

Study of inter- and transgenerational effects of in ovo nutriepigenetic stimulation on chicken cecal mucosa transcriptome

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1. Introduction



Green-legged Partridgelike chicken

In ovo Chick Model for Epigenetic Research

- Early Sexual Maturity
- External Embryo Development
- Large Eggs for Easy Manipulation
- Manipulability at Any Developmental Stage
- Short Interval Between Generation
- Well-Characterized Genome
- Economic and Ethical Advantages



- Slow growing
- Well adapted to extensive management
- Low susceptibility to unfavorable conditions.
- Resistant to diseases
- Well-developed maternal traits.

Epigenetic effect inheritance

Paternal epigenetic transmission

Maternal epigenetic transmission

Environmental change

Sperm cell

F0

F0

F1

PGCs

Intergenerational Inheritance

F1

F1

F2

F2

F3

F3

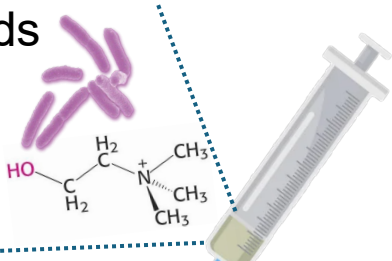
Transgenerational Inheritance

2. Hypothesis and Aim



Epigenetic stimulators

Bioactive
compounds



In ovo
injection

Embryonic development
stimulation

Epigenetic
changes

Life-long
impacts on
adult chicken
and
subsequent
generations

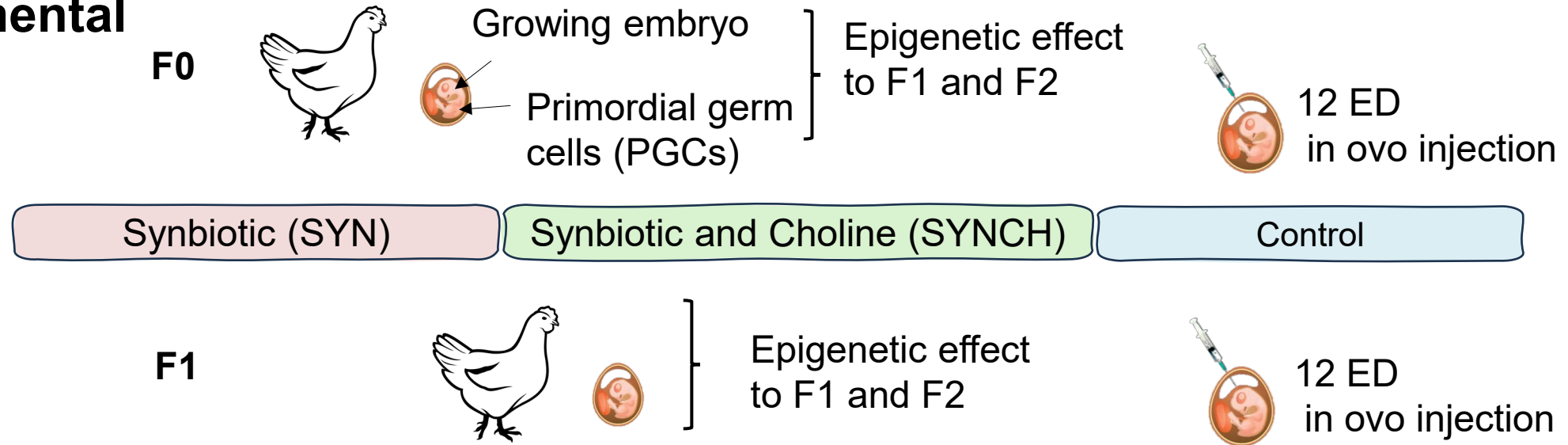
Aim

Investigate the effect of in ovo stimulation on the **cecal mucosa** transcriptome of green-legged partridge-like adult chicken in three subsequent generations.

