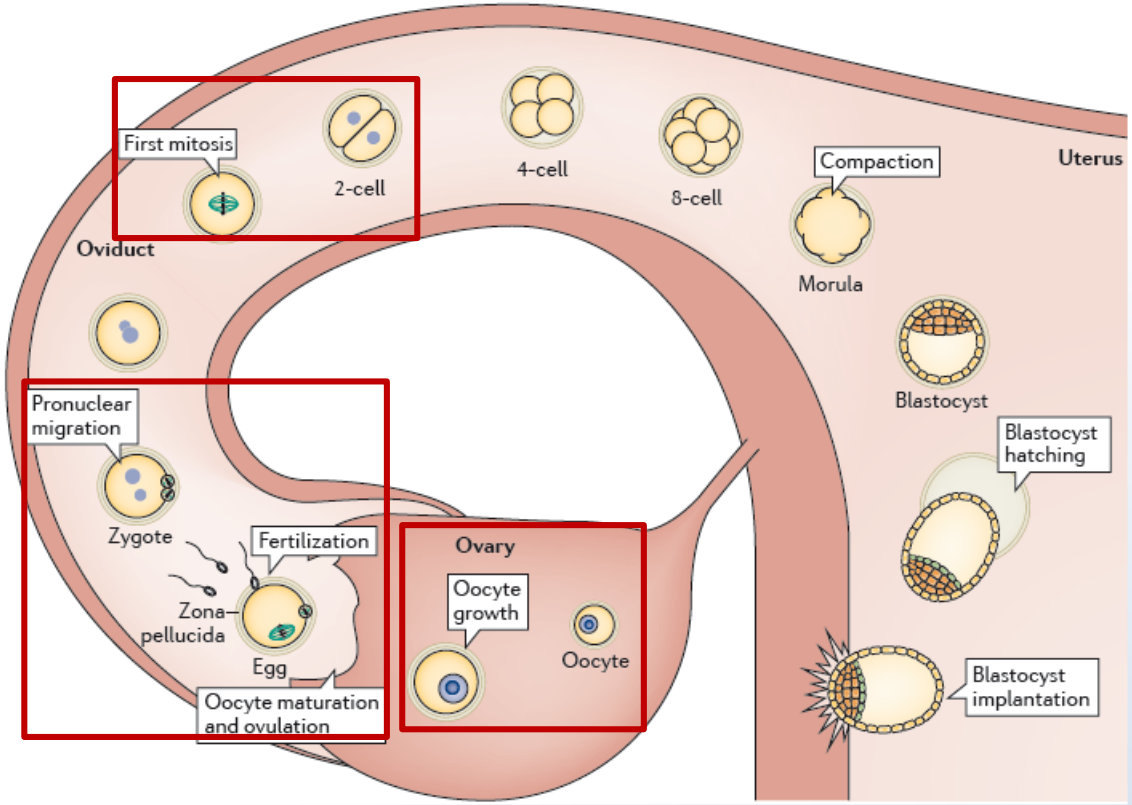
CELL CYCLE REGULATION IN MAMMALIAN OOCYTES AND EARLY EMBRYOS

- **standard somatic cell cycle is modulated to meet the specific requirements of different developmental stages**

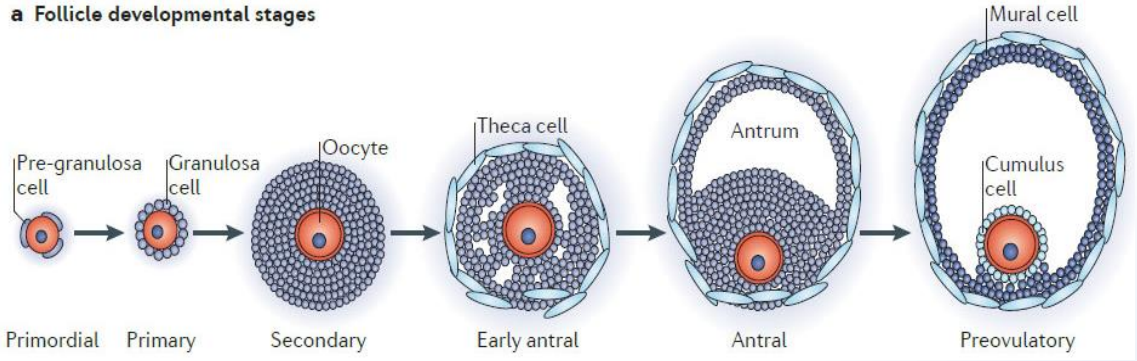


An overview of preimplantation development

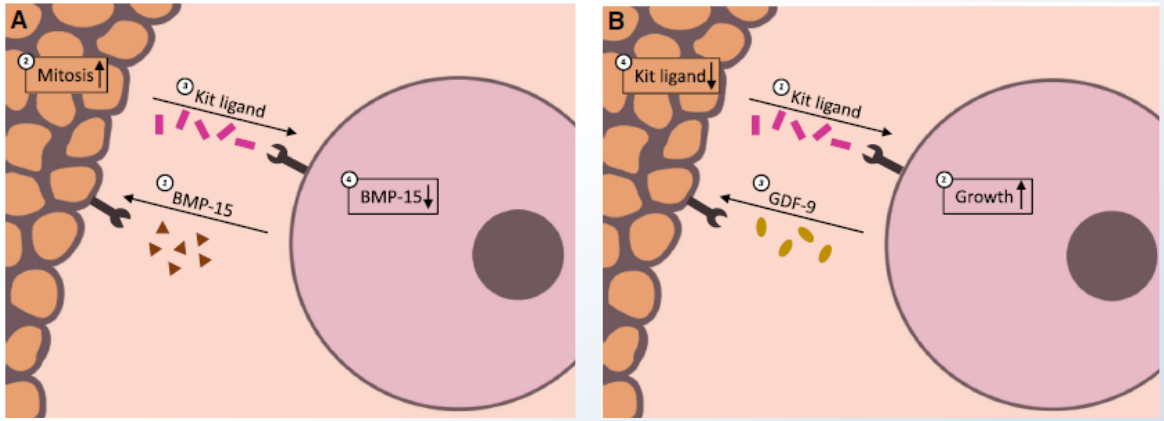
Clift, Schuh, 2013, Nat Rev Mol Cell Biol., PMID: 23942453

- oocyte–somatic cell interactions
- **cell cycle arrests**
- pairing and **recombination** between homologous chromosomes
- **meiotic resumption**
- **MI** - inhibition of sister-chromatid separation
- **MII** - absence of DNA replication
- **meiotic-to-mitotic transition**
- **cell cycle adaptations**

FOLLICULOGENESIS AND OOGENESIS



Li, Albertini, 2013, Nat Rev Mol Cell Biol., PMID: 23429793

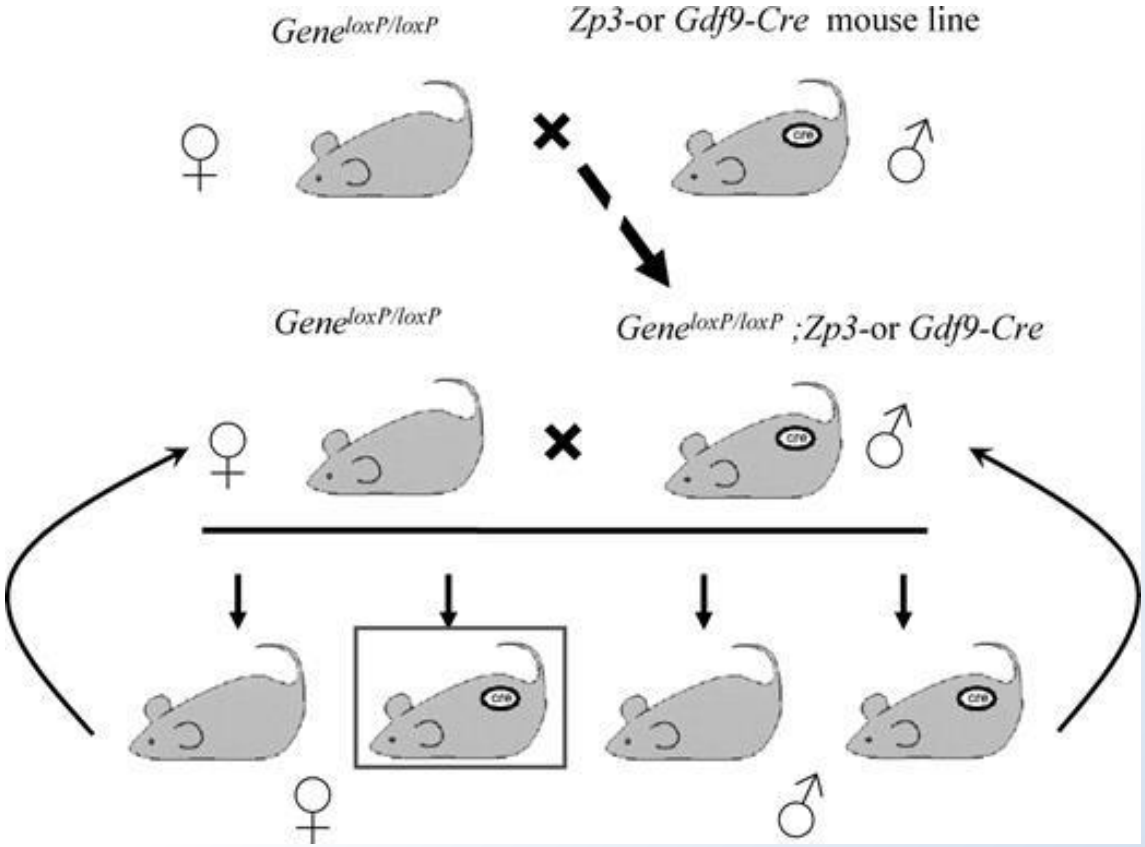


Oocyte can initiate, modulate, and terminate follicle growth and maturation

Jones, Shikanov, 2019, J Biol Eng., PMID: 30647770

- oocyte–somatic cell interactions
- **gonadotropin-dependent** antral follicle growth

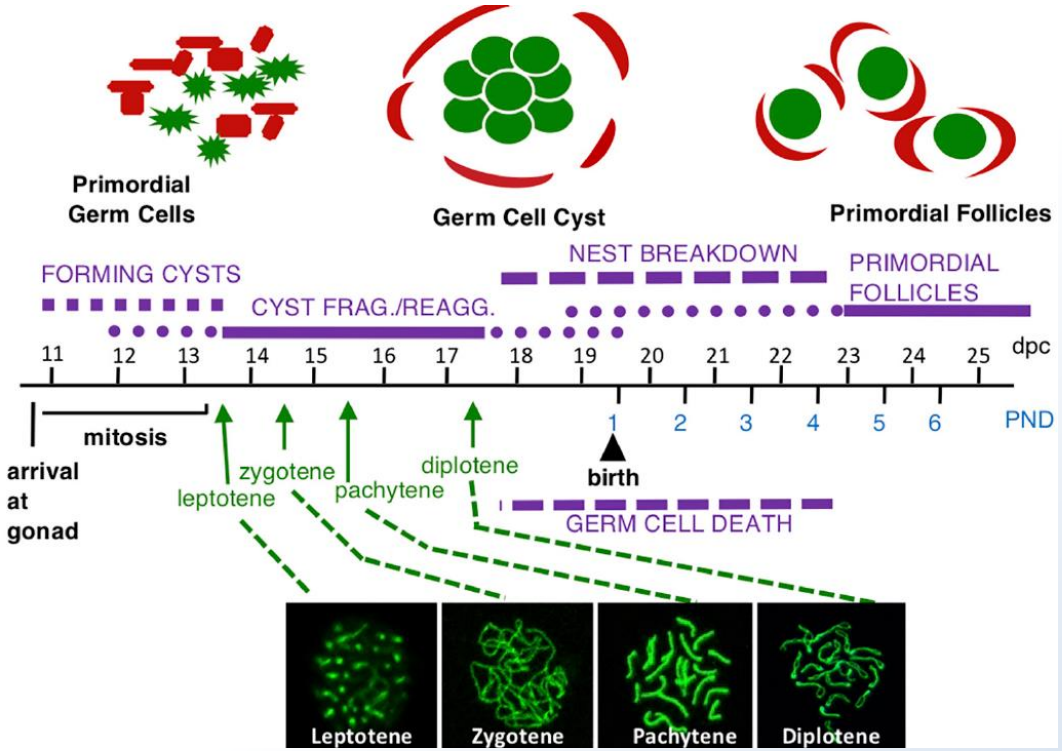
GENERATION OF OOCYTE-SPECIFIC GENE KNOCKOUT MOUSE LINES



Sun et al, 2008, Biol Reprod., PMID: 18753607

- **Cre-recombinase** expression is **driven by** oocyte-specific zona pellucida 3 **(*Zp3*)** promoter or growth differentiation factor 9 **(*Gdf9*) promoter**

