A Combination of Simvastatin and Methylprednisolone Improve the Quality of Vitrified-Warmed Ovarian Tissue after Transplantation

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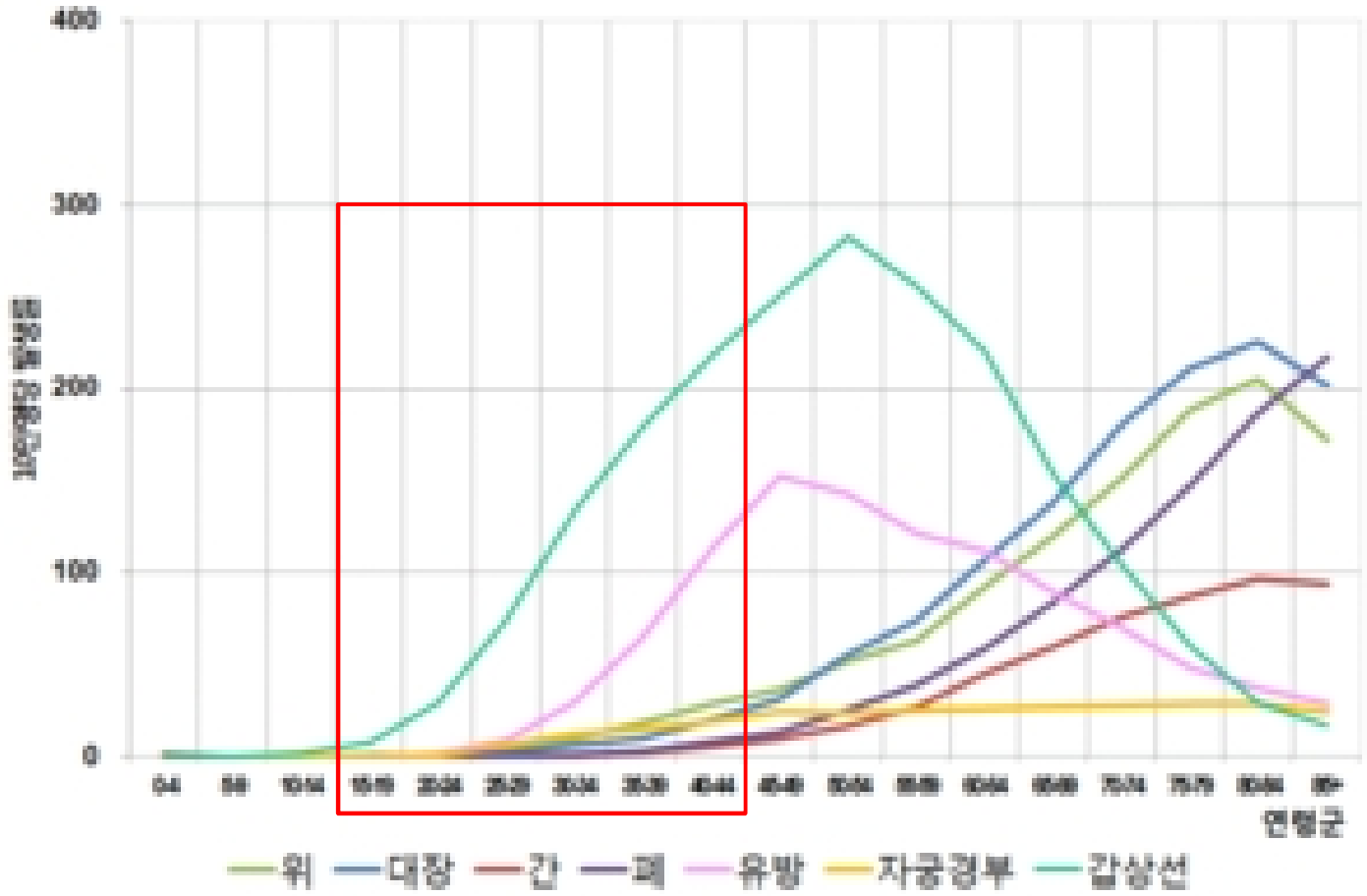
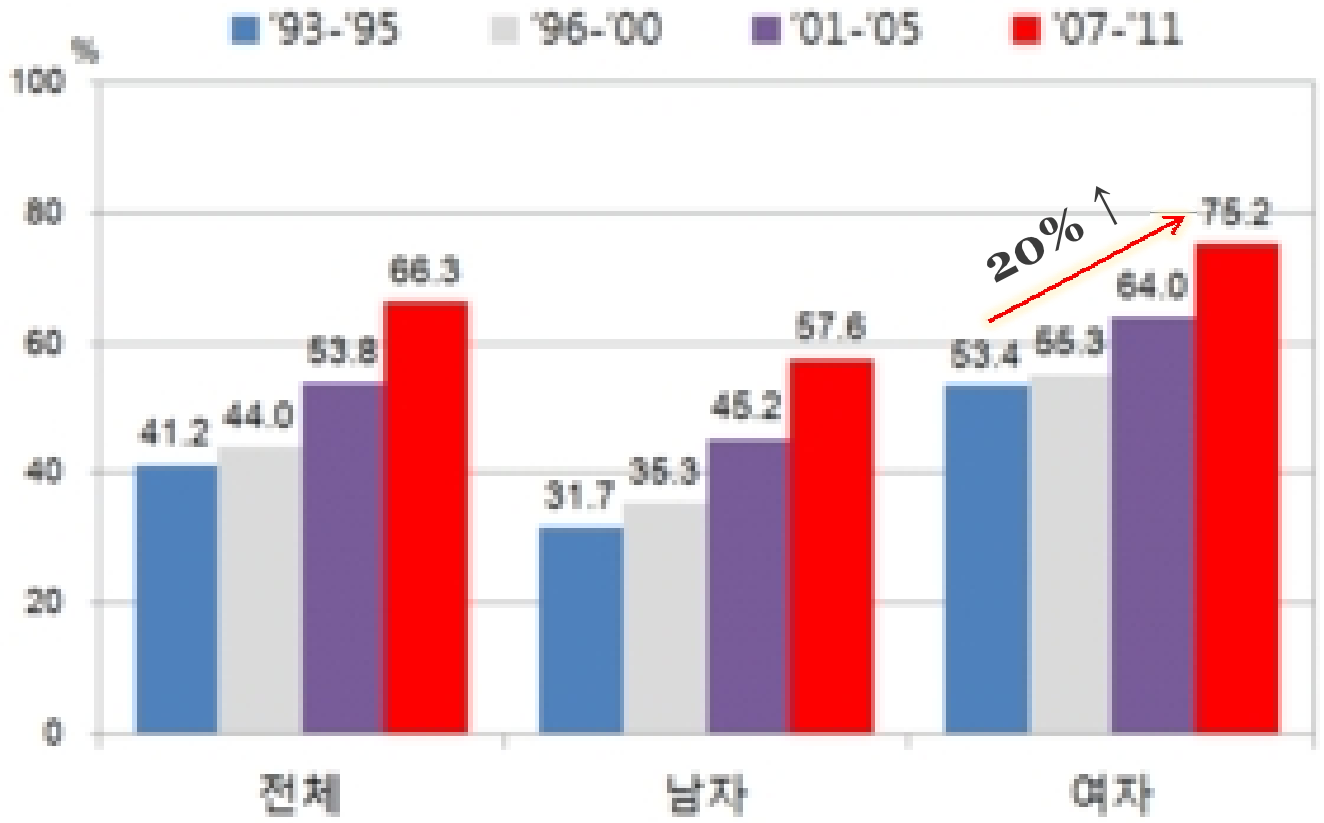
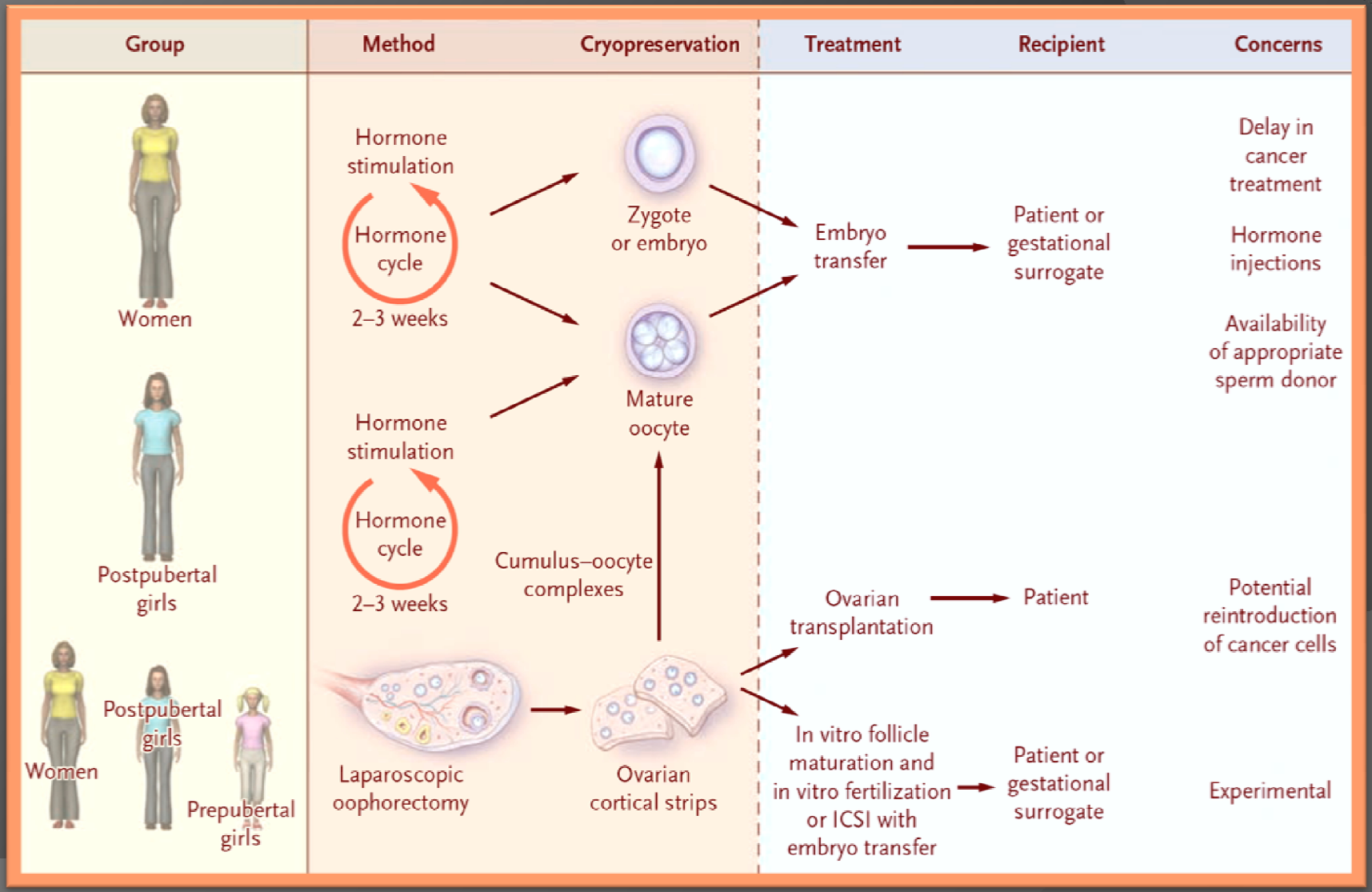
주요 암종 연령군별 발생률: 여자, 2011