3. Materials and Methods



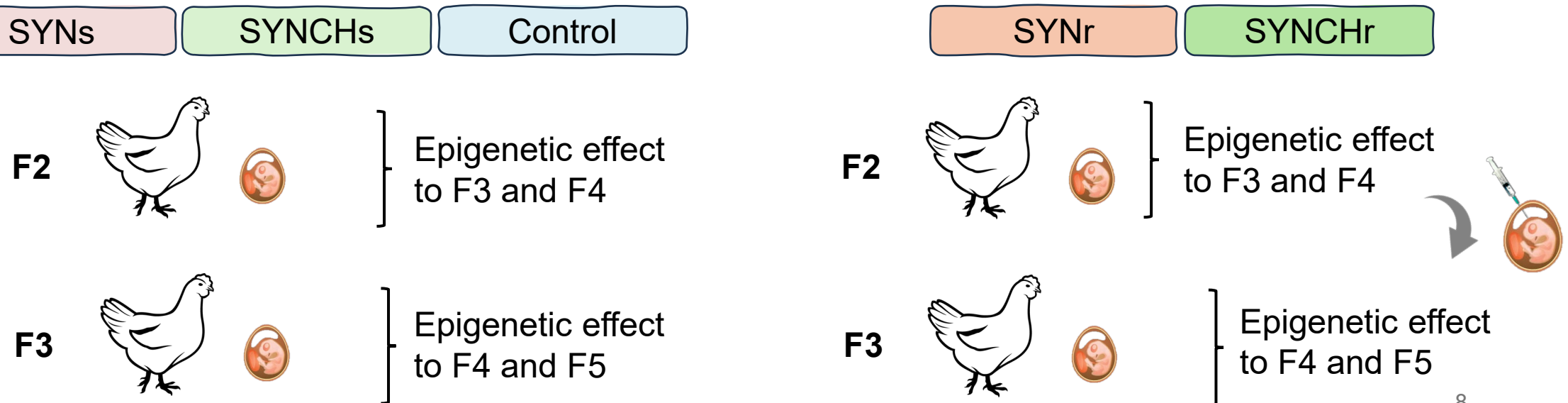
Experimental design

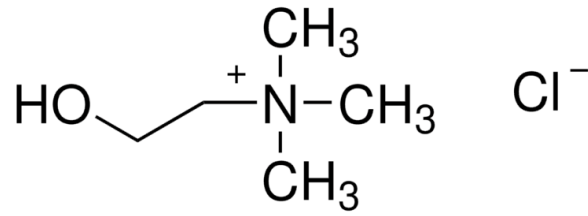


Splitting the group treatments for F2-F3

Single in ovo injection at F1 ↙

↘ Repeated in ovo injection





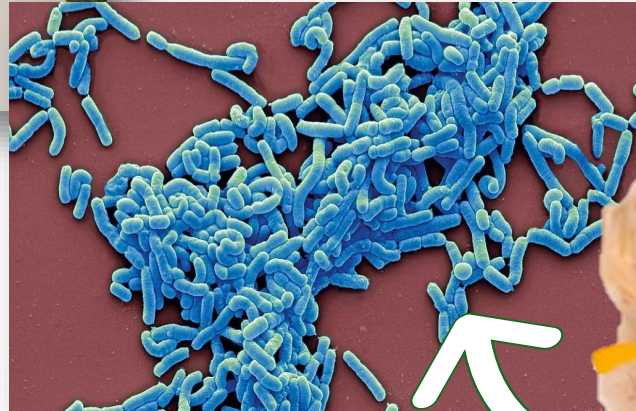
Choline



Choline:
0.25mg/embryo



Prebiotic
Fructooligosaccharides



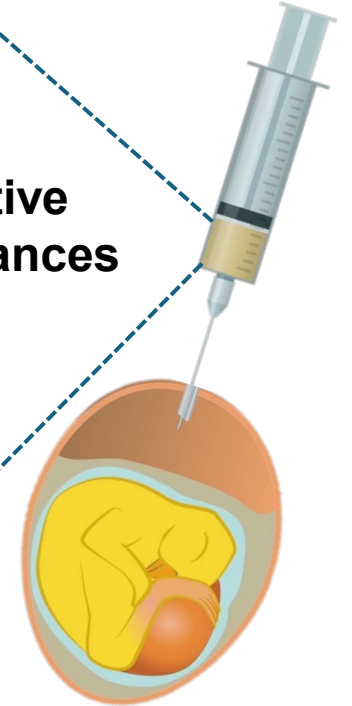
BIOMIN
PoultryStar®
Synbiotic

Probiotic strains belonging
to the genera *Enterococcus*,
Pediococcus, *Lactobacillus*,
Bifidobacterium