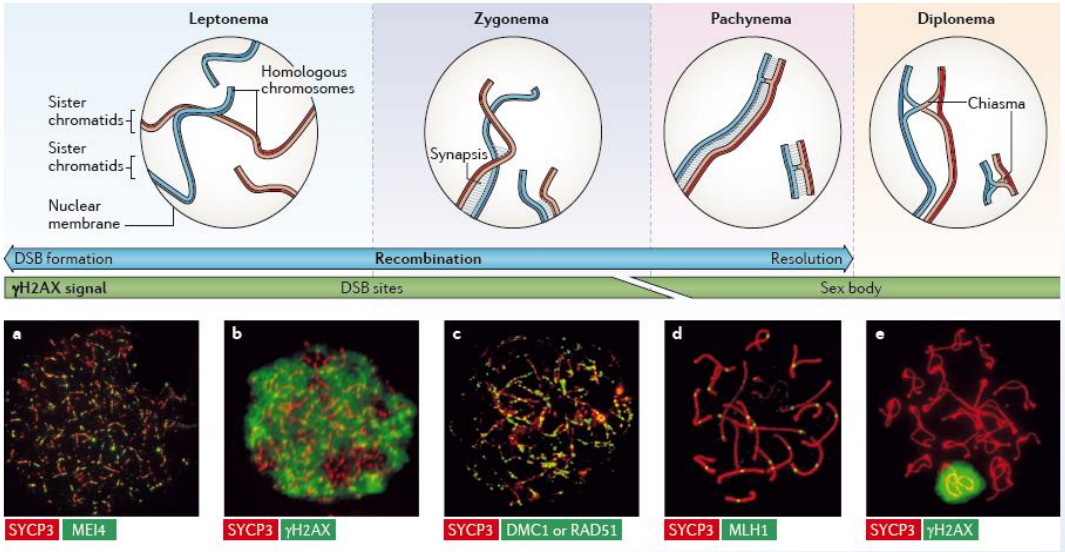
CELL CYCLE ARREST - MEIOTIC PROPHASE I IN MAMMALIAN OOCYTES



Wang, Pepling, 2021, Front Cell Dev Biol., PMID: 34095134

- **fetal development**
- at postnatal day 5, most of mouse oocytes have reached the late diplotene (in humans from 8- to 28-week of fetal development)
- mammalian oocytes are **arrested at prophase I until puberty**

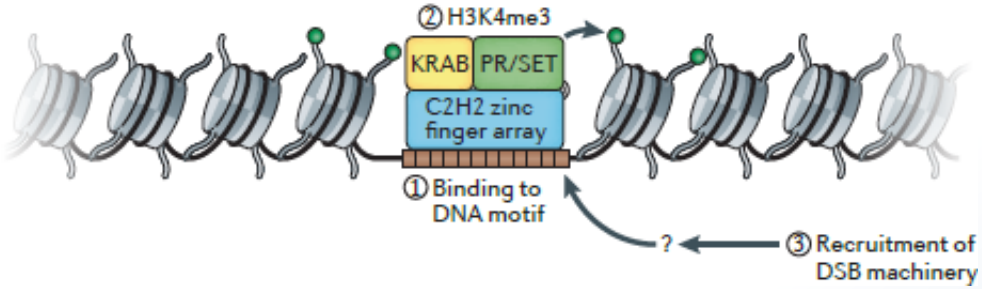
MEIOTIC RECOMBINATION DURING MEIOTIC PROPHASE I IN MAMMALIAN OOCYTES



Baudat et al, 2013, Nat Rev Genet., PMID: 24136506

- **programmed induction of DNA double-strand breaks** (**DSBs**) leading to the exchange of genetic material between homologous chromosomes
- essential for **genome diversity** and **proper chromosome segregation**
- synaptonemal complex - **SYCP3**
- **RAD51, DMC1** – DNA recombinases – **DSB repair** – 200-400 foci/cell
- **MEI4** – DSB induction
- **MLH1** – crossover

REGULATION OF MEIOTIC RECOMBINATION IN MAMMALS

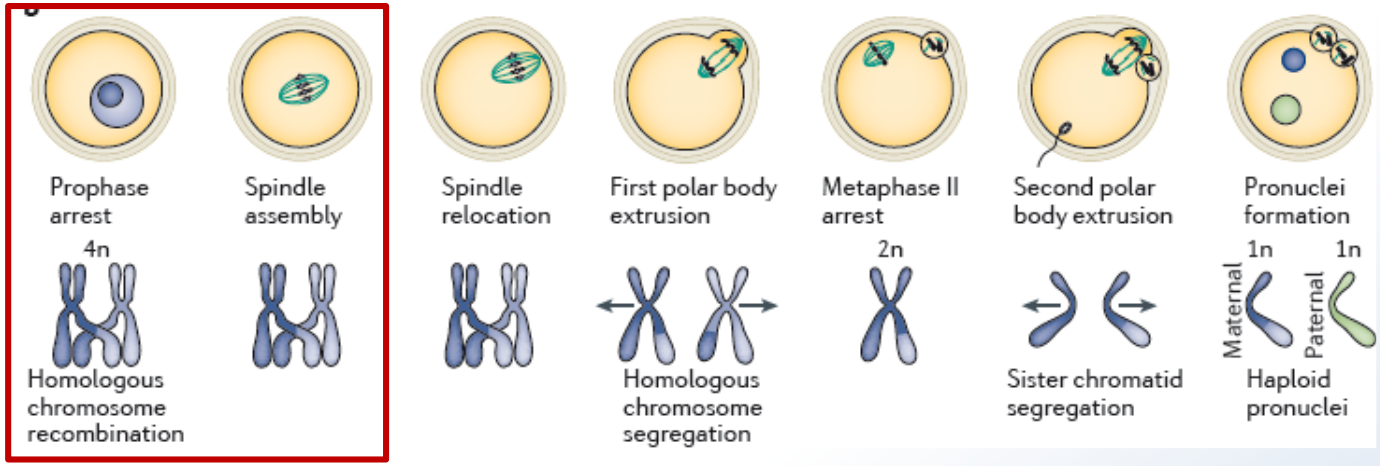


Model for the role of PRDM9 in meiotic DSB localization

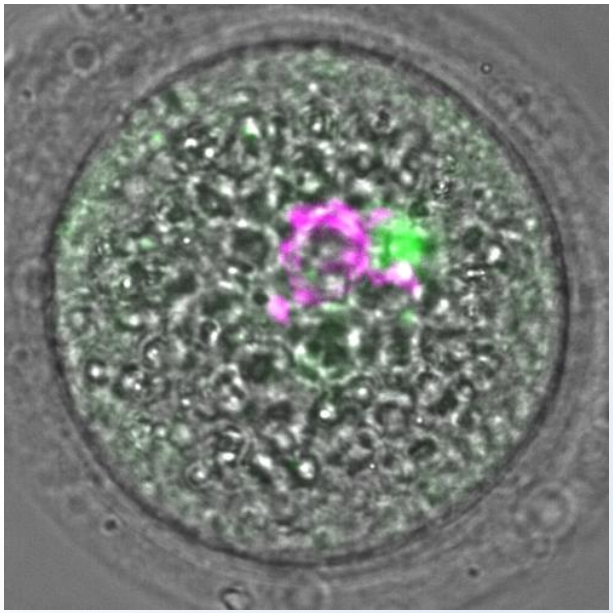
Baudat et al, 2013, Nat Rev Genet., PMID: 24136506

- **PRDM9 –** sequence-specific DNA-binding histone methyltransferase
- several domains
- SPO11 recruitment
- **„hotspots“ –** 1-2 kbp - short chromosomal intervals with C2H2 zinc finger array
- **more than 40 PRDM9 alleles in humans**
- more than 25,000 crossover hot spots in humans
- **evolution** of hotspots and **hotspot paradox**

MAMMALIAN OOCYTE MATURATION

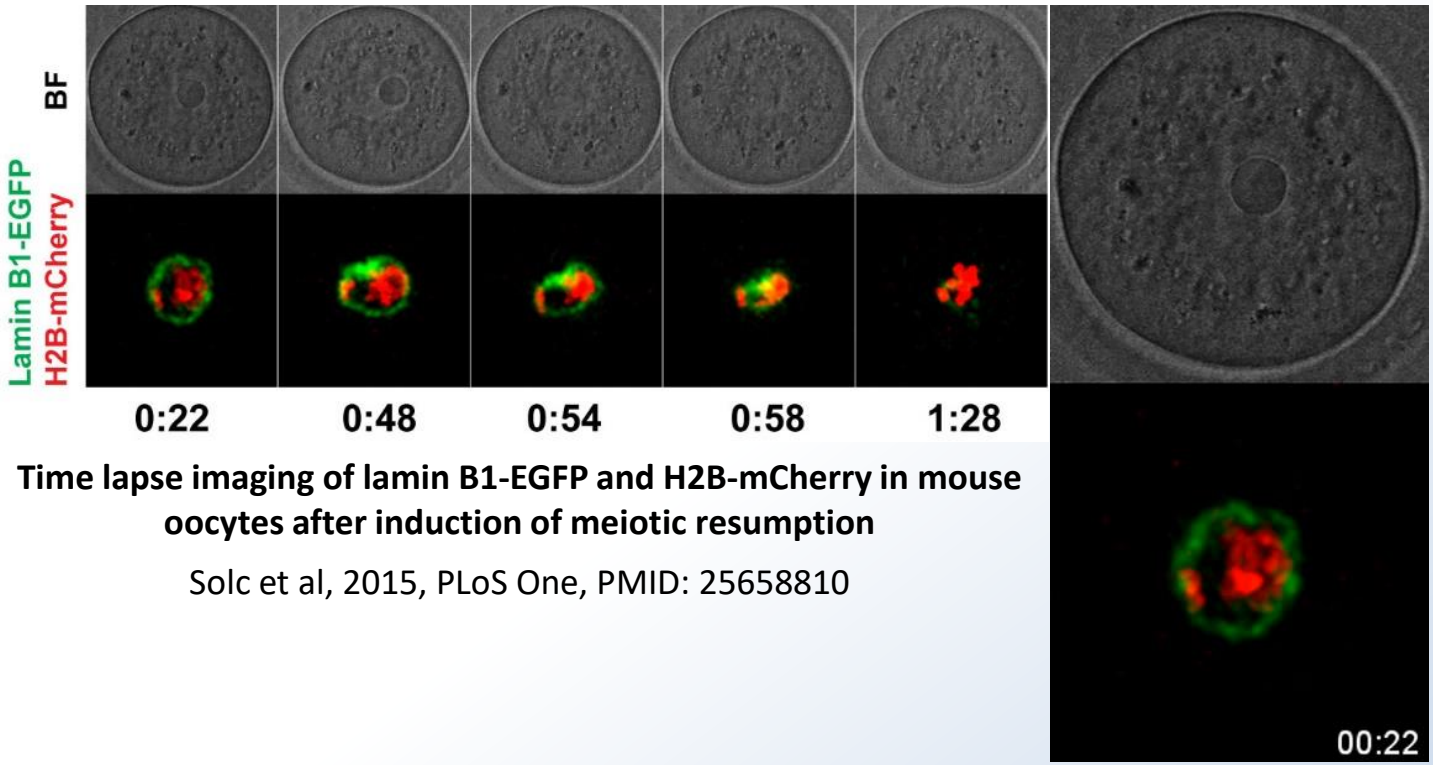


Clift, Schuh, 2013, Nat Rev Mol Cell Biol., PMID: 23942453



- meiosis I and meiosis II – **primary vs. secondary oocyte**
- cell cycle arrest at **prophase I** and **metaphase II**
- asymmetric cell division
- spinde formation