성별 5년 생존율 추이: 모든 암



Options for Fertility Preservation in Female Cancer Patients



J.S.Jeruss et al., N Engl J Med, 2009

Problems during Ovarian Tissue Cryopreservation & Transplantation

- There are many **hurdles** during ovarian tissue (OT) cryopreservation and transplantation
  - Cryodamage (Cryopreservation)
  - **Ischemic injury (Transplantation)**
  - Re-implantation of malignancy cells (After transplantation)

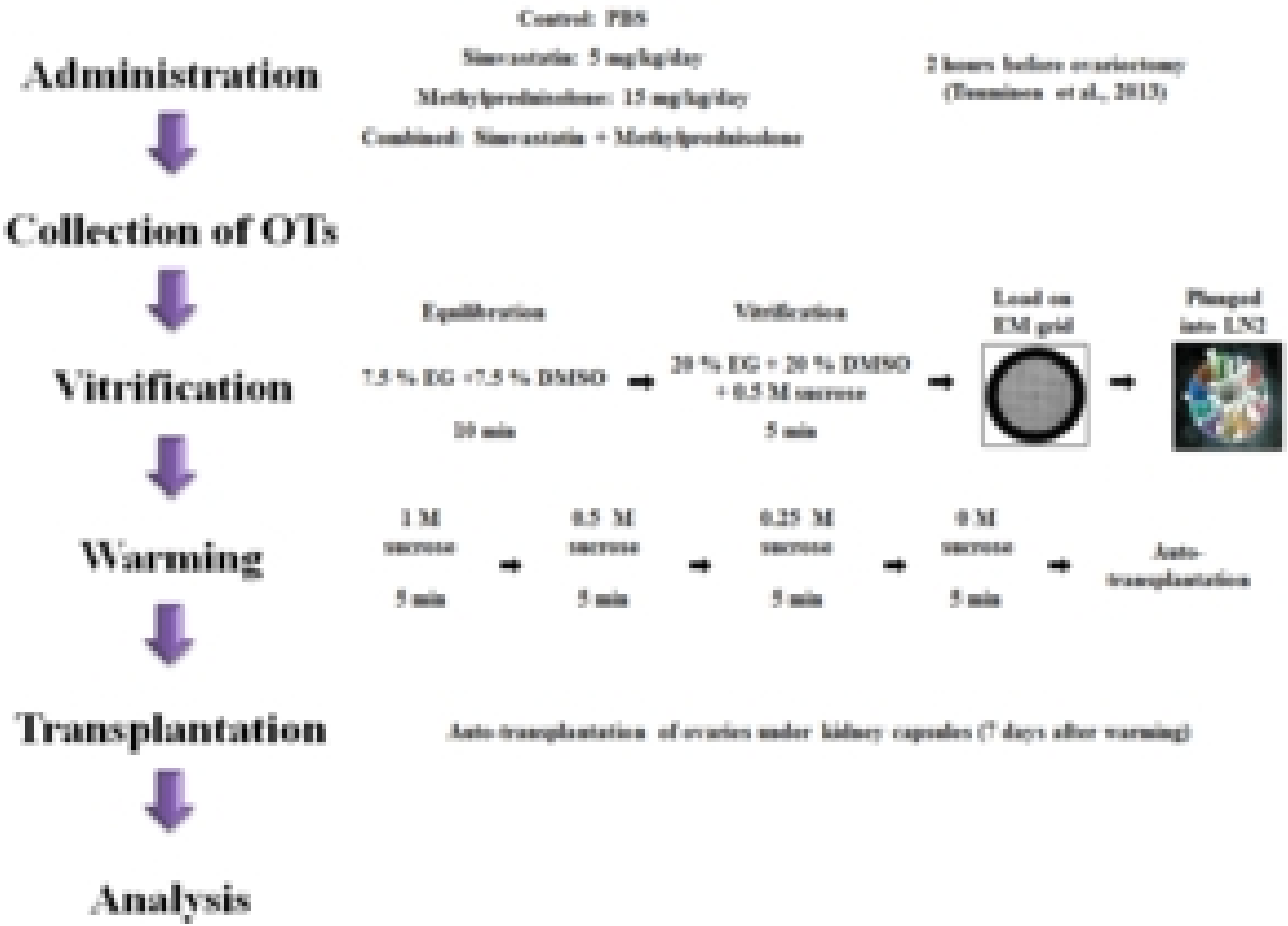
- In general, after transplantation, ovarian revascularization occurs and numerous ovarian follicles are lost during the ischemic period (Israely et al., 2003, Jeremias et al., 2002)

- At least 25% of the primordial follicles are lost as a result of transplantation (TPL) of cryopreserved xenografts of human OT into mice (Newton et al., 1996).

