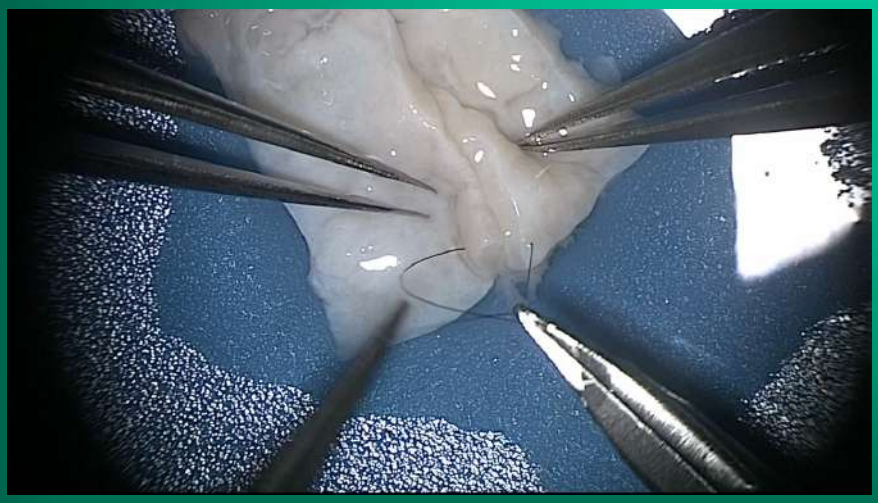
## HumanTissue Post Vitrification



## Post-thaw Survival of Oocytes: Vitrification vs Slow Freeze

| | Ovarian tissues | Number Collected | (%) of oocytes Surviving |
|---|---|---|---|
| Vitrified | 8 | 1122 | 1000 (89.1%) |
| Fresh | 2 | 358 | 329 (91.9%) |
| Slow-Freeze | 6 | 821 | 342 (41.7%) |

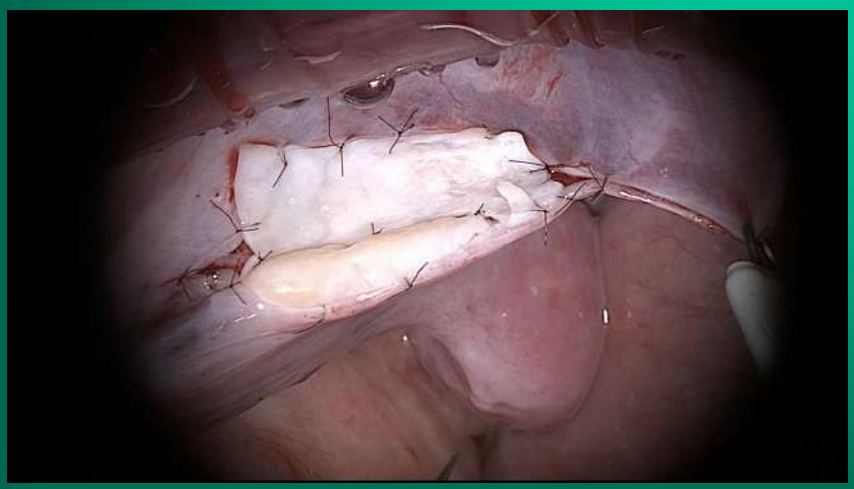
## Ovary Transplant
*Tissue Quilting Clip 1*



## Ovary Transplant
*Tissue Quilting Clip 2*

## Ovary Transplant
*Tissue Quilting Clip 3*

## Ovary Transplant
*Transplant Clip 5*



## Why do the ovarian transplants maintain endocrine function for such extented periods of time?

- The recruitment rate of primordial follicles becomes reduced in parallel to a reduced ovarian reserve.
- By replacing one single piece of cortex at a time recruitment rate will constantly be very low and utilisation of follicles may be augmented compared to nature.


- It is likely that one ovary or part of an ovary will have the capacity to provide menstrual cycles for a considerable period of time.

Wallace H *et al*. *PlosOne*, 2010;5:e8772

## Factors associated with premature ovarian failure, early menopause and earlier onset of menopause in Japanese women

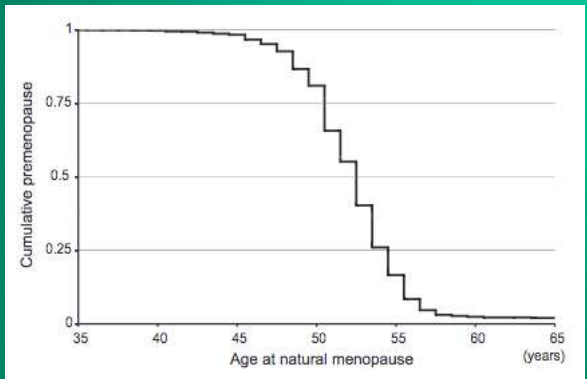



**Fig. 1.** Kaplan–Meier cumulative estimates of natural menopause in Japanese women.

Yasui et al 2012

## Factors associated with premature ovarian failure, early menopause and earlier onset of menopause in Japanese women

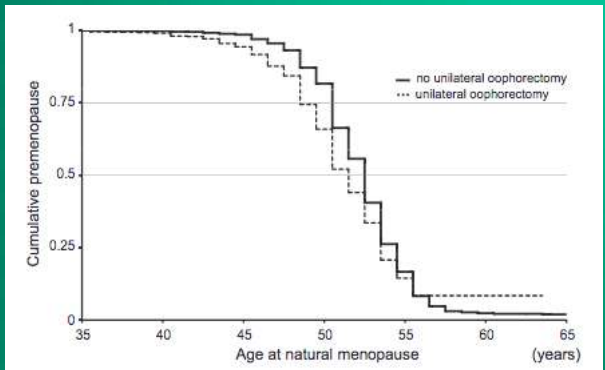



**Fig. 2.** Adjusted Kaplan–Meier cumulative estimates of natural menopause stratified by unilateral oophorectomy in Japanese women. Solid line: no unilateral oophorectomy, Broken line: unilateral oophorectomy.