MEIOTIC RESUMPTION FROM THE PROPHASE I



Time lapse imaging of lamin B1-EGFP and H2B-mCherry in mouse oocytes after induction of meiotic resumption

Solc et al, 2015, PLoS One, PMID: 25658810

- germinal vesicle breakdown **(GVBD)** and chromosome condensation followed by the microtubule assembly
- often compared with the **G2-M transition in somatic cells**

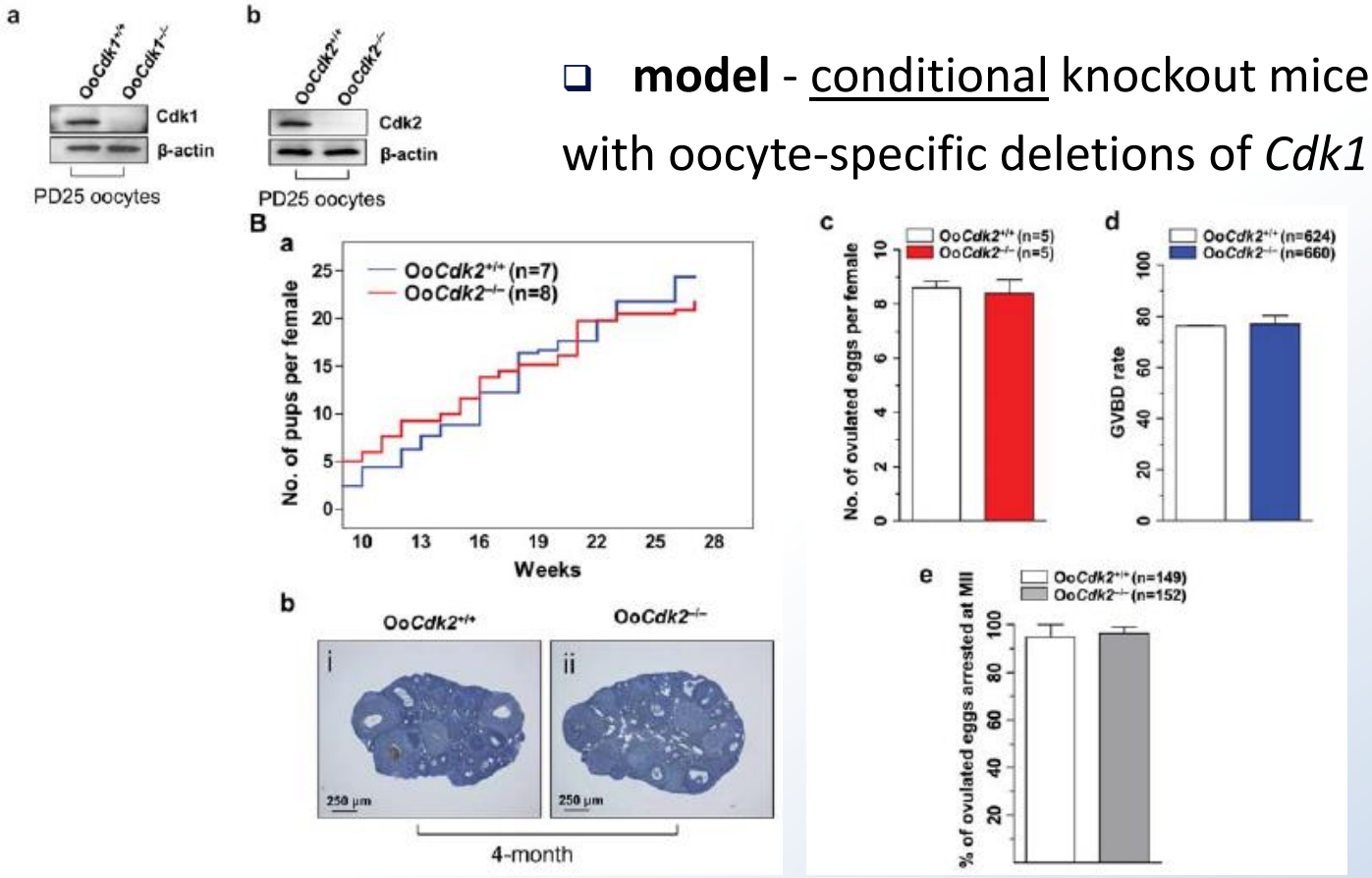
SPECIFIC REQUIREMENTS OF CDKS DURING THE MEIOTIC RESUMPTION

Table 1 | **Representative mouse models carrying gene-targeted CDK alleles***

Kinase	Genotype§	Phenotype
Loss-of-function strains		
CDK1	*Cdk1*$^{mut/mut}$	Deficiency in CDK1 results in embryonic lethality in the first cell divisions
CDK2	*Cdk2*$^{-/-}$	Sterility due to defective meiosis; no effect on mitotic cells
CDK4	*Cdk4*$^{-/-}$	Diabetes and defective postnatal proliferation of endocrine cells such as pancreatic β-cells or pituitary hormone-producing cells
CDK6	*Cdk6*$^{-/-}$	Slight anaemia and defective proliferation of some haematopoietic cells
CDK11	*Cdk11*$^{-/-}$	Embryonic lethality in peri-implantation embryos accompanied by mitotic aberrations
CDK2; CDK4; CDK6	*Cdk2*$^{-/-}$; *Cdk4*$^{-/-}$; *Cdk6*$^{-/-}$	Deficiency in all these interphase CDKs provokes embryonic lethality by mid-gestation due to haematopoietic defects