**Bioactive
substances**

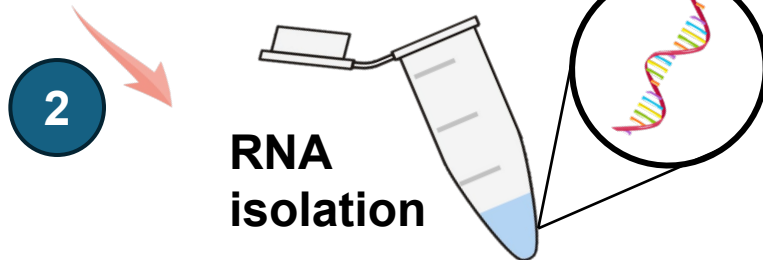
Synbiotic:
2mg/embryo



Total injected volume:
200µl



**1 Tissue sampling
Cecal mucosa**



**3 Transcriptomic
sequencing
(RNA-Seq)**
Platform: Illumina
Novaseq6000
Strategy: PE150



4

Bioinformatic analysis

Quality control of raw data
FastQC software

Trimming
Fastp software

Quality control
FastQC software

Mapping
(Gallus gallus
bGalGal1.mat.broiler.GRCg7b),
STAR v2.7.10a software

Differential expression analysis
DESeq2 v.1.34.0, an R/Bioconductor
package

5

Enrichment



GO:
Gene
Ontology

KEGG:
Kyoto
Encyclopedia of
Genes and
Genomes

4. Main Results



Differentially expressed genes (DEGs)