Yasui et al 2012

## Intrinsic Fertility of Human Oocytes

ORIGINAL ARTICLE: INFERTILITY

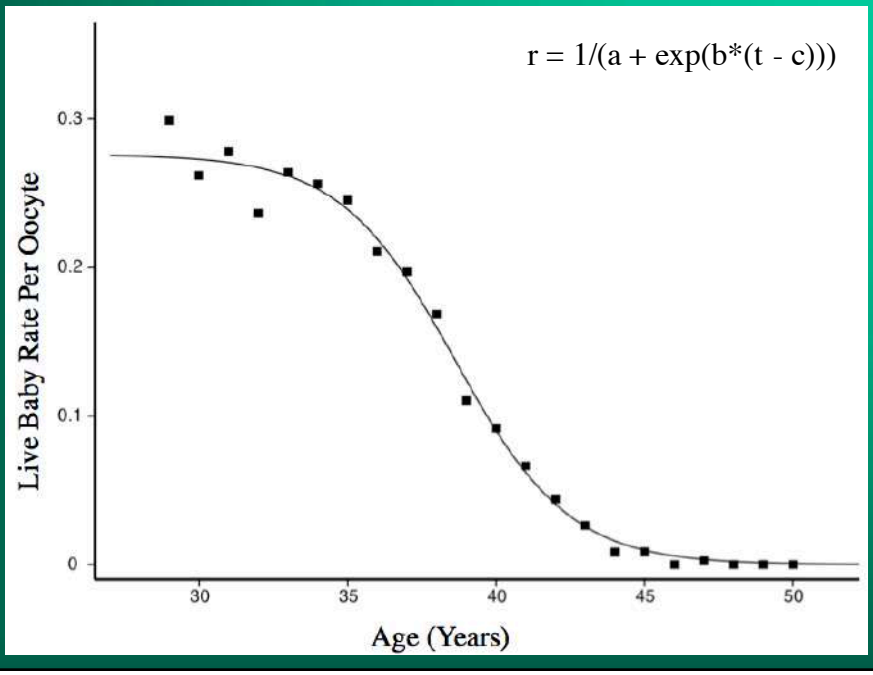
### Intrinsic fertility of human oocytes

Sherman J. Silber, M.D.,[a] Keiichi Kato, M.D., Ph.D.,[b] Naoki Aoyama, M.Sc.,[b] Akiko Yabuuchi, Ph.D.,[b] Helen Skaletsky, Ph.D.,[c] Yuting Fan, M.D.,[c] Kazunori Shinohara, M.D.,[b] Noriyuki Yatabe, M.D., Ph.D.,[b] and Tamotsu Kobayashi, M.D.[b]

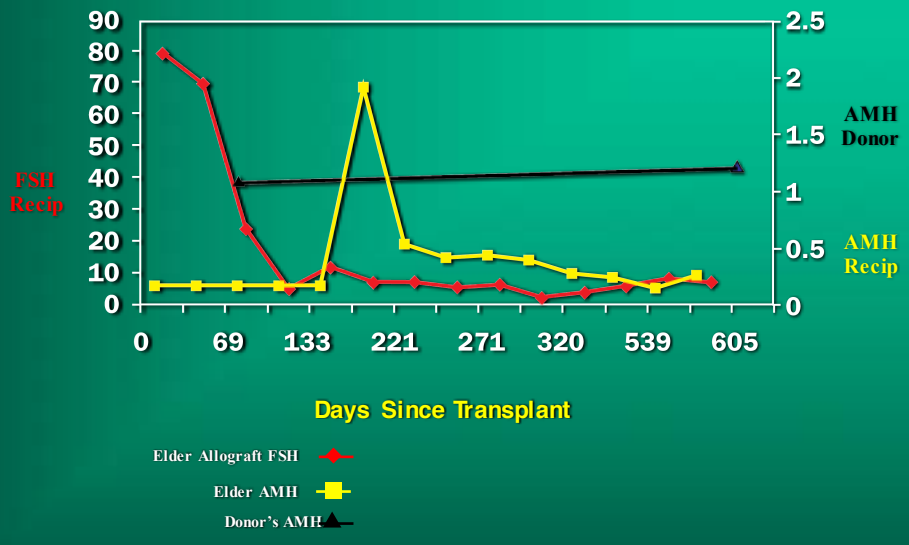
[a] Infertility Center of St. Louis, St. Louis, Missouri; [b] Kato Ladies Clinic, Tokyo, Japan; and [c] Massachusetts Institute of Technology, Whitehead Institute for Biomedical Research, Cambridge, Massachusetts

Silber et al. 2017

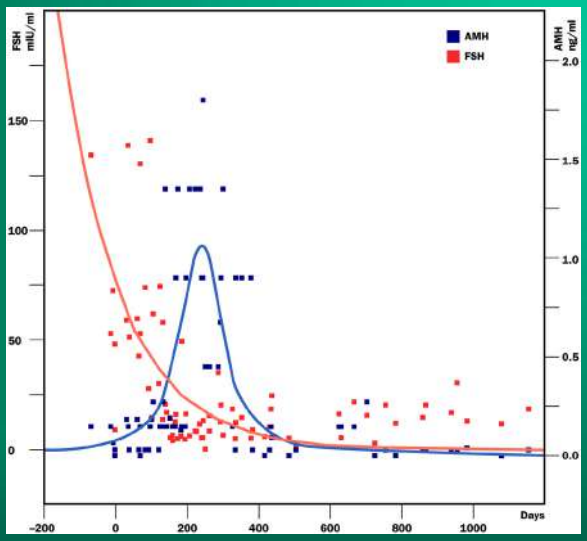
## Fertility Of The Human Oocyte Related to Age