Malumbres, Barbacid, 2009, Nature Reviews

SPECIFIC REQUIREMENTS OF CDK1 AND CDK2 DURING THE MEIOTIC RESUMPTION



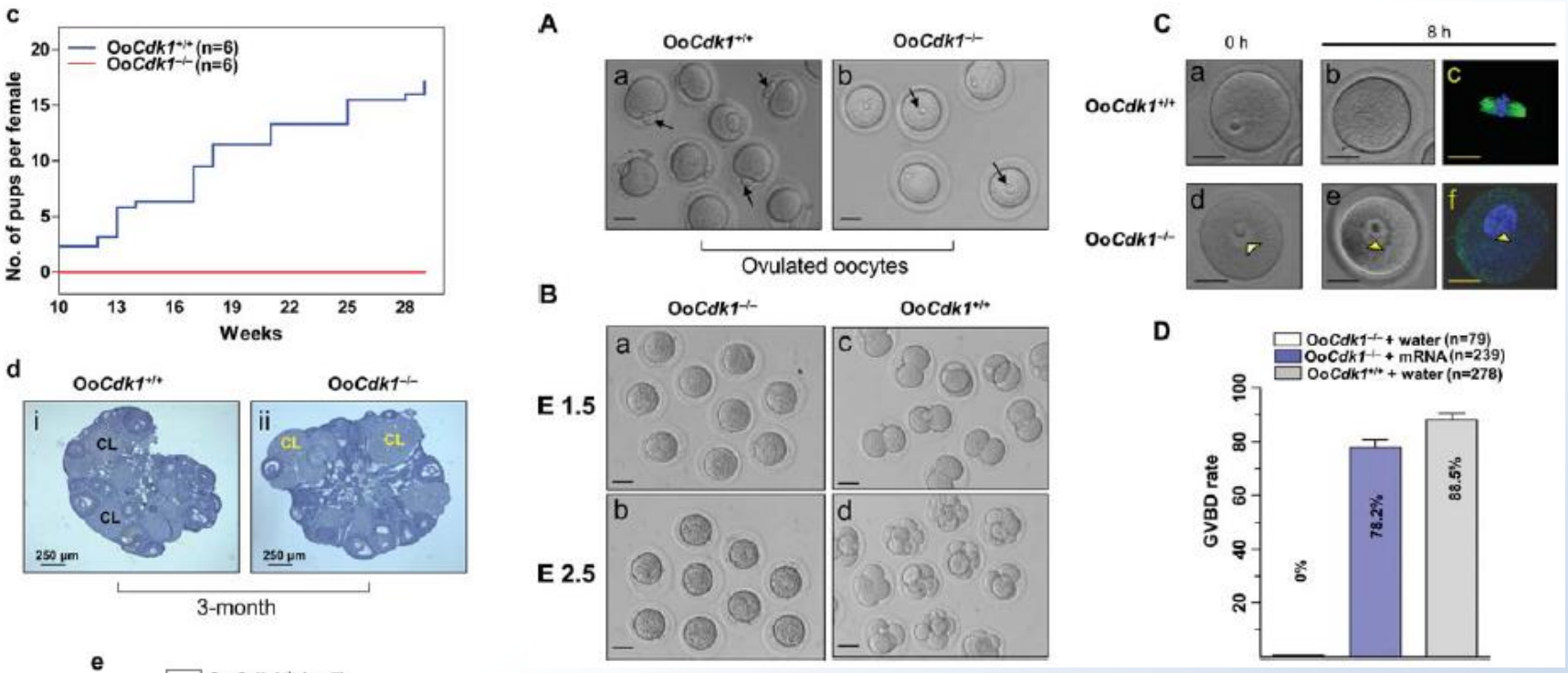
- **model** - conditional knockout mice

with oocyte-specific deletions of *Cdk1* or *Cdk2*

Adhikari et al, 2012, Hum Mol Genet, PMID: 22367880

- *Cdk2* is not required for the resumption of meiosis in mouse oocytes

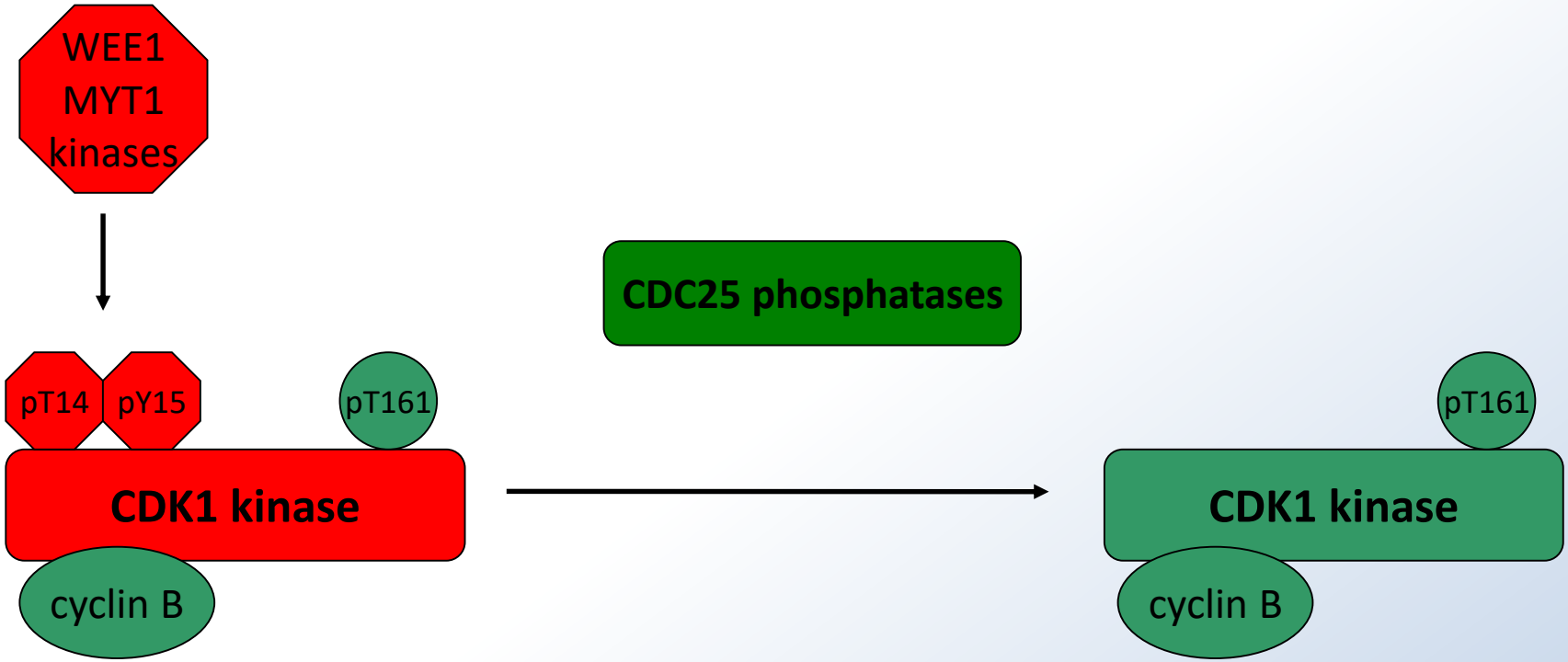
SPECIFIC REQUIREMENTS OF CDK1 AND CDK2 DURING THE MEIOTIC RESUMPTION



Adhikari et al, 2012, Hum Mol Genet, PMID: 22367880

- ***Cdk1*** **is indispensable for the triggering of the resumption of meiosis in mammalian oocytes**

MECHANISMS OF CDK1 REGULATION

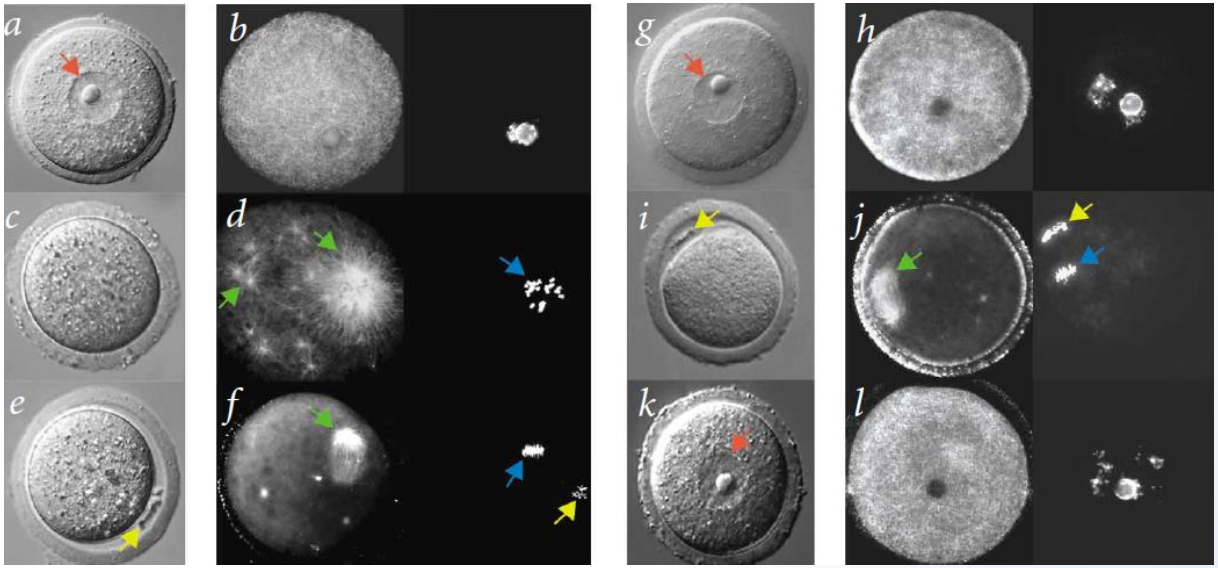


- cyclins, **activating** and **inhibitory phosphorylation**

MECHANISMS OF CDK1 REGULATION IN MAMMALIAN OOCYTES

Cdc25b +/+ oocytes

Cdc25b -/- oocytes



+ *Cdc25b* cRNA

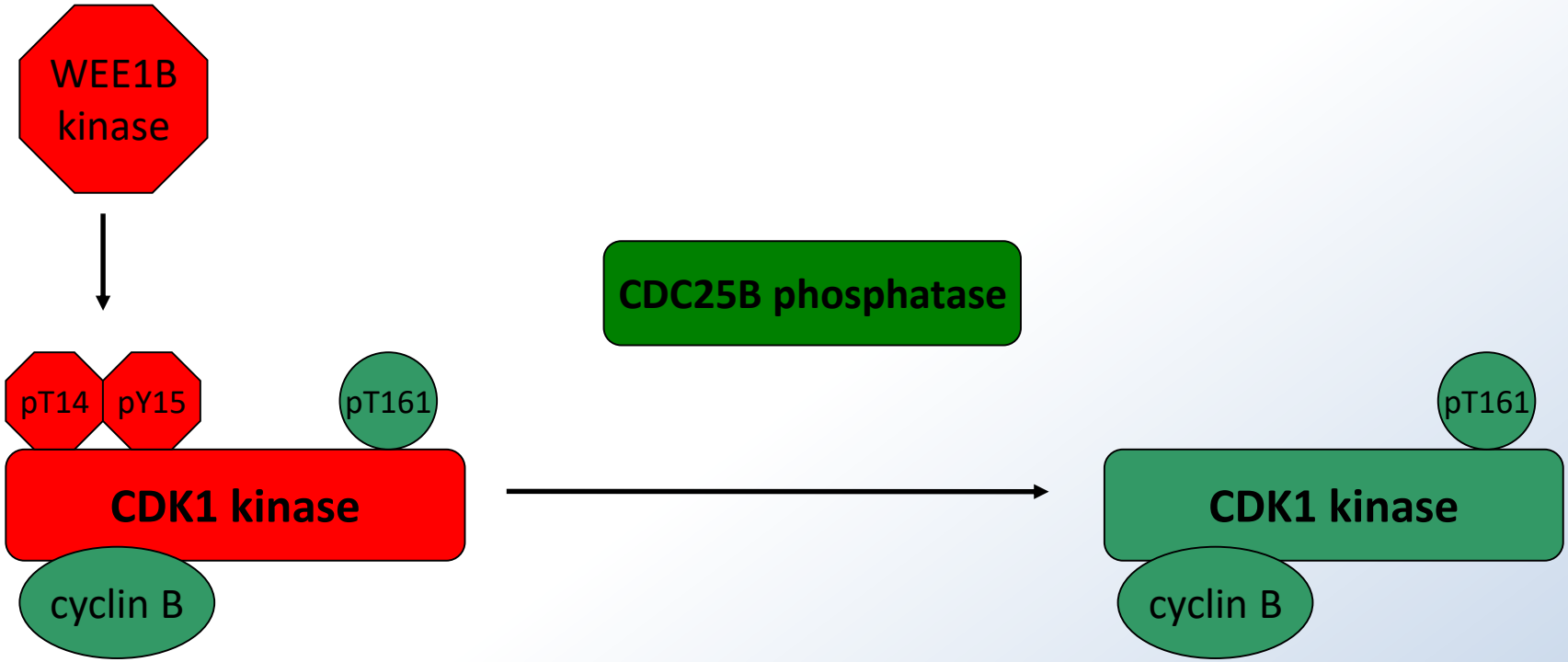
+ catalytically inactive *Cdc25b* cRNA

Cdc25b–/– oocytes are permanently arrested at the germinal vesicle stage

Lincoln et al, 2002, Nat Genet. PMID: 11912493

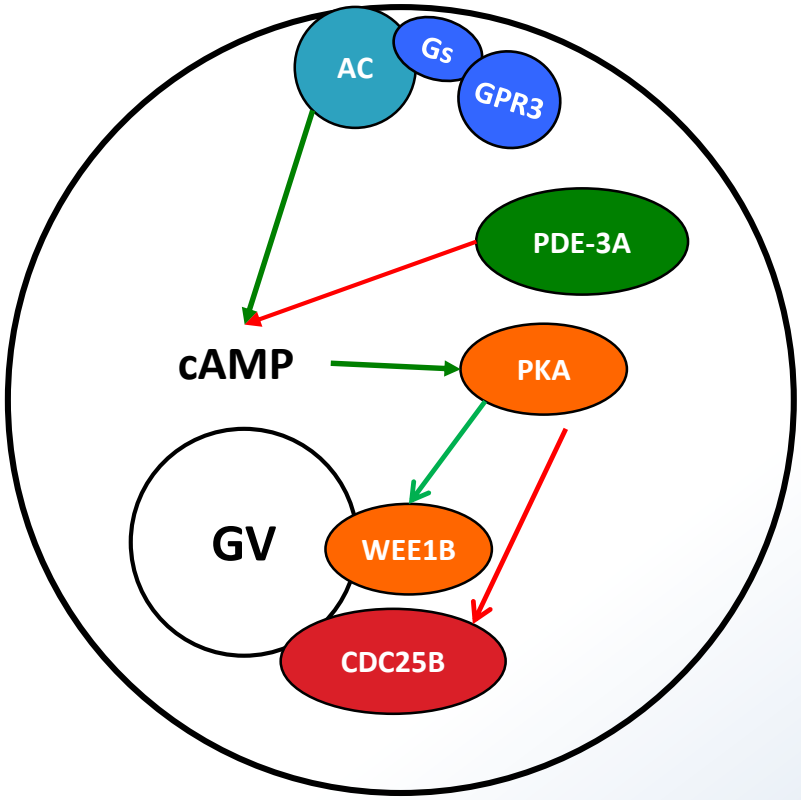
- **CDC25B phosphatase is required for resumption of meiosis**

MECHANISMS OF CDK1 REGULATION IN MAMMALIAN OOCYTES



- CDC25B is also required for the MI-MII transition in mouse oocytes (Ferencova et al, 2022, *J Cell Sci.*, PMID: 35237831)
- **How to restore CDK1 activity in *Cdc25b* -/- oocytes?**

MAINTAINING OF PROPHASE I ARREST

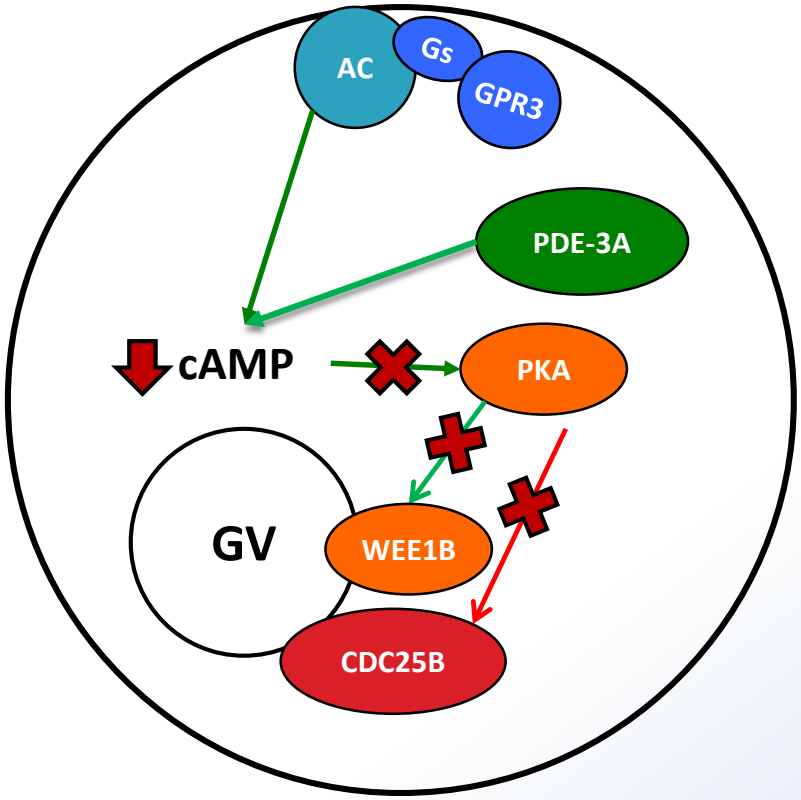


- **cAMP** – cyclic adenosine monophosphate
- **PDE-3A** - phosphodiesterase 3A
- **AC** -adenylate cyclase
- **PKA** – protein kinase A

cytoplasm **nucleus**

CDC25B is sequestered in the cytoplasm by the 14-3-3 adaptor protein
Pirino et al, 2009, Cell Cycle, PMID: 19223768