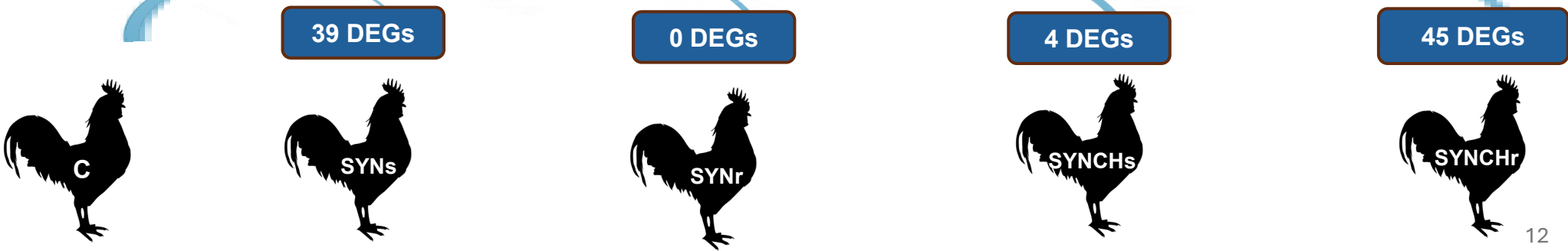
F1 generation



F2 generation

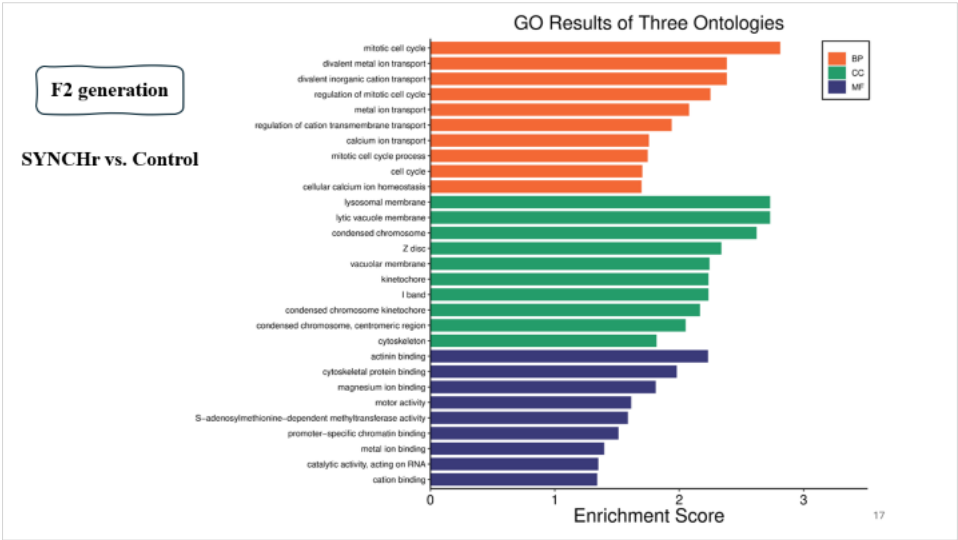
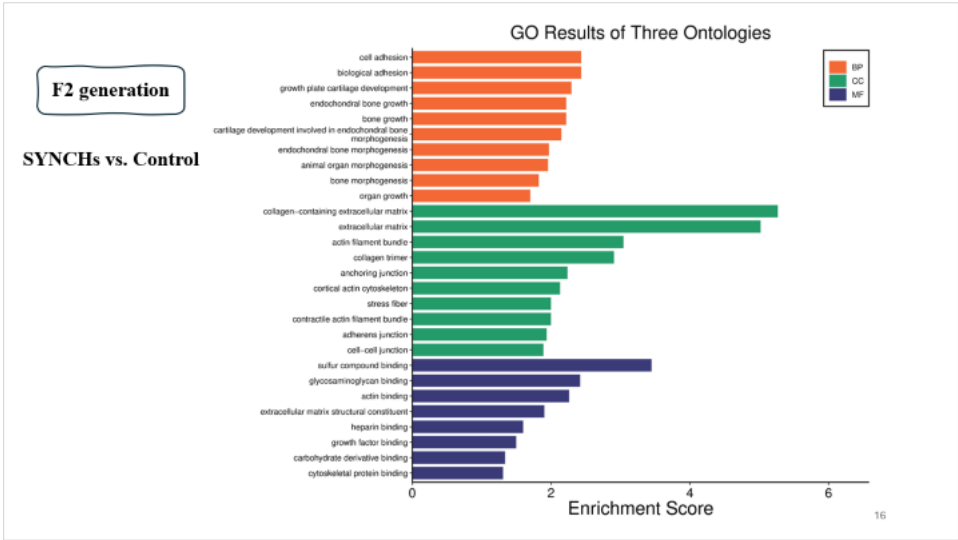
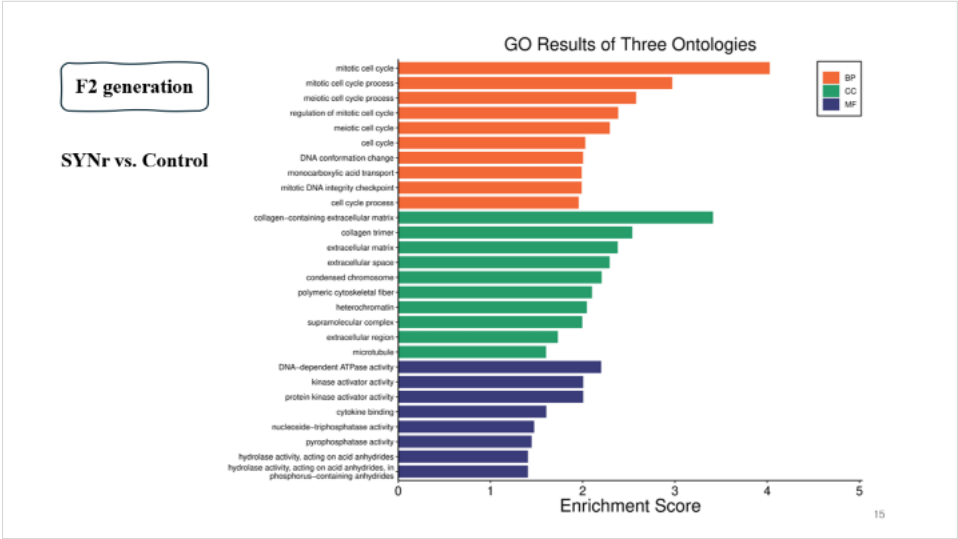
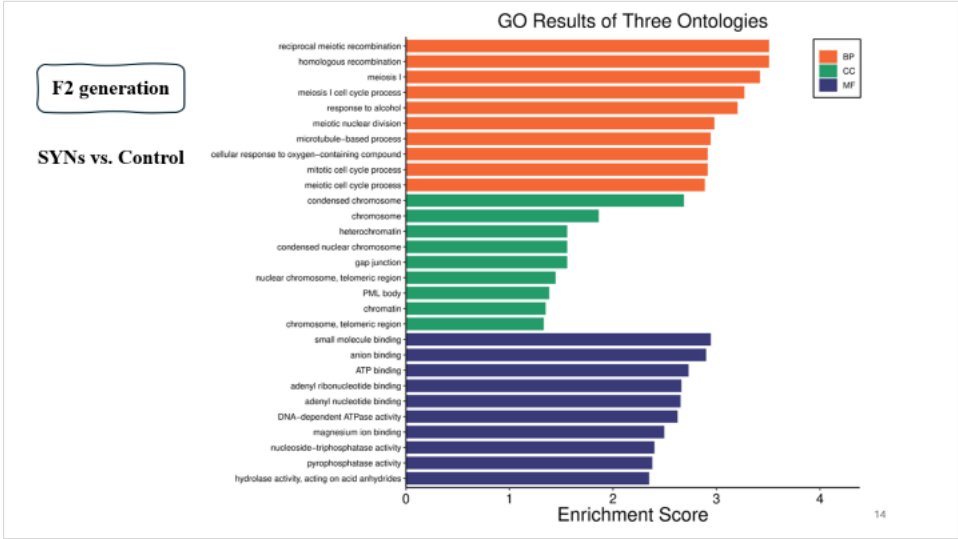