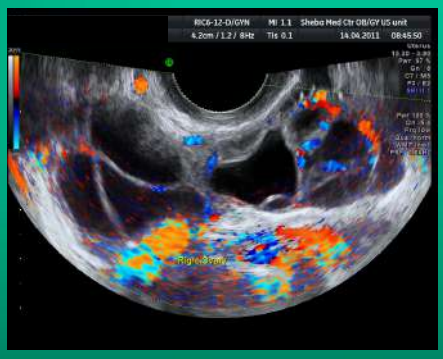

## FSH & AMH in Donor and Recipient: First Allograft



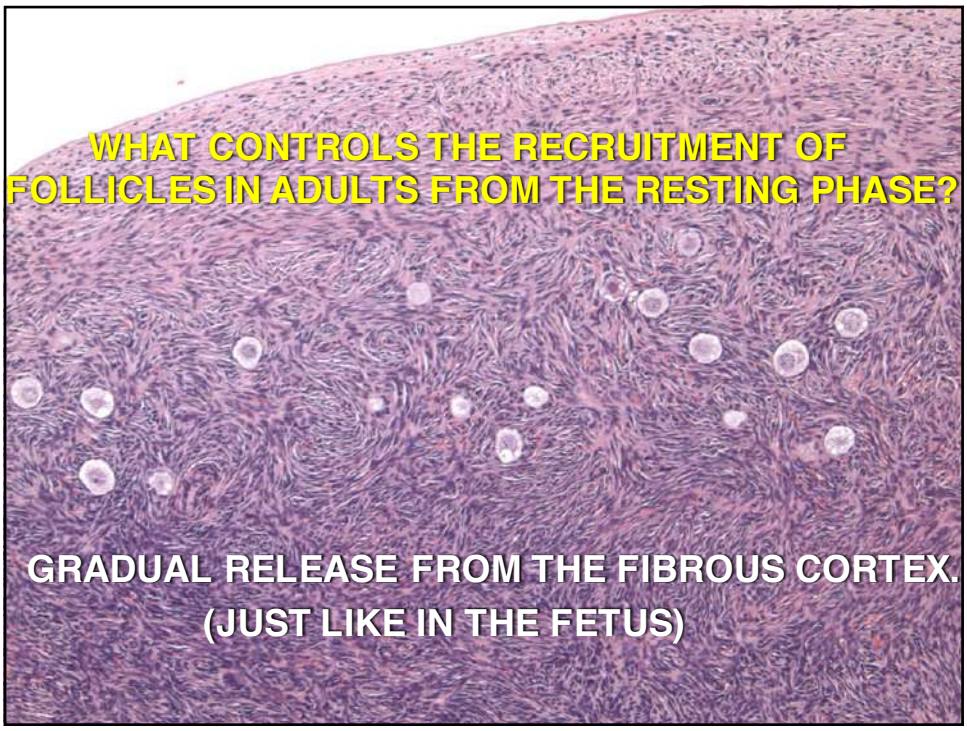
# RELATIONSHIP OF FSH AND AMH AFTER OVARY TRANSPLANTATION

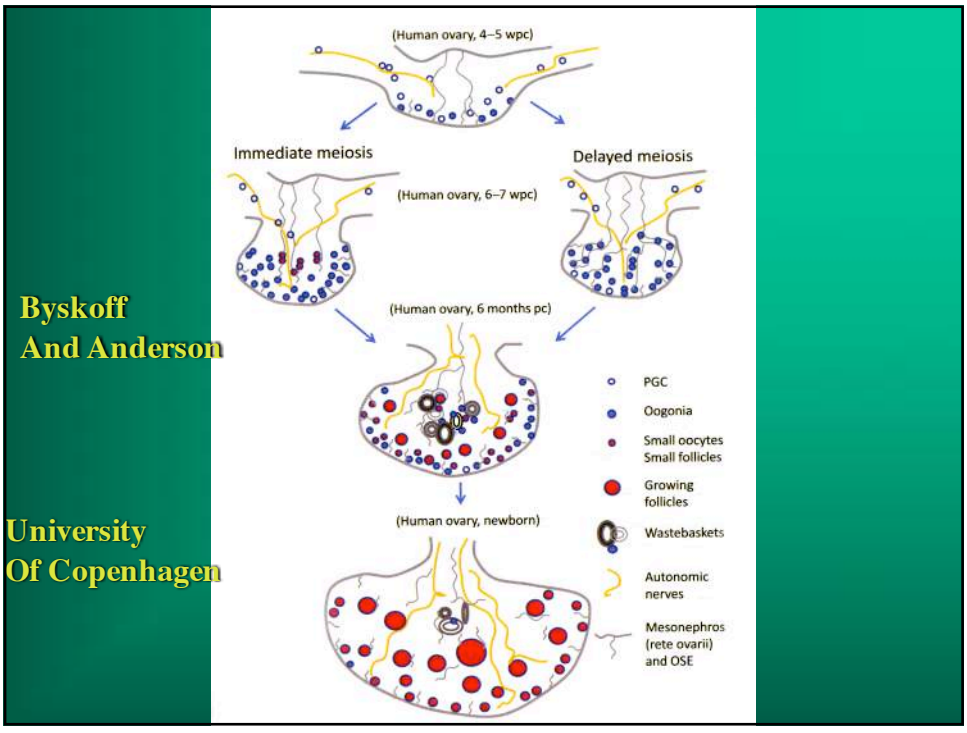



## Stimulated ovary post transplantation of fresh ovarian tissue



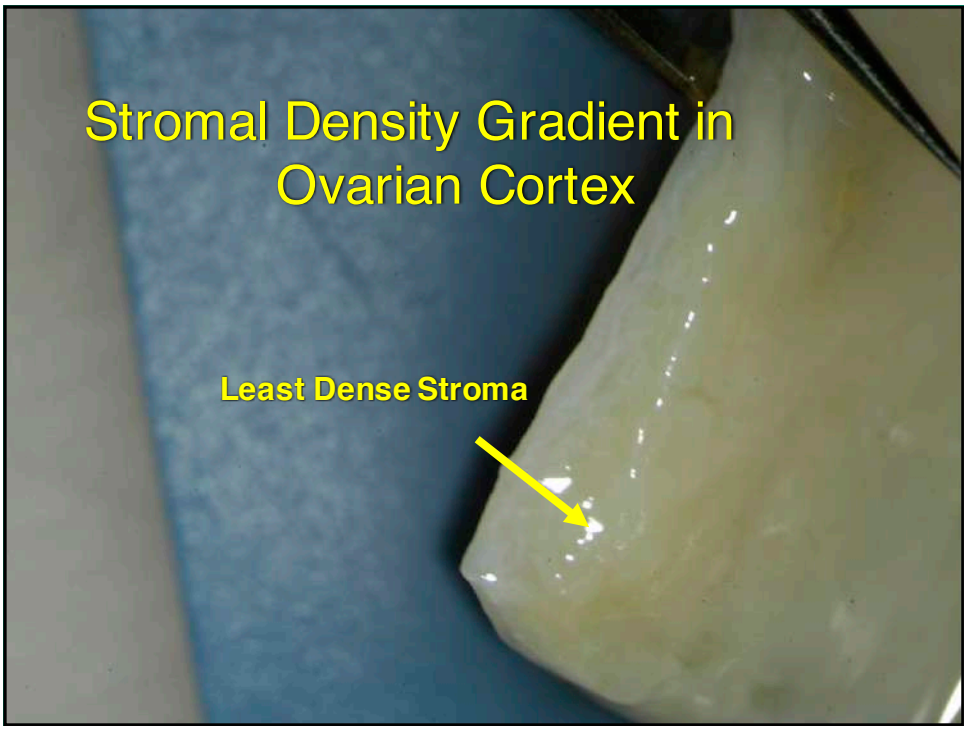
**APPEARS TO BE NO FOLLICLE LOSS FROM ISCHEMIA**


WHAT CONTROLS THE RECRUITMENT OF FOLLICLES IN ADULTS FROM THE RESTING PHASE?
GRADUAL RELEASE FROM THE FIBROUS CORTEX.
(JUST LIKE IN THE FETUS)


Byskoff
And Anderson
University
Of Copenhagen
(Human ovary, 4–5 wpc)
Immediate meiosis
Delayed meiosis
(Human ovary, 6–7 wpc)
(Human ovary, 6 months pc)
(Human ovary, newborn)
PGC
Oogonia
Small oocytes
Small follicles
Growing follicles
Wastebaskets
Autonomic nerves
Mesonephros (rete ovarii) and OSE