MAINTAINING OF PROPHASE I ARREST

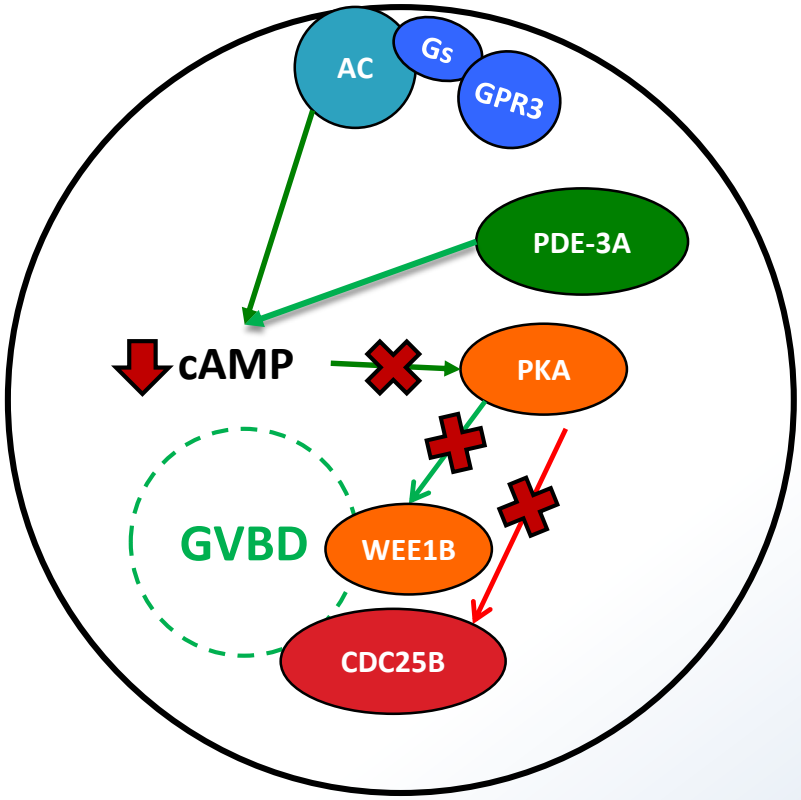


- **cAMP** – cyclic adenosine monophosphate
- **PDE-3A** - phosphodiesterase 3A
- **AC** -adenylate cyclase
- **PKA** – protein kinase A

cytoplasm **nucleus**

CDC25B is sequestered in the cytoplasm by the 14-3-3 adaptor protein
Pirino et al, 2009, Cell Cycle, PMID: 19223768

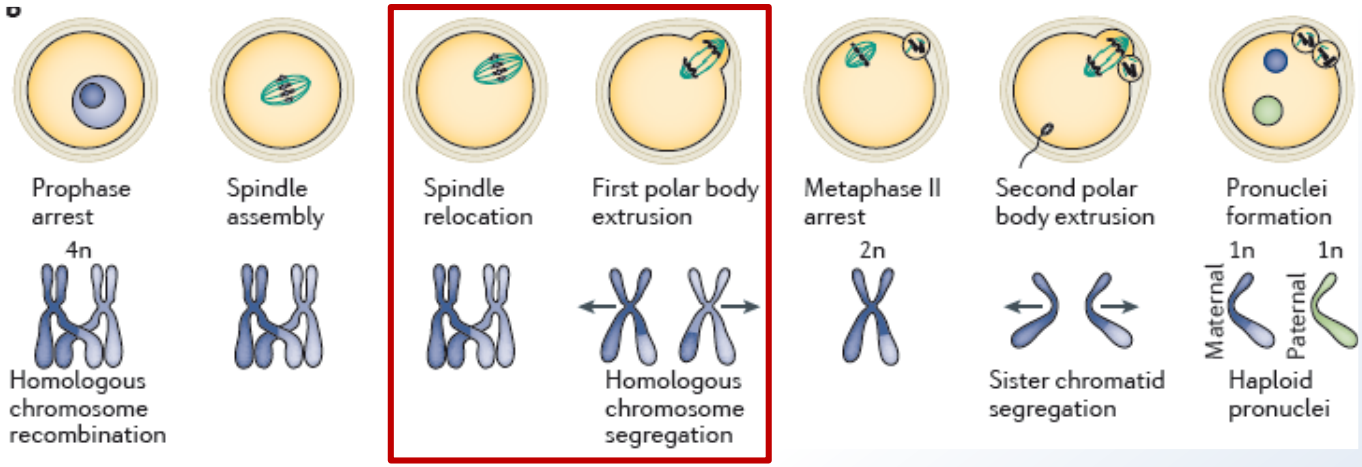
MAINTAINING OF PROPHASE I ARREST



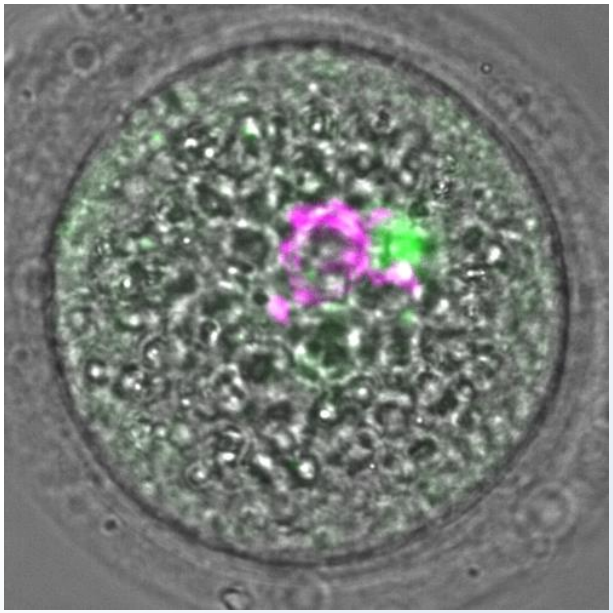
- **cAMP** – cyclic adenosine monophosphate
- **PDE-3A** - phosphodiesterase 3A
- **AC** -adenylate cyclase
- **PKA** – protein kinase A

CDC25B is sequestered in the cytoplasm by the 14-3-3 adaptor protein
Pirino et al, 2009, Cell Cycle, PMID: 19223768