- Therefore, **reducing the ischemic damage** during OT TPL is **important** for **successful tissue TPL.**

	Simvastatin	Methylprednisolone
Chemical Structure		
Main Function	·HMG-CoA reductase inhibitor	·Corticosteroid
Main Targets	·Hyperlipidemia ·Coronary artery disease	·Arthritis ·Auto-immunedisease
Other Effects	·Angiogenesis ·Anti-inflammation ·Inhibition of apoptosis	·Decrease nitric oxide (NO)

Materials & Methods



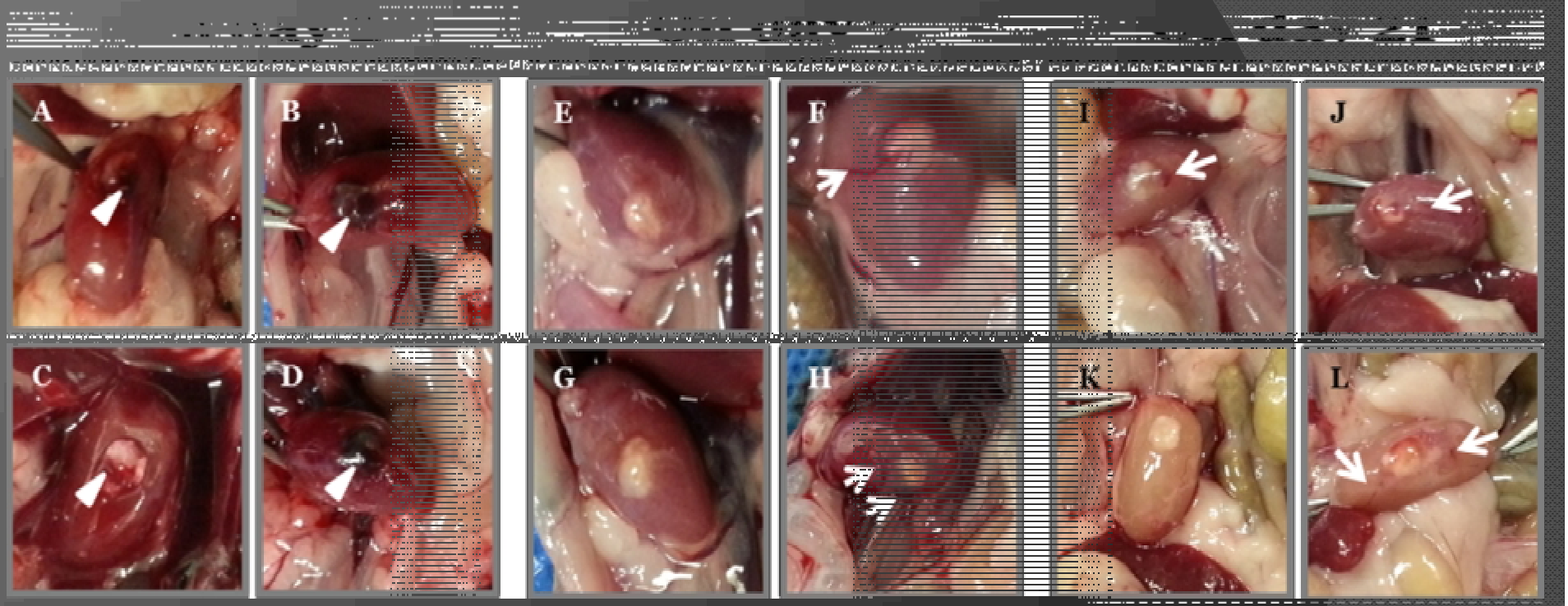
Materials & Methods

- **Animal**
  - ✓ 6-week-old aged BD F-1 mice
  - ✓ Sacrifice on **day 2, 7 and 21** after transplantation