Stromal Density Gradient in Ovarian Cortex
Most Dense Stroma


Stromal Density Gradient in Ovarian Cortex
Less Dense Stroma

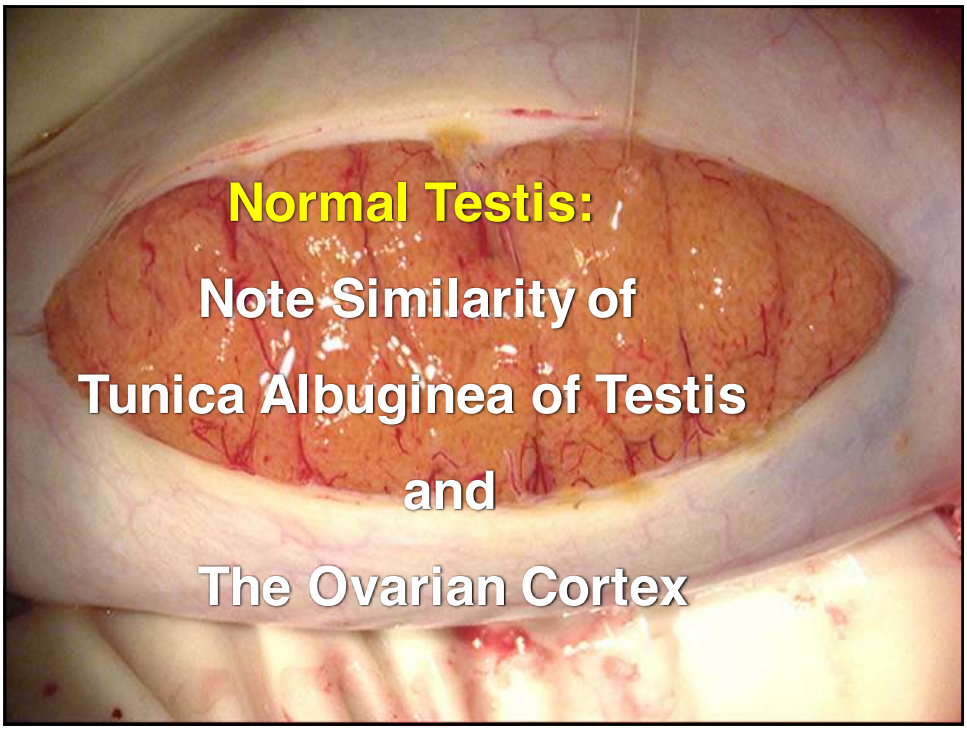



## CONCLUSIONS

- 1) OVARY FREEZING AND TRANSPLANTATION SHOULD NOT BE CONSIDERED EXPERIMENTAL
- 2) A UNIFYING THEORY FOR ADULT RESTING FOLLICLE RECRUITMENT, AND FOR FETAL OOCYTE ARREST.
- 3) WITHOUT A STEADY CONTROLLED RELEASE OF "RESTING" FOLLICLES IN THE ADULT, WOMEN WOULD RUN OUT OF EGGS PREMATURELY.
- 4) IF FETAL OOCYTE ARREST DID NOT ACCUR AFTER
- MEIOTIC ACTIVATION, THERE WOULD BE NO OOCYTES AT BIRTH.
- 5) IMMEDIATELY AFTER TRANSPLANT THERE IS AN OVER-RECRUITMENT OF PRIMORDIAL FOLLICLES, WITH SUBSEQUENT DEPLETION.
- BUT THEN THE GRAFTS LAST A VERY LONG TIME DESPITE VERY LOW AMH.

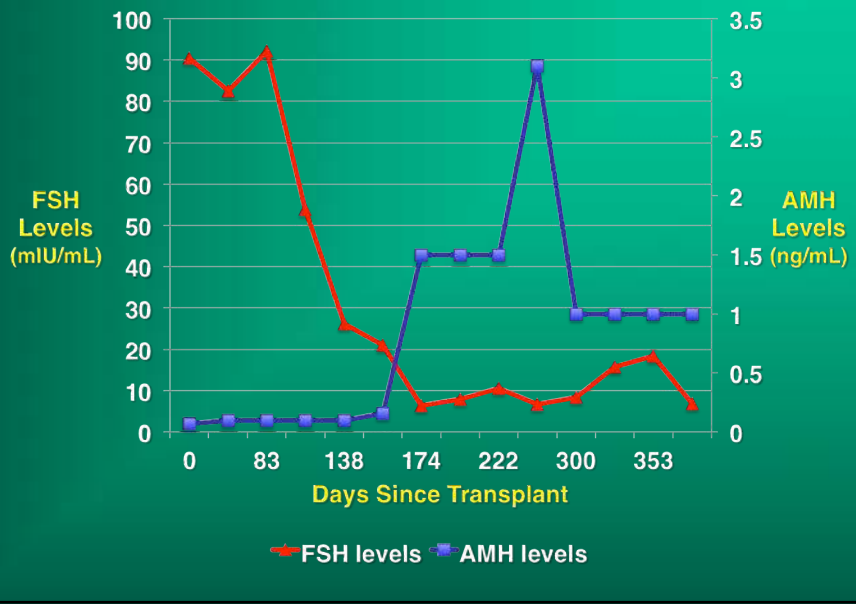
## WHAT CAN THE PRIMORDIAL OOGONIA DO:

TO PREVENT THEMSELVES FROM CONTINUING THROUGH MEIOSIS AND DISAPPEARING BY THE TIME OF BIRTH?



## Leukemia, Tissue Frozen 15 Years Earlier

## FSH AND AMH AFTER FROZEN OVARY TRAN



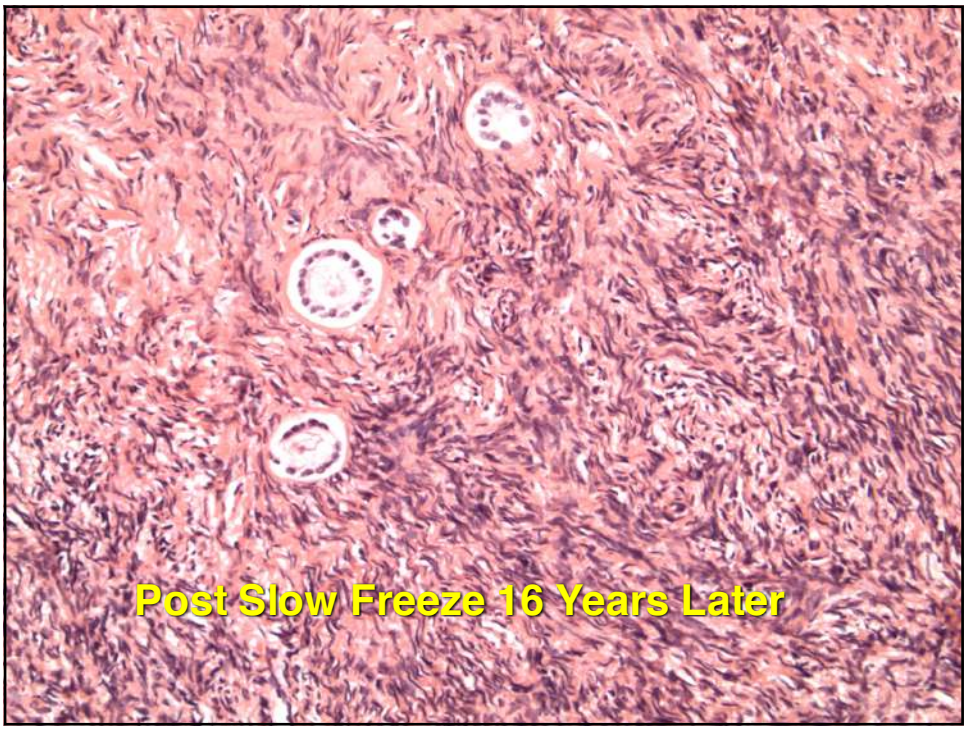

MEDISON
01-26-2016-0002
SCHAIKEWITZ, ... 28y6m
The Childbirth Center
12/06/2015, 7w2d
FPS40D
C2-8
12.0cm
Gen
MI 0.84
TIs 0.1
01-26-2016
05:56:23 PM
Leukemia Case: SECOND pregnancy
6 weeks 5 days, January 26, 2016
CRL 0.82cm
GA 6w5d±4d
Pctl. 24.22
EDD 09-15-2016