MAMMALIAN OOCYTE MATURATION

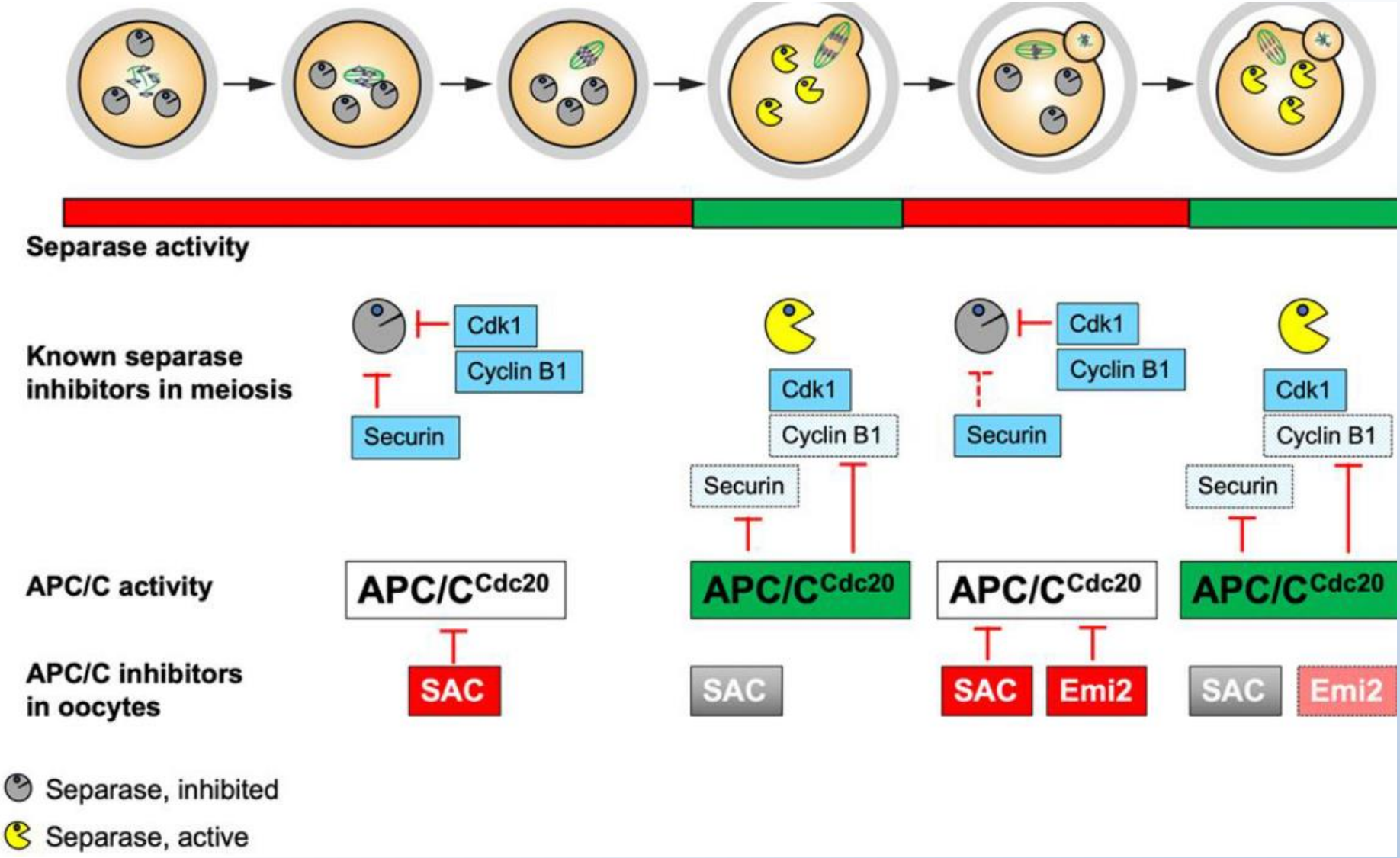


Clift, Schuh, 2013, Nat Rev Mol Cell Biol., PMID: 23942453



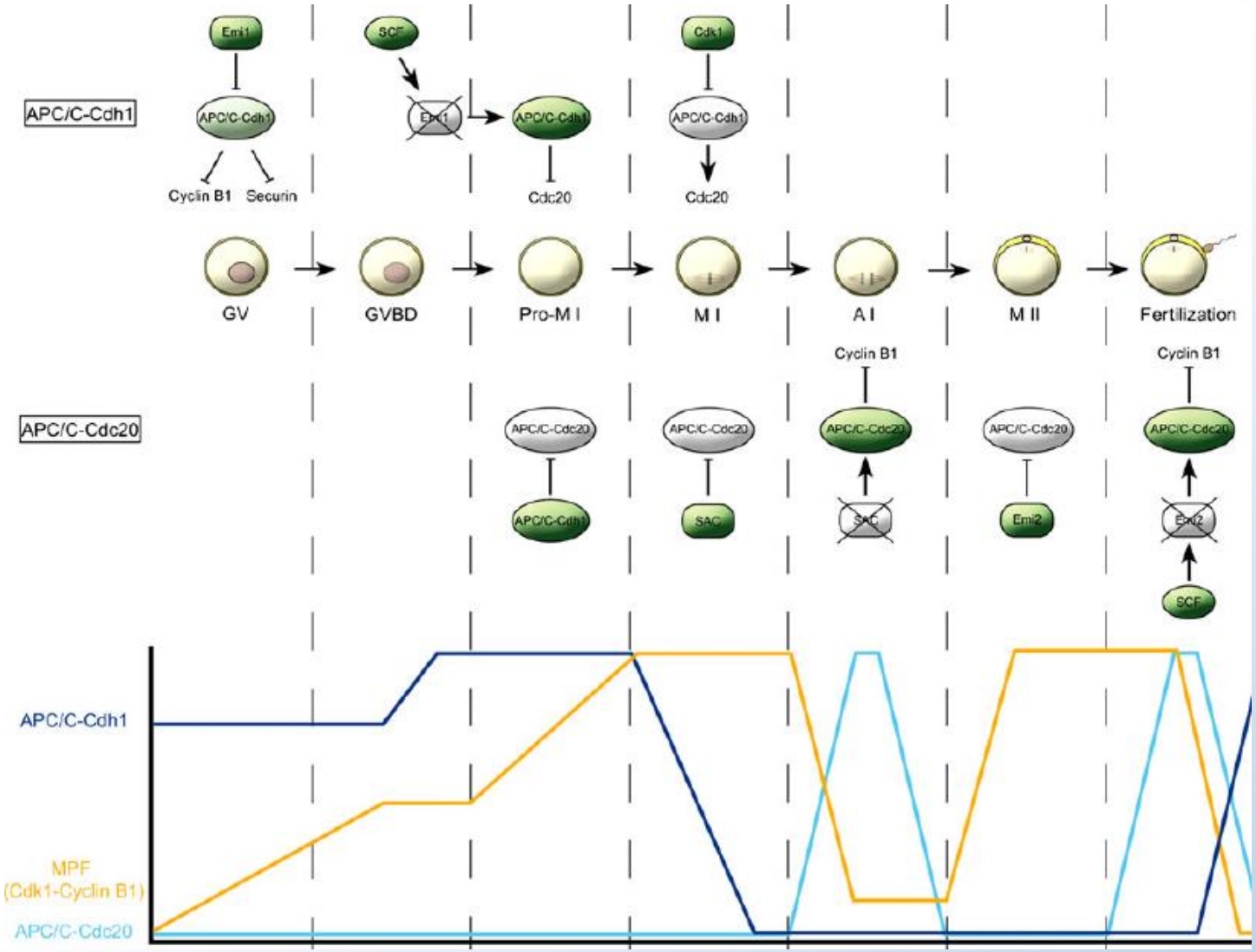
- meiosis I and meiosis II – **primary vs. secondary oocyte**
- cell cycle arrest at **prophase I** and **metaphase II**
- asymmetric cell division
- spinde formation

REGULATION OF SEPARASE ACTIVITY IN HOMOLOGOUS CHROMOSOMES SEGREGATION



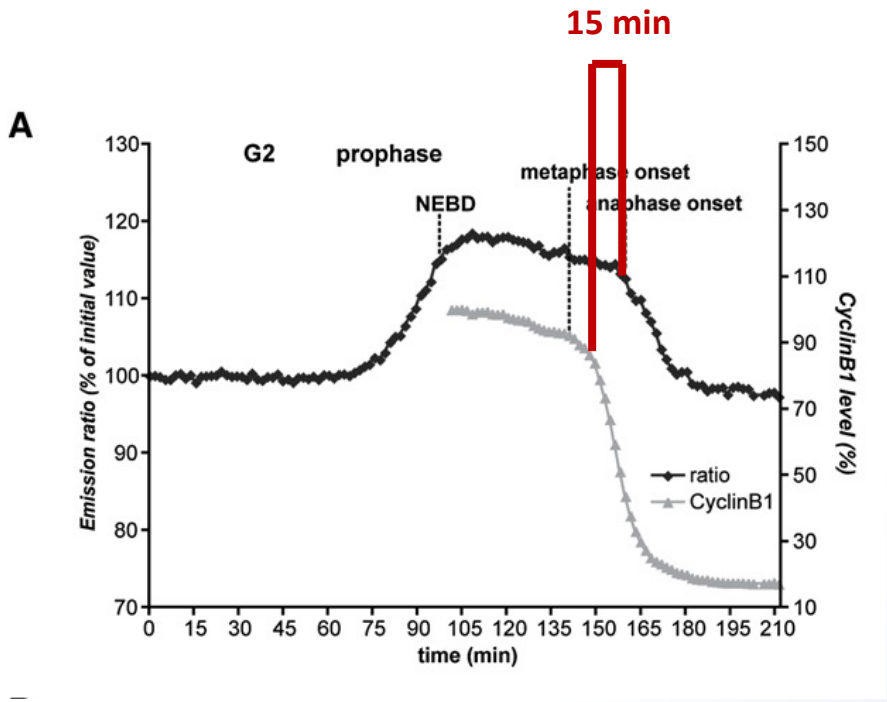
Wassmann, 2022, Cells

ACTIVITY OF APC/C-CDH1, APC/C-CDC20 AND MPF DURING MEIOSIS



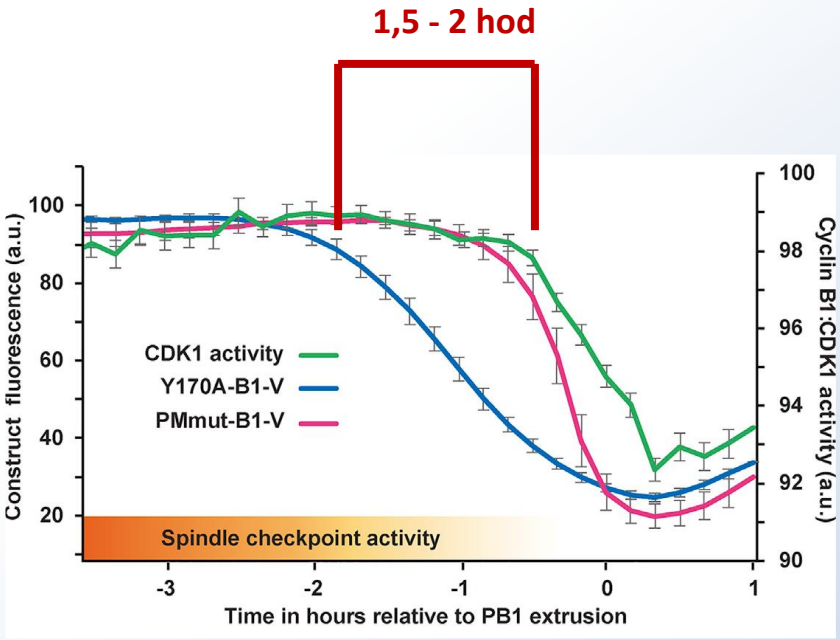
Karabinova et al, 2011, Cell Tissue Res., PMID: 21969023

CYCLIN B1 DEGRADATION IN SOMATIC CELLS VS MOUSE OOCYTES



Cyclin B1 level vs CDK1 activity in somatic cell...

Gavet, Pines, 2010, Dev Cell., PMID: 20412769



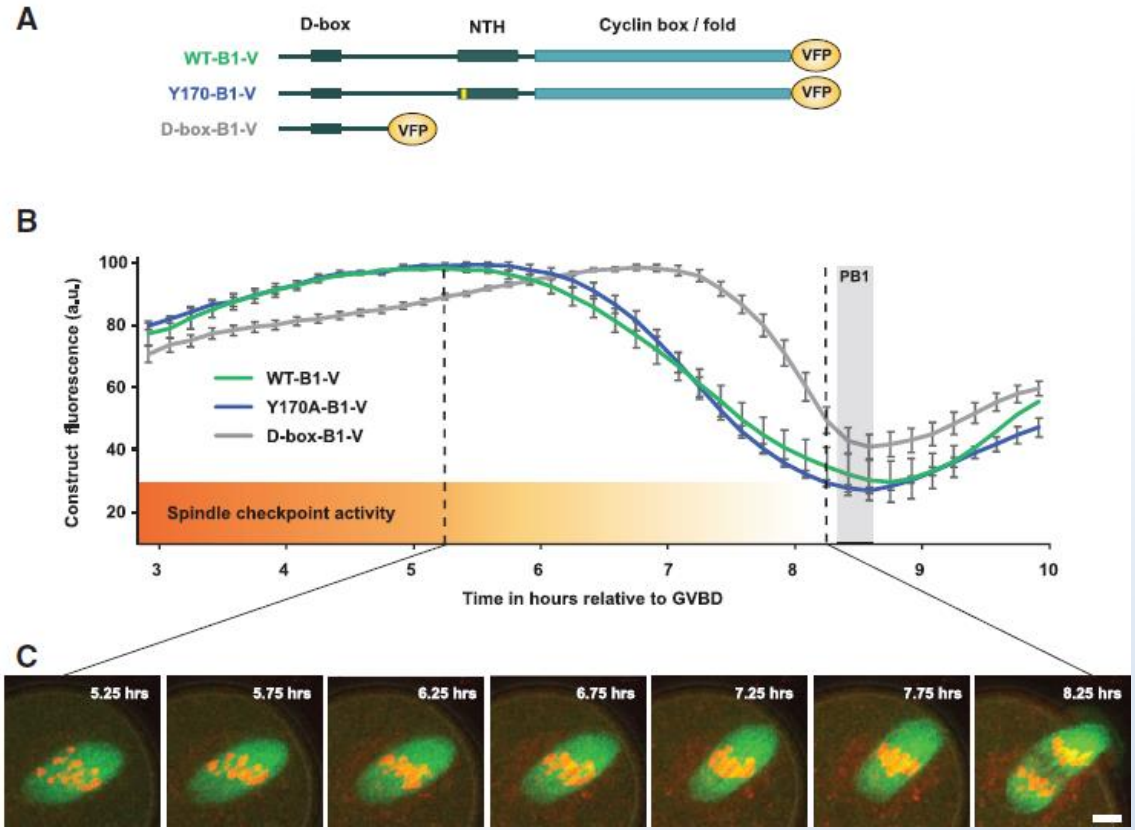
Y170-B1-V = full length cyclin B1 that cannot bind CDK1

...and mammalian oocyte

Levasseur et al, 2019, Dev Cell., PMID: 30745144

- triggered **in metaphase in mitotic cells**, but **in prometaphase I in mammalian oocytes**