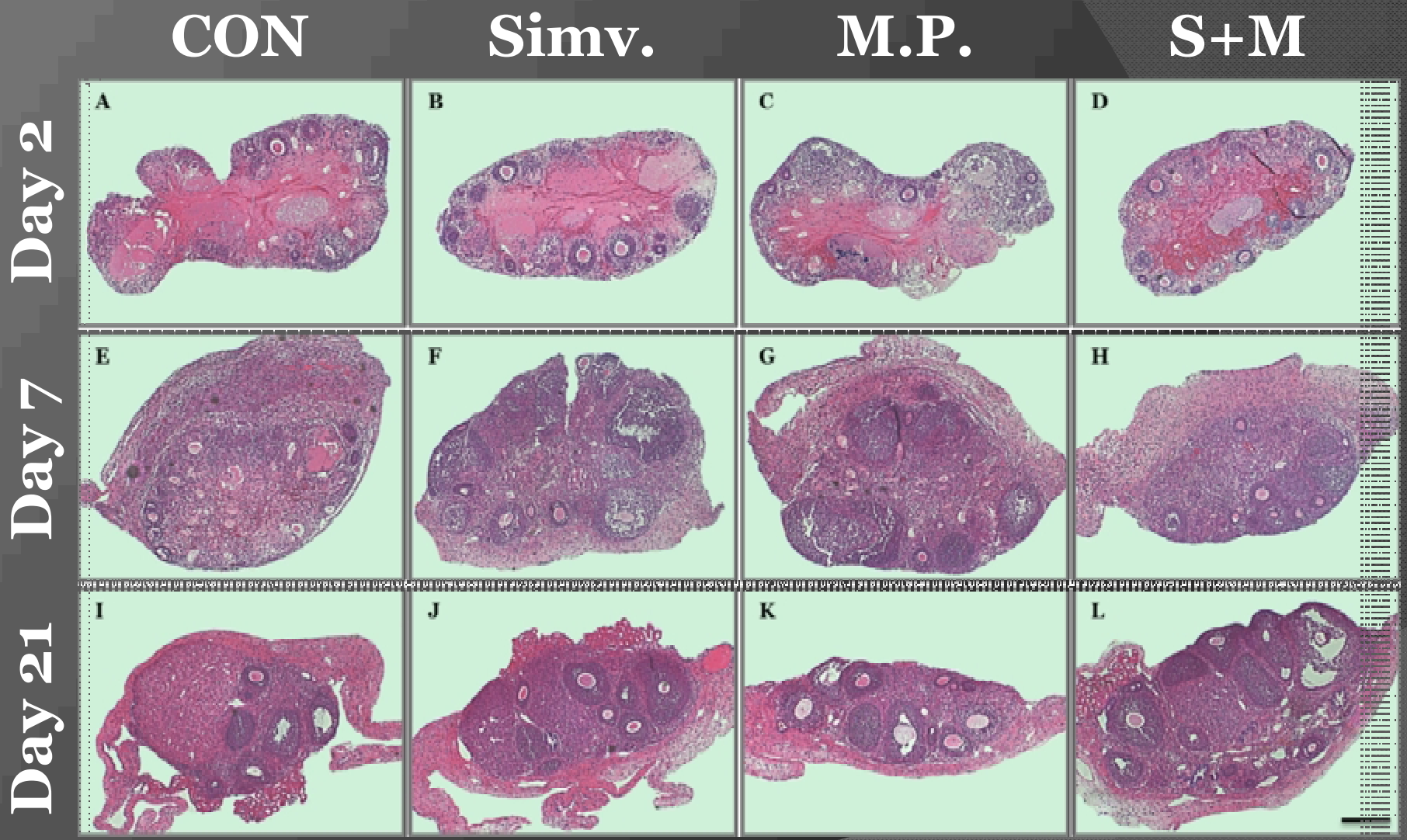
- **Assessment Methods**
  - ✓ Gross Observation
  - ✓ Histological Analysis – **H&E Stain**
  - ✓ Follicle Classification – **Grade 1 Follicle**
  - ✓ Apoptosis – **TUNEL Assay**
  - ✓ Vessel Density – **IHC for CD31**
  - ✓ Immune Cell Infiltration – **Flow Cytometry for CD45**
  - ✓ Ovarian Reserve – **ELISA for Serum AMH**
  - ✓ Embryonic Development – **OPU, IVF and DST**