Baby Girl Born 9/16/16
7lbs, 1oz, apgar 9

## Leukemia-January 1997: 24 Years Old

## Cured and Married October 2012: 41 Years Old Now


Pre-Slow Freeze in 1997


Post Slow Freeze 16 Years Later

## Autologous Ovary Transplant O' Brien

| | Recipient | Donor |
|---|---|---|
| Pre-BMT HLA Molecular Typing Results | HLA-A2<br>HLA-A3<br>HLA-B7<br>HLA-B60<br>HLA-DRB1* 0401<br>HLA-DRB1* 1501<br>HLA-DQB1* 0301<br>HLA-DQB1* 0602<br>HLA-Cw7<br>HLA-Cw-- | HLA-A2<br>HLA-A3<br>HLA-B7<br>HLA-B60<br>HLA-DRB1* 0401<br>HLA-DRB1* 1501<br>HLA-DQB1* 0301<br>HLA-DQB1* 0602<br>HLA-Cw--<br>HLA-Cw-- |
| ABO/RH Blood Type | A+ | B- |

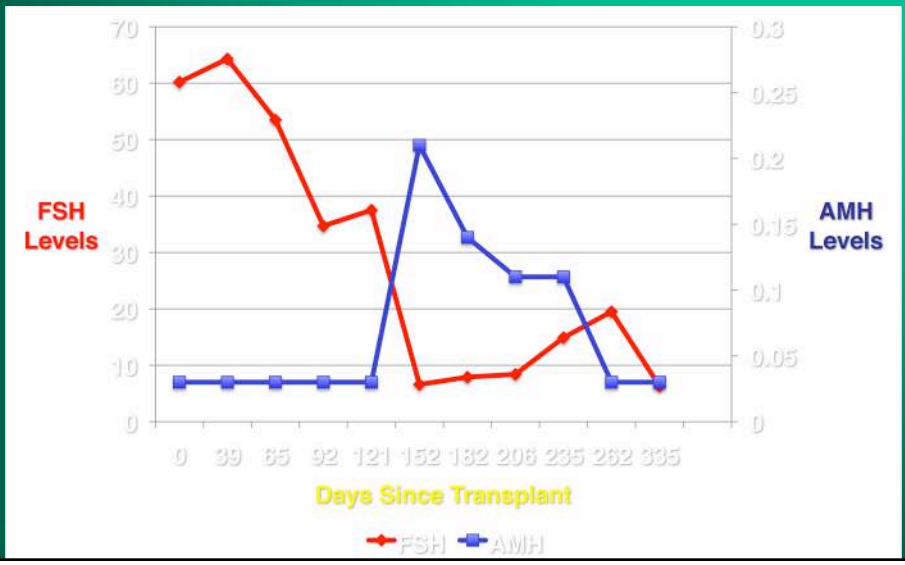
## Autologous Ovary Transplant O' Brien

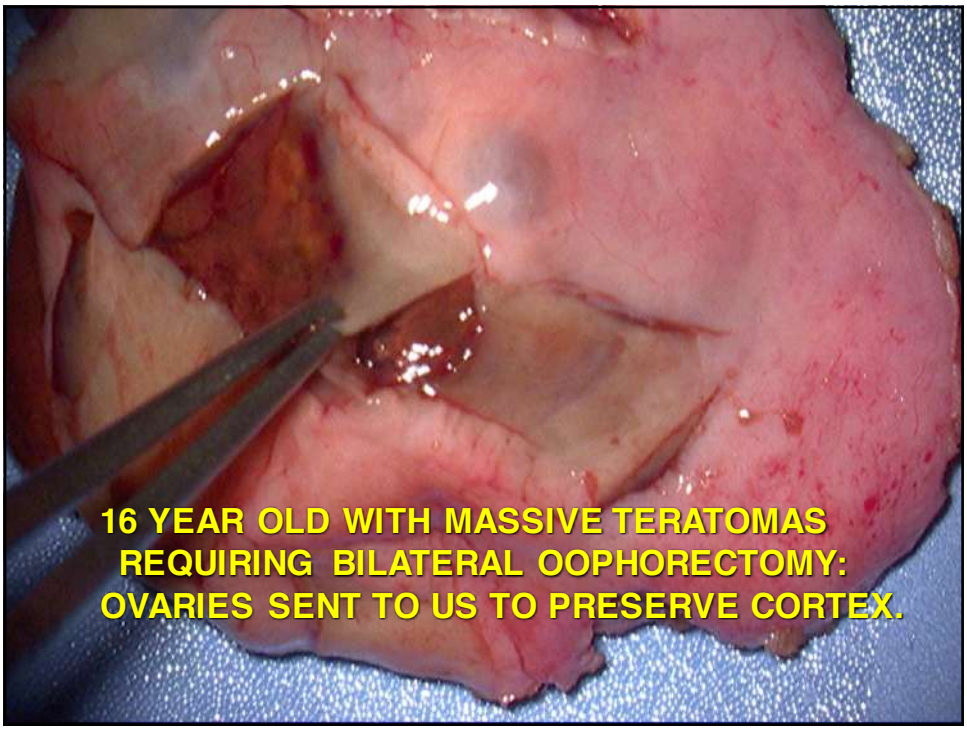
| | Recipient | Donor |
|---|---|---|
| Diagnosis | Acute Lymphocytic Leukemia | |
| AFC | Not done | |
| Date of OTF | 7/17/97 – Patient had chemo prior to freeze | |
| Type of Freeze | Slow | |
| Date of BMT | 9/27/97 | |
| Date of BMT | 6/17/99 | |
| After 2nd BMT | Patient showed some signs of GVHD | |
| Post-BMT HLA Molecular Typing Results | Not Done | Not Done |
| ABO/RH Blood Type | B- | B- |
| Date of Ovary Transplant | 10/7/13 | |