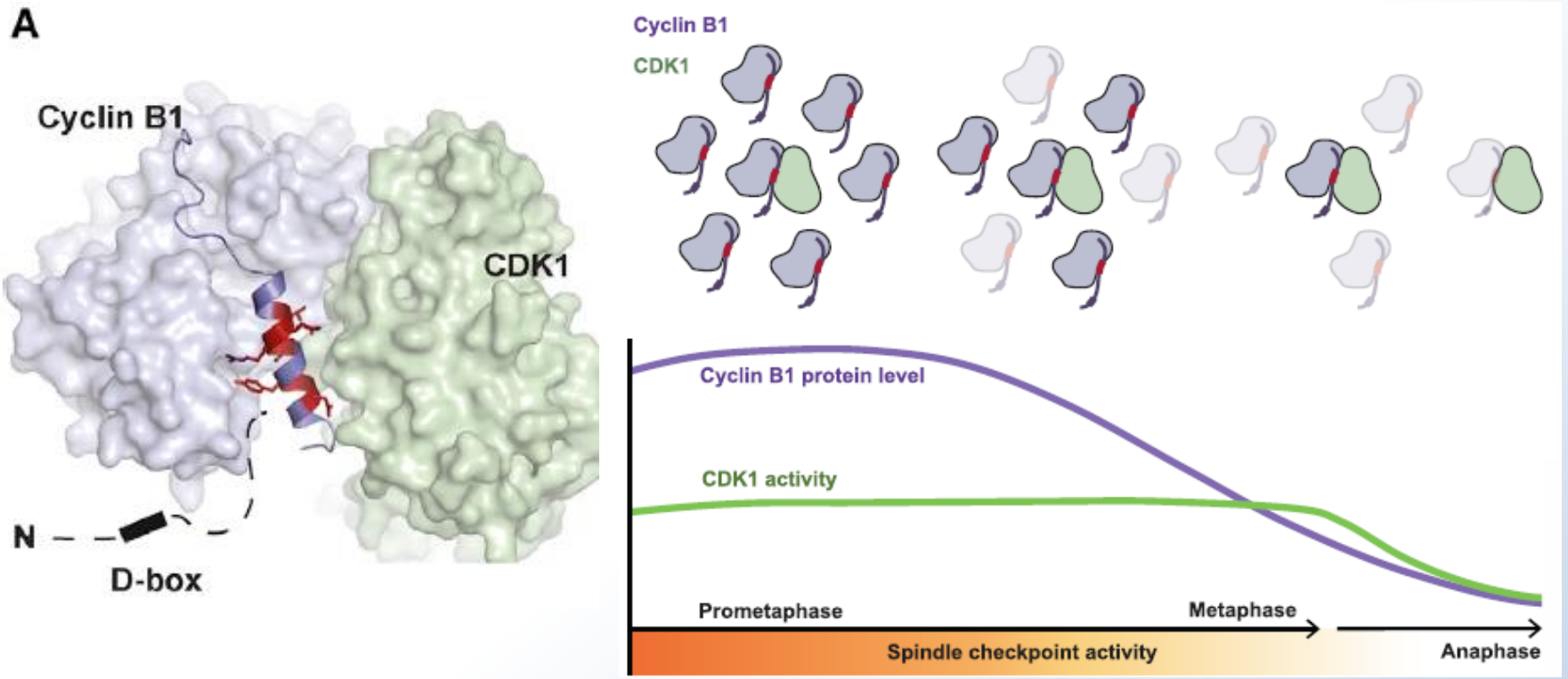
CYCLIN B1 DEGRADATION IN MOUSE OOCYTES



Levasseur et al, 2019, Dev Cell., PMID: 30745144

- **destruction motives:**
- **NTH** – not SAC sensitive, **masked within the cyclin B1:CDK1 interface**
- **D-box**- SAC sensitive need high APC activity for destruction

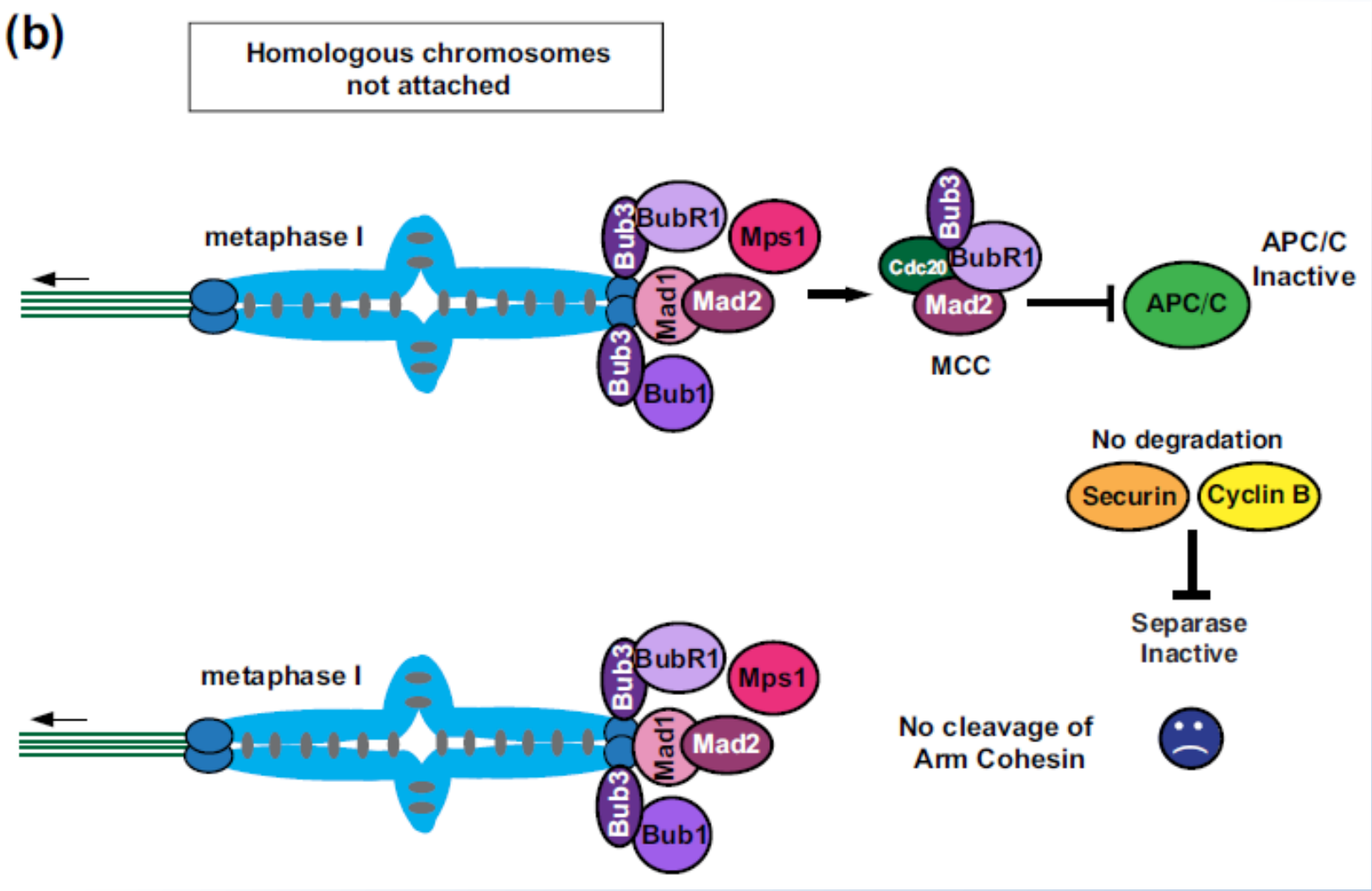
CYCLIN B1 DEGRADATION IN MOUSE OOCYTES



Levasseur et al, 2019, Dev Cell., PMID: 30745144

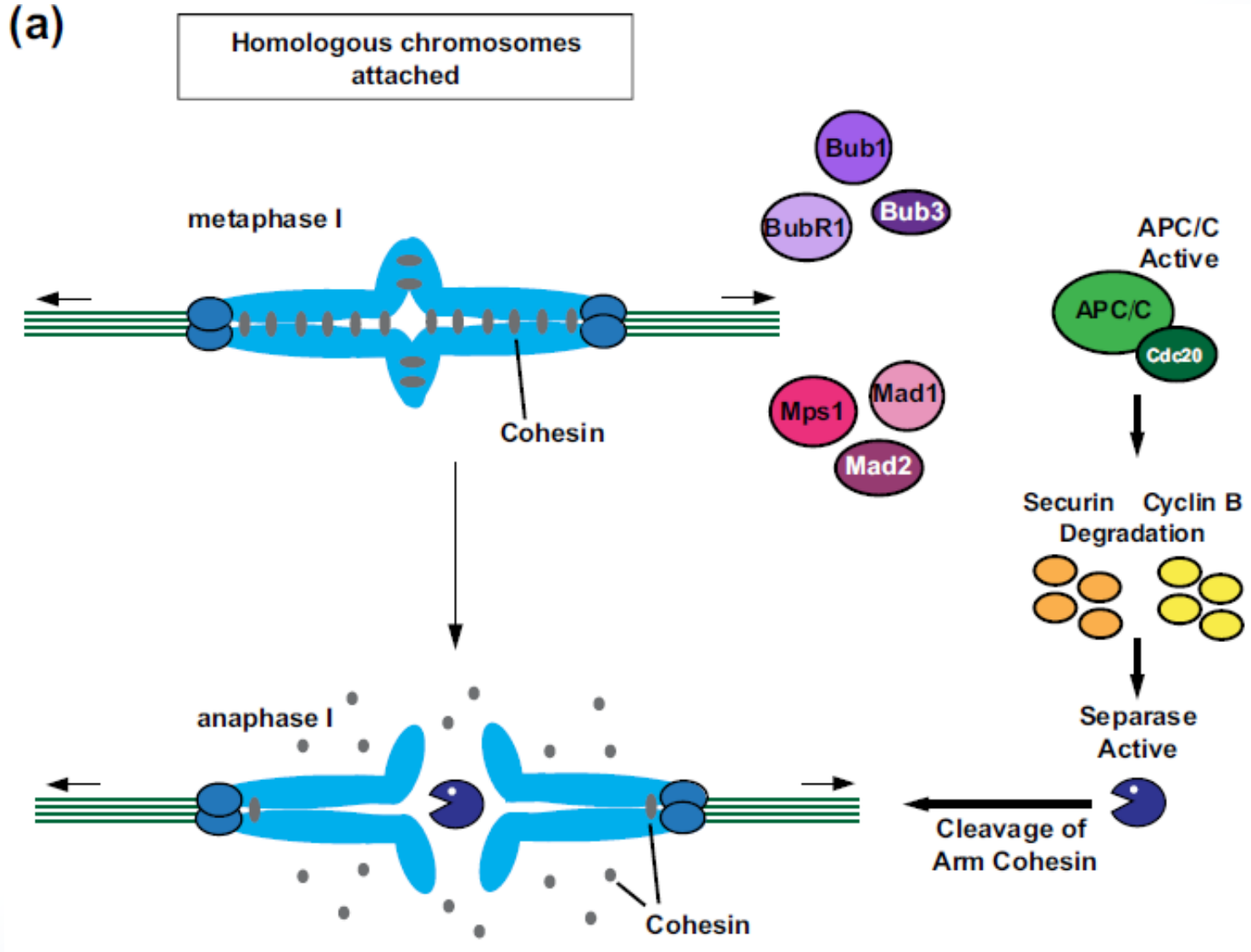
- Total cyclin B1 level does not reflect CDK1 activity in MI oocytes because an **excess of free cyclin B1 is destroyed ahead of CDK1-bound cyclin B1**
- Prolonged **CDK1 activity assists the SAC and prevents aneuploidy**