Results

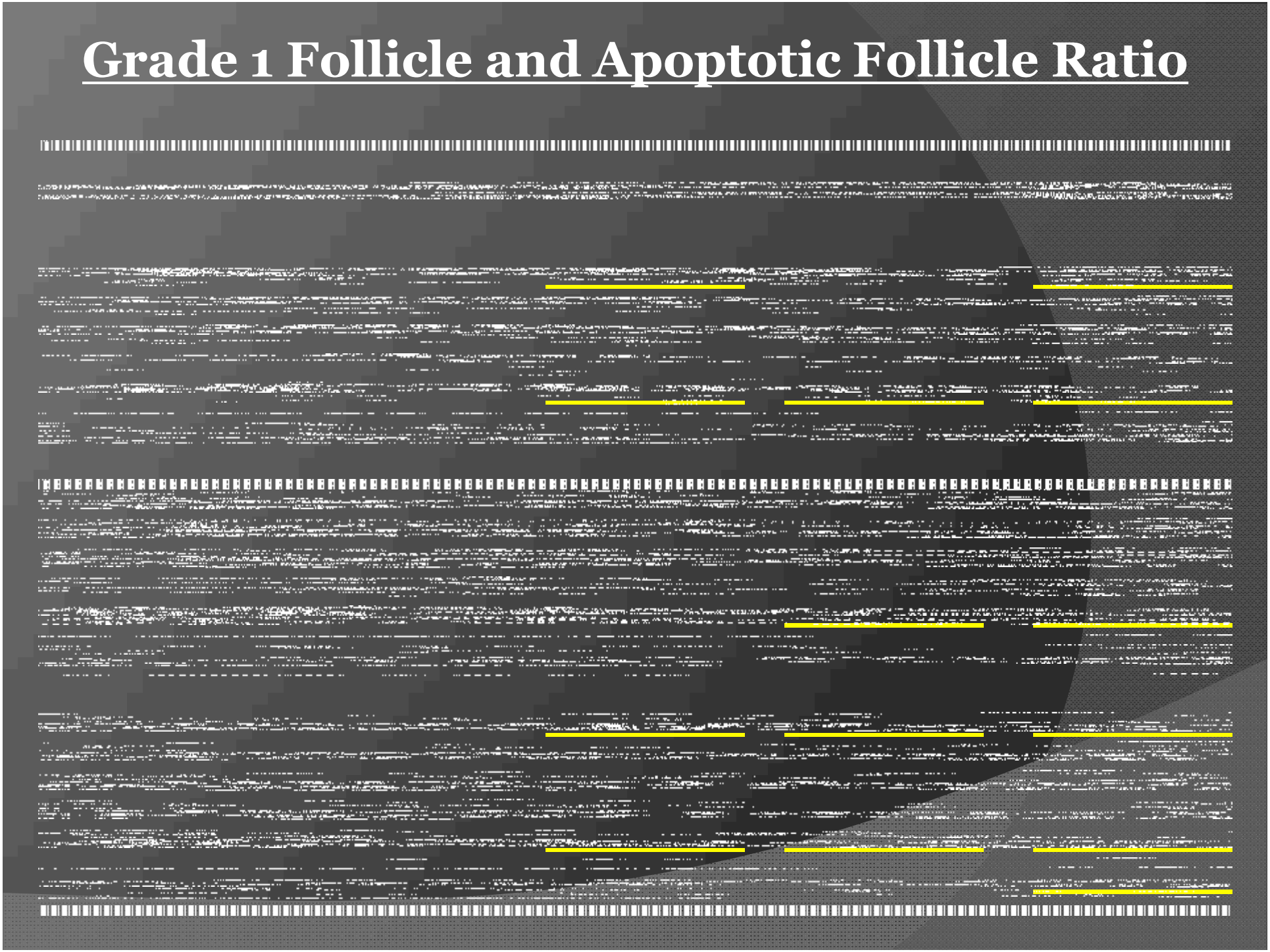
Gross Observation



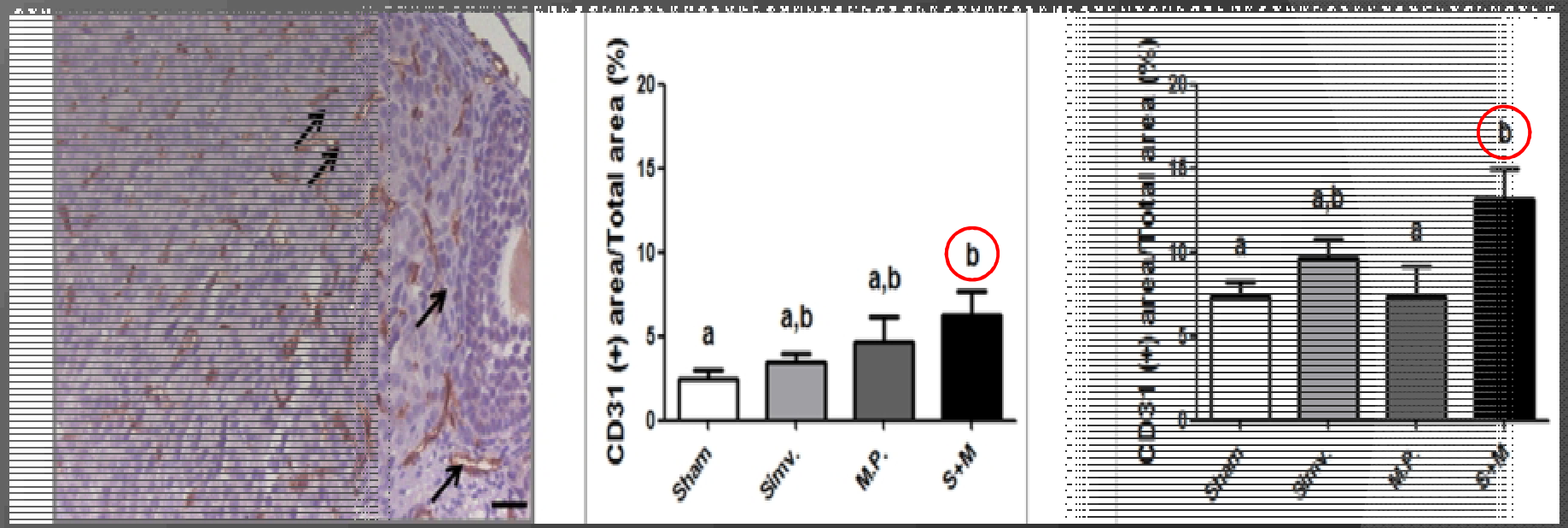
Histological Assessment



Grade 1 Follicle and Apoptotic Follicle Ratio

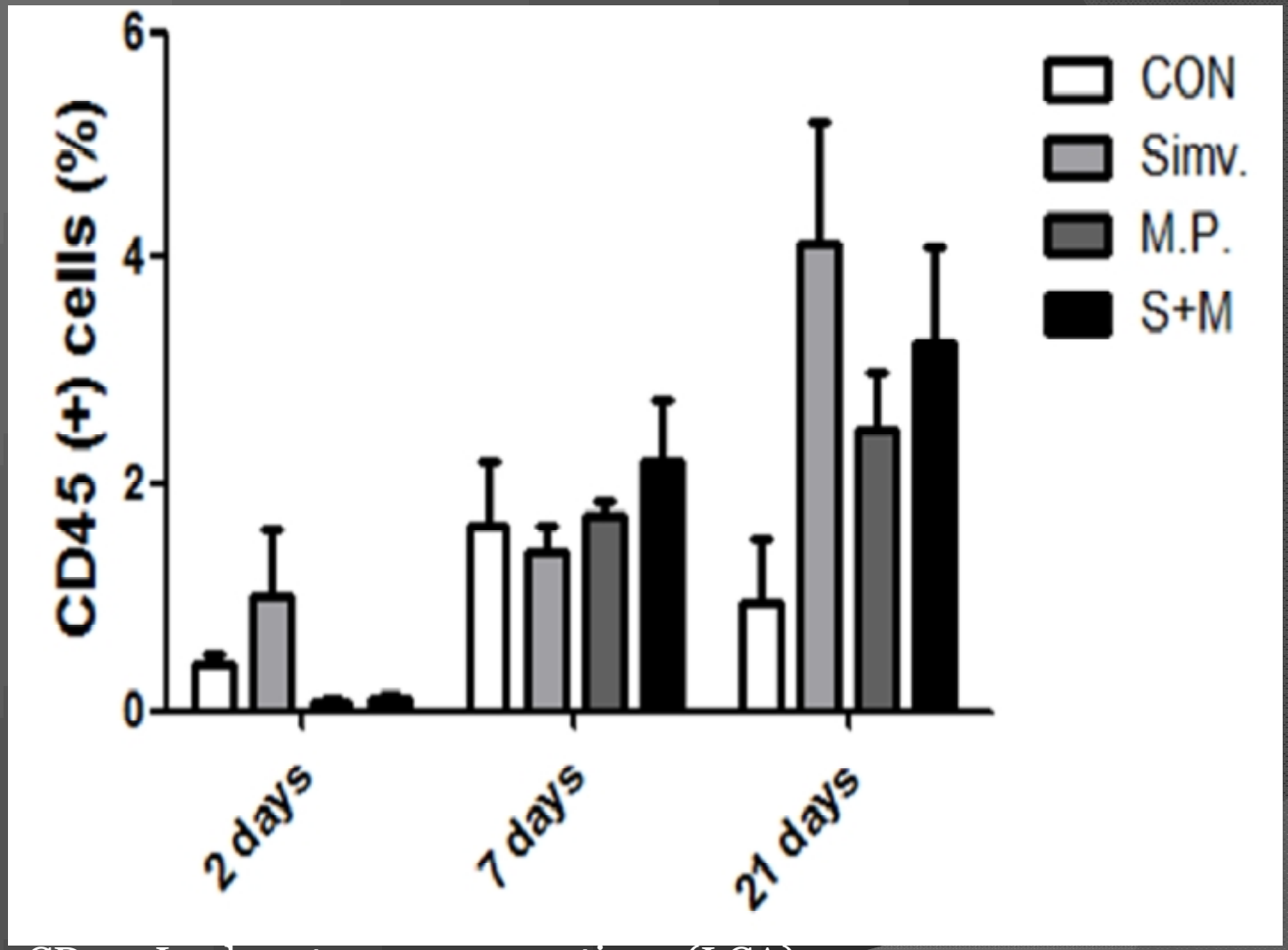


The Proportion of CD31 Area



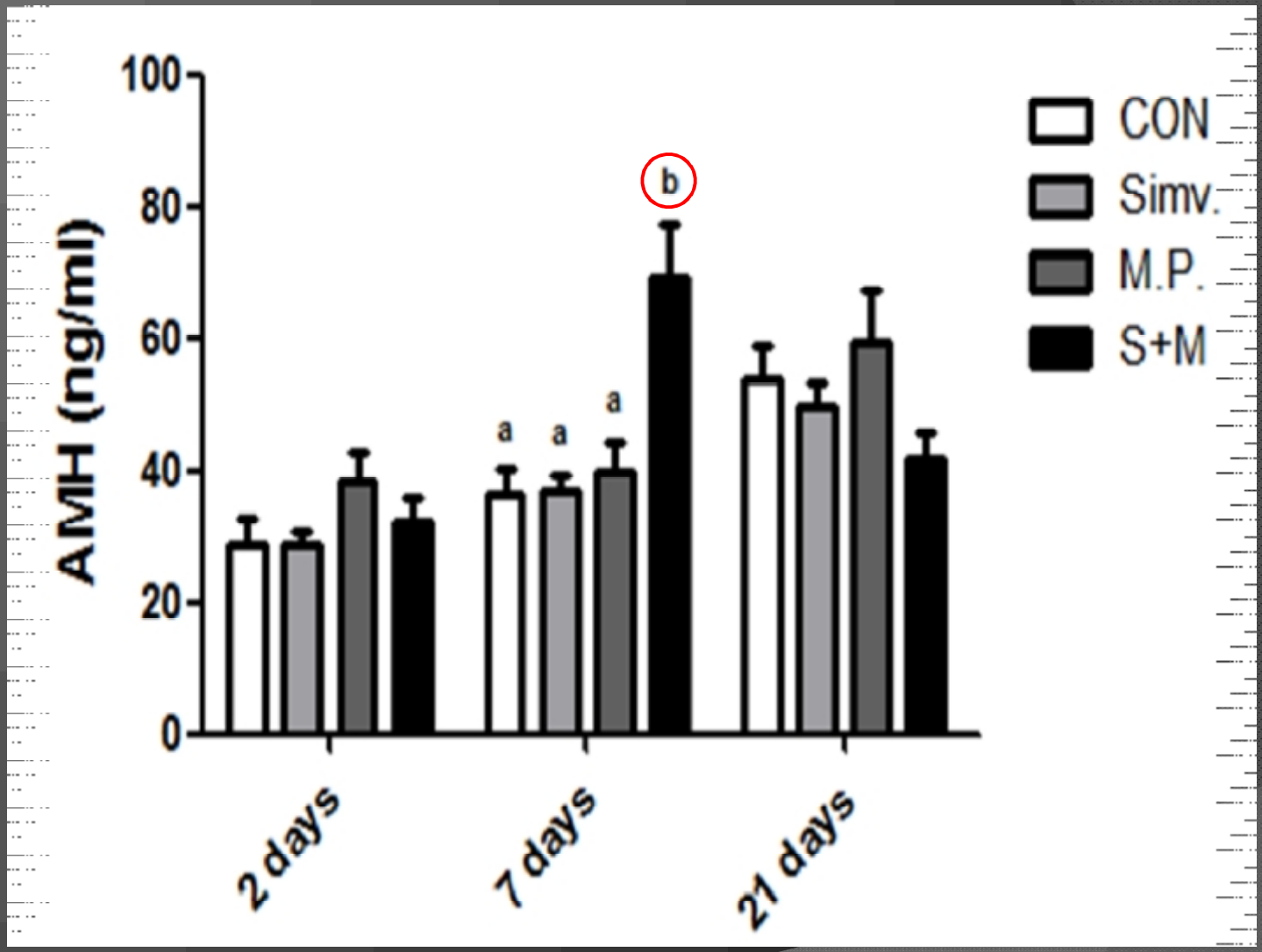
Blood Vessels

The Proportion of CD45 Cells



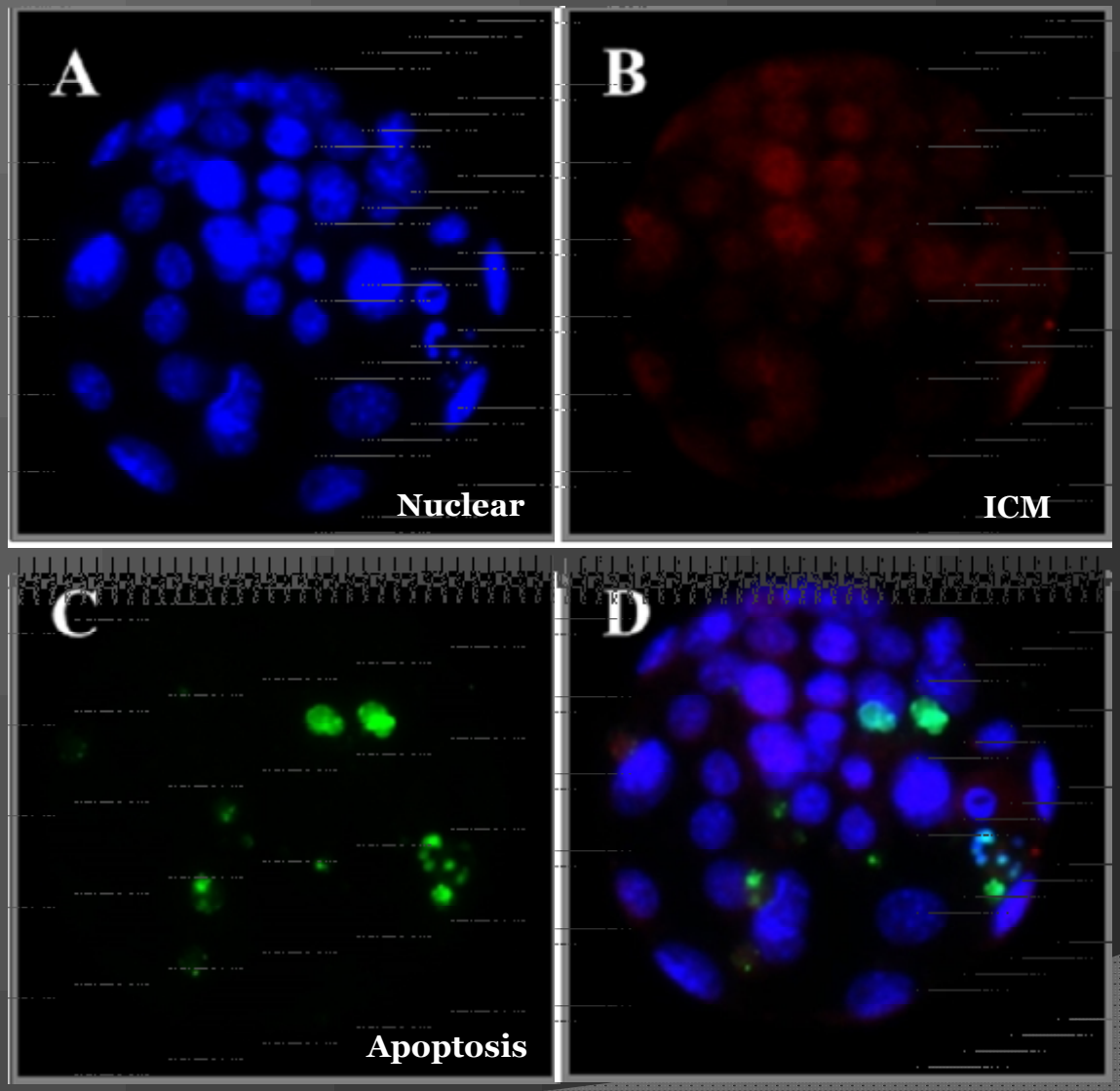
CD45: Leukocyte common antigen (LCA) De Vitro et al., Int J Mol Med (2012)

Serum AMH Levels



Anti-Mullerian Hormone (AMH): A biomarker for ovarian reserve

Differential Staining



A: Hoechst33342
B: Anti-Oct4
C: TUNEL
D: Merged

Embryonic Development after IVF

ICM/TE (ea)	7.9±1.9/ 21.5±1.7	7.3±2.6/ 23.4±2.7	7.9±1.9/ 25.4±2.6	9.1±2.4/ 24.9±2.5

Conclusion

- Treatment with combined Simvastatin and Methylprednisolone enhances the quality of ovarian tissues and reserve quantity.
  - ✓ **Primordial G1**
  - ✓ **Apoptotic Follicle**
  - ✓ **CD31-positive area**
  - ✓ **Serum AMH**

- Although we evaluated the beneficial effects of Simvastatin and Methylprednisolone in the present study, we **could not unravel** the corresponding **protective mechanisms**

- This combinatorial treatment can be **clinically applied** to **humans** and **domestic animals** subject to further studies.