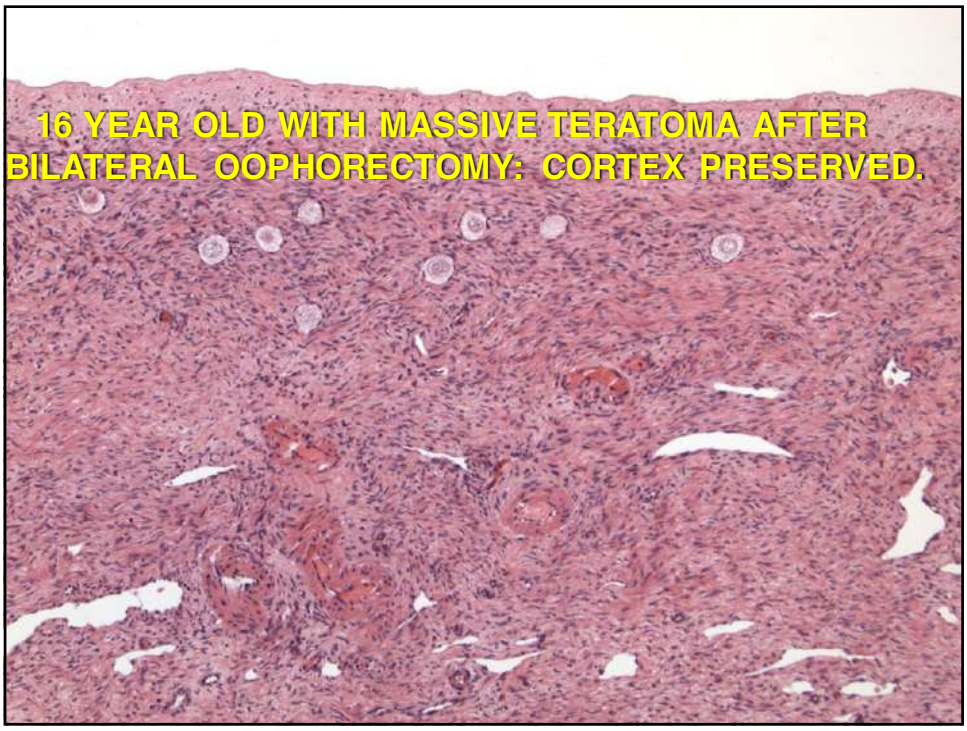
## September 2013

Transplantation of thawed and quilted ovarian slices

## TRANSPLANT OF THAWED OVARIAN TISSUE
October 7, 2013 (24 Y.O. TISSUE IN 40 Y.O.)
Our very 1st ovary freeze 1996





16 YEAR OLD WITH MASSIVE TERATOMAS
REQUIRING BILATERAL OOPHORECTOMY:
OVARIES SENT TO US TO PRESERVE CORTEX.


16 YEAR OLD WITH MASSIVE TERATOMA AFTER
BILATERAL OOPHORECTOMY: CORTEX PRESERVED.


14 YEAR OLD GIRL OSTEOGENIC SARCOMA-
NEVER SAW PATIENT–
OVARY SENT OVERNIGHT FROM TEXAS:
(ANDERSEN APPROACH)


Ovary Transplant
Healthy Women

## OTF Breast Cancer Patient 35 y.o. And "not yet ready to have children"

EVEN IF SHE DID NOT HAVE CANCER SHE WILL BE INFERTILE SOON

## SO OBVIOUSLY OVARY FREEZING AND TISSUE GRAFTING WORKS

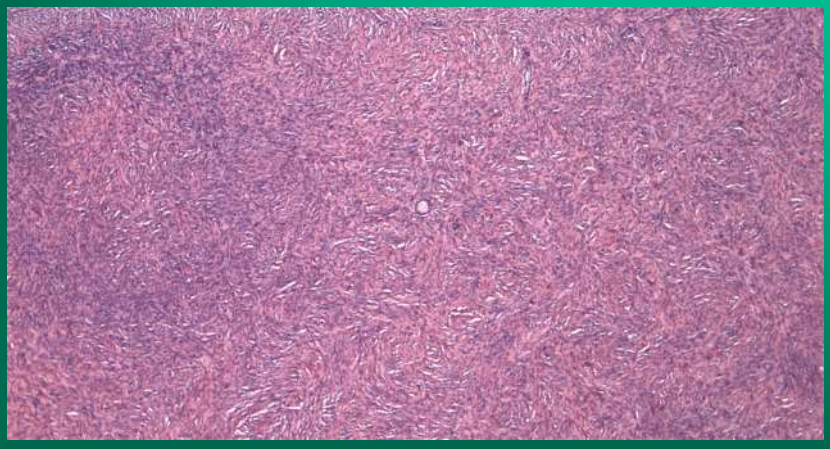
FOR ASRM TO RULE THIS AS "EXPERIMENTAL" FOR CANCER PATIENTS IS NO LONGER APPROPRIATE.