SPINDLE ASSEMBLY CHECKPOINT (SAC) IN MEIOSIS



Touati & Wassmann, 2016, Chromosoma

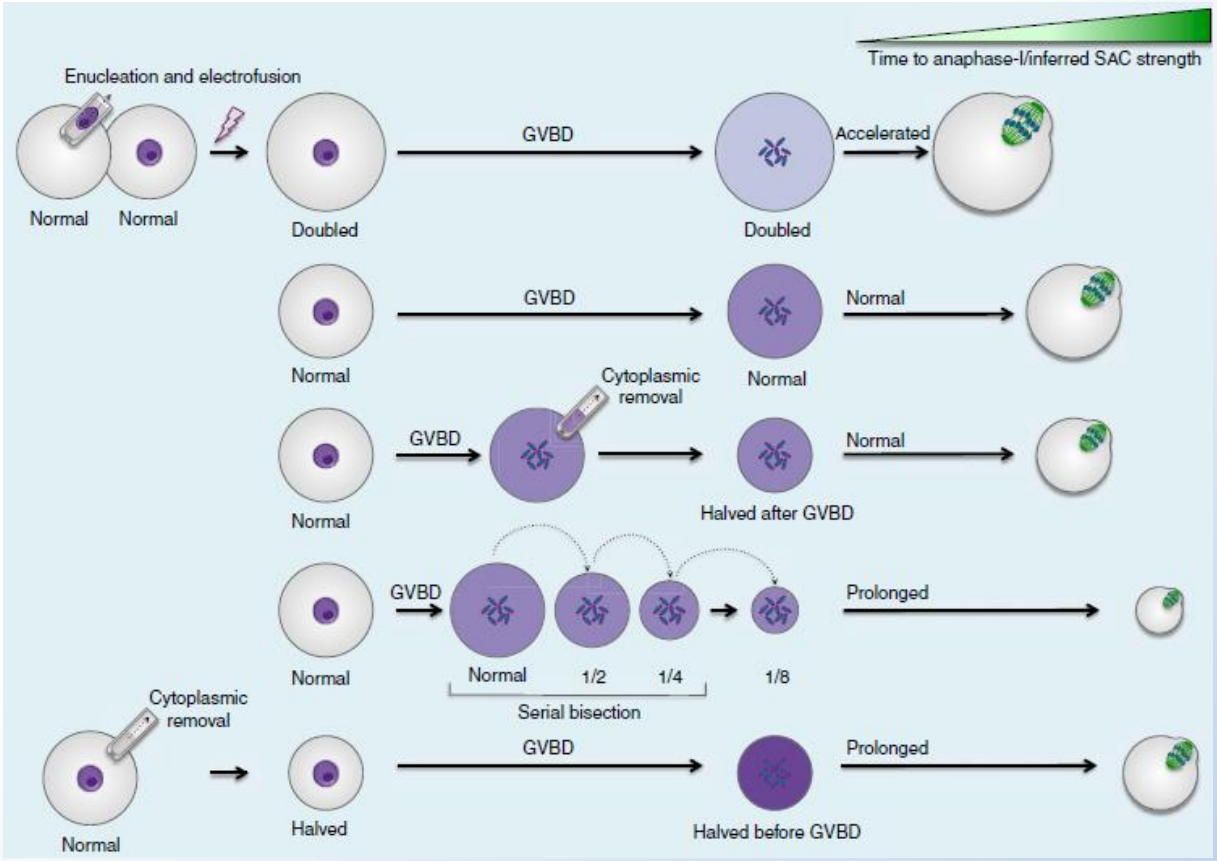
SPINDLE ASSEMBLY CHECKPOINT (SAC) IN MEIOSIS



Touati & Wassmann, 2016, Chromosoma

SAC SIGNALING IN MAMMALIAN OOCYTES LACKS STRINGENCY

- large cytoplasmic volume

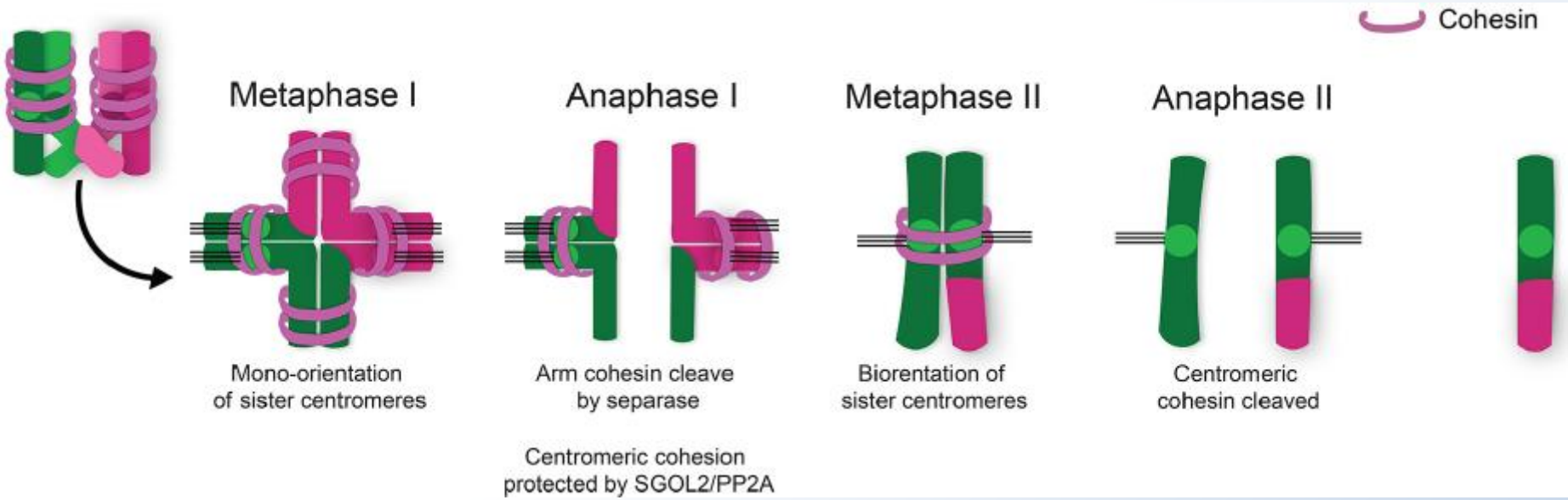


The effects of altered cytoplasmic volume on SAC strength during meiosis I

Mihajlovic and FitzHarris, 2018, *Current Biology*

- oocyte aging,...

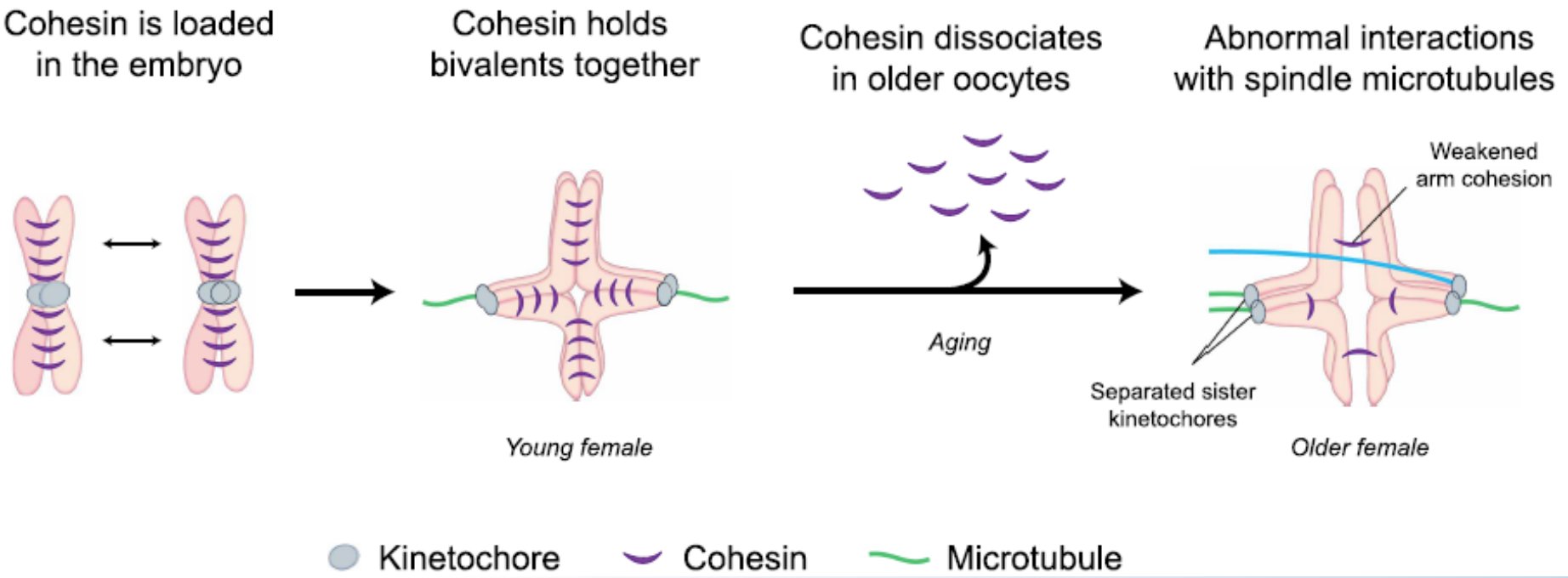
REGULATION IN HOMOLOGOUS CHROMOSOMES SEGREGATION IN MAMMALIAN OOCYTES



Beverley et al, 2021, Frontiers in Cell and Developmental Biology

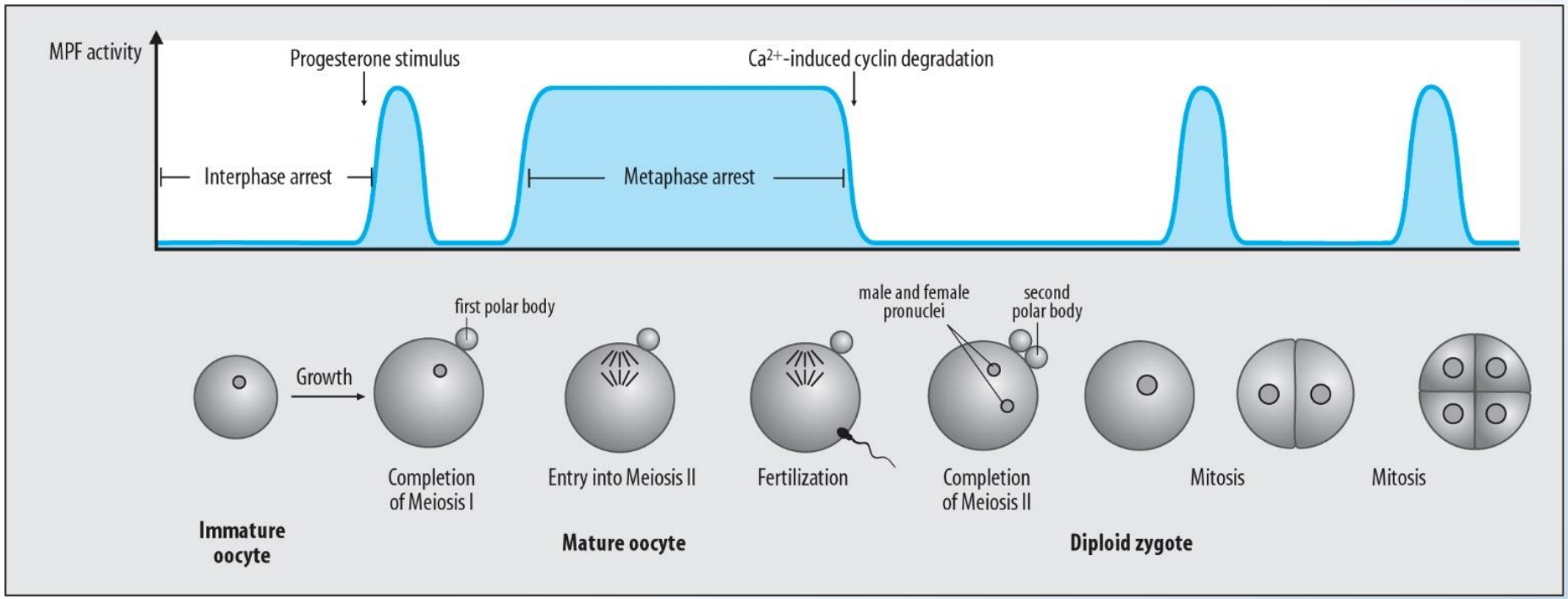
- Cohesin subunit **Rec8** at centromeres **is protected from cleavage** due to the presence of shugoshin.

AGE-RELATED DECREASE OF MEIOTIC COHESINS IN HUMAN OOCYTES



Schematic showing cohesin dissociation from chromosomes in oocytes from older women
Thomas et al,, 2021, Biochemical Society Transactions

CDK1 ACTIVITY DURING MEIOTIC MATURATION



https://socratic.org/questions/where-in-the-body-do-oocytes-mature

- Mouse oocytes proceed through meiosis I and arrest at **second meiotic metaphase with high CDK1-cyclin B1 activity.**