## MASSIVE PRIMORDIAL FOLLICLE RECRUITMENT

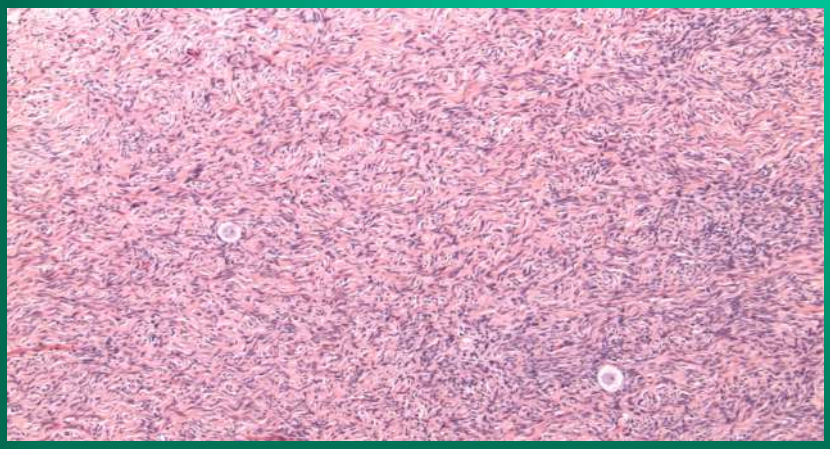
DUE TO SUDDEN DECREASE IN STROMAL TISSUE PRESSURE WHICH IS THE MECHANISM OF OOCYTE ARREST.

## "Dead" Tissue in Menopausal Women ?

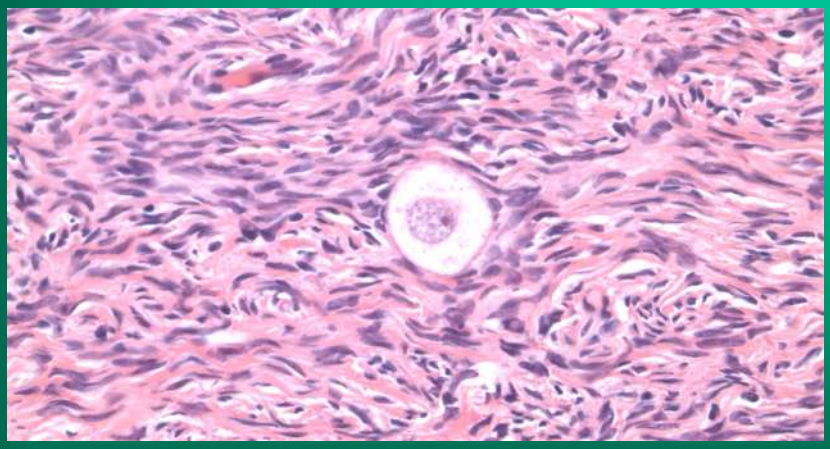
## There Are Usually A Few Eggs Trapped in The Cortex of Menopausal Women with POF



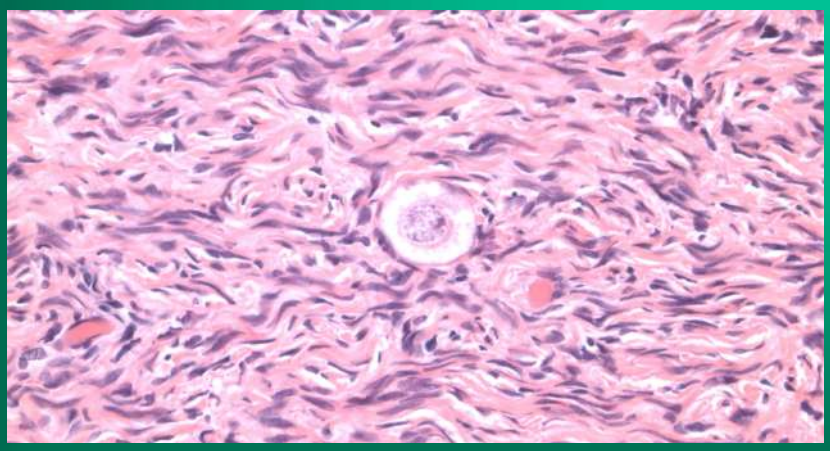
## Or Even Several Eggs



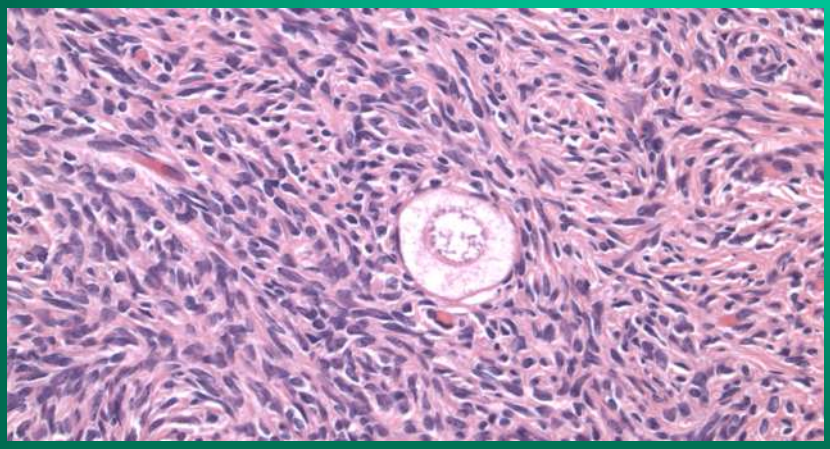
## Occasional Egg



## Occasional Egg



## Good Quality Occasional Egg



# CONCLUSIONS

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- MEIOTIC ACTIVATION, THERE WOULD BE NO OOCYTES AT BIRTH.

# Autotransplant For Premature Ovarian Failure

- Our main effort has been to avoid premature ovarian failure with ovarian tissue freezing.
- However, what if the patient never had her ovarian tissue frozen?
- If we remove her existing "dead" cortex, and re-implant it back onto her medulla, can we release these few trapped resting follicles,
- and allow them to be recruited for IVF or even ovulation and spontaneous pregnancy.

## A Great Egg In A MenopausalWoman With POF