F3 generation



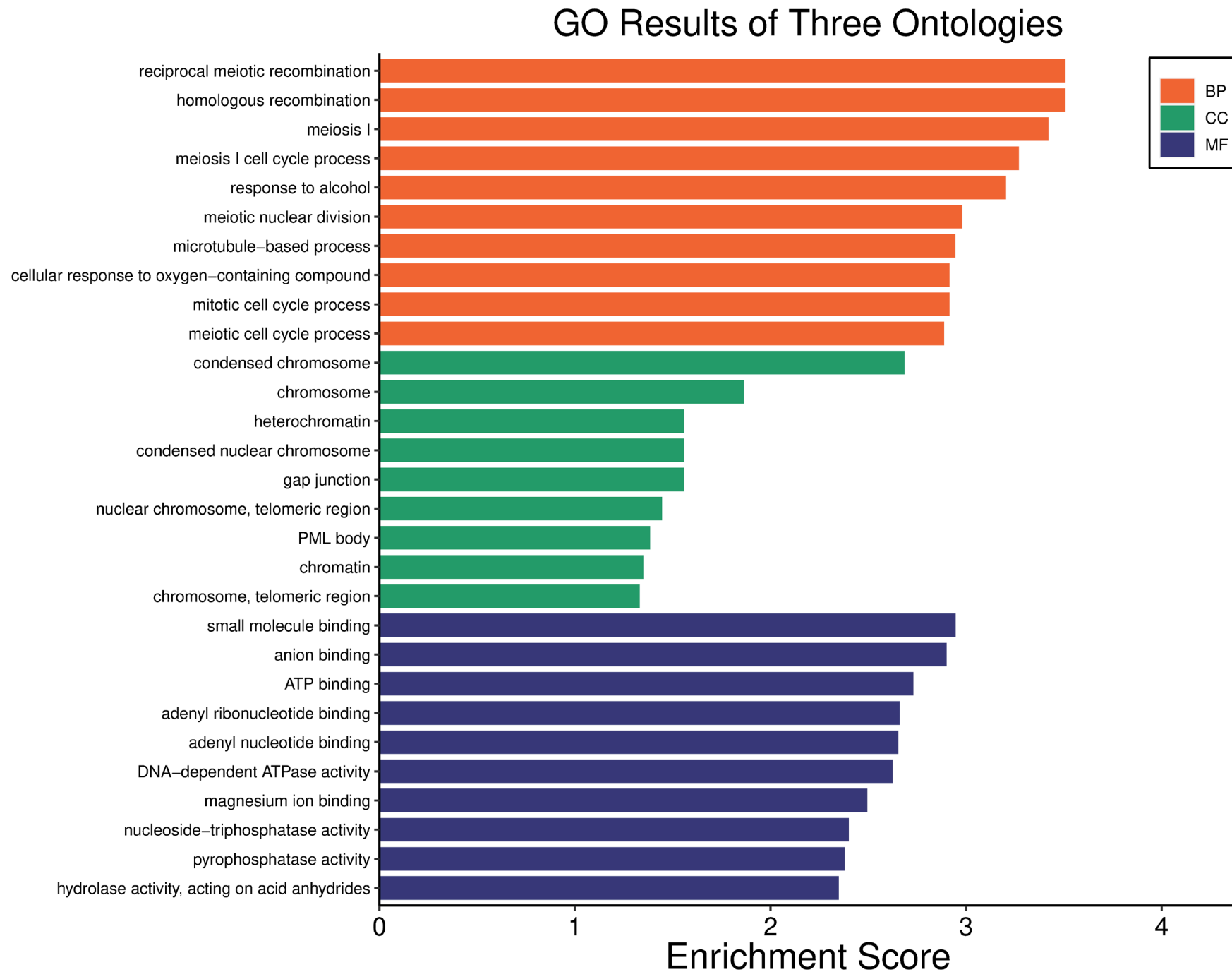
GO terms

F2 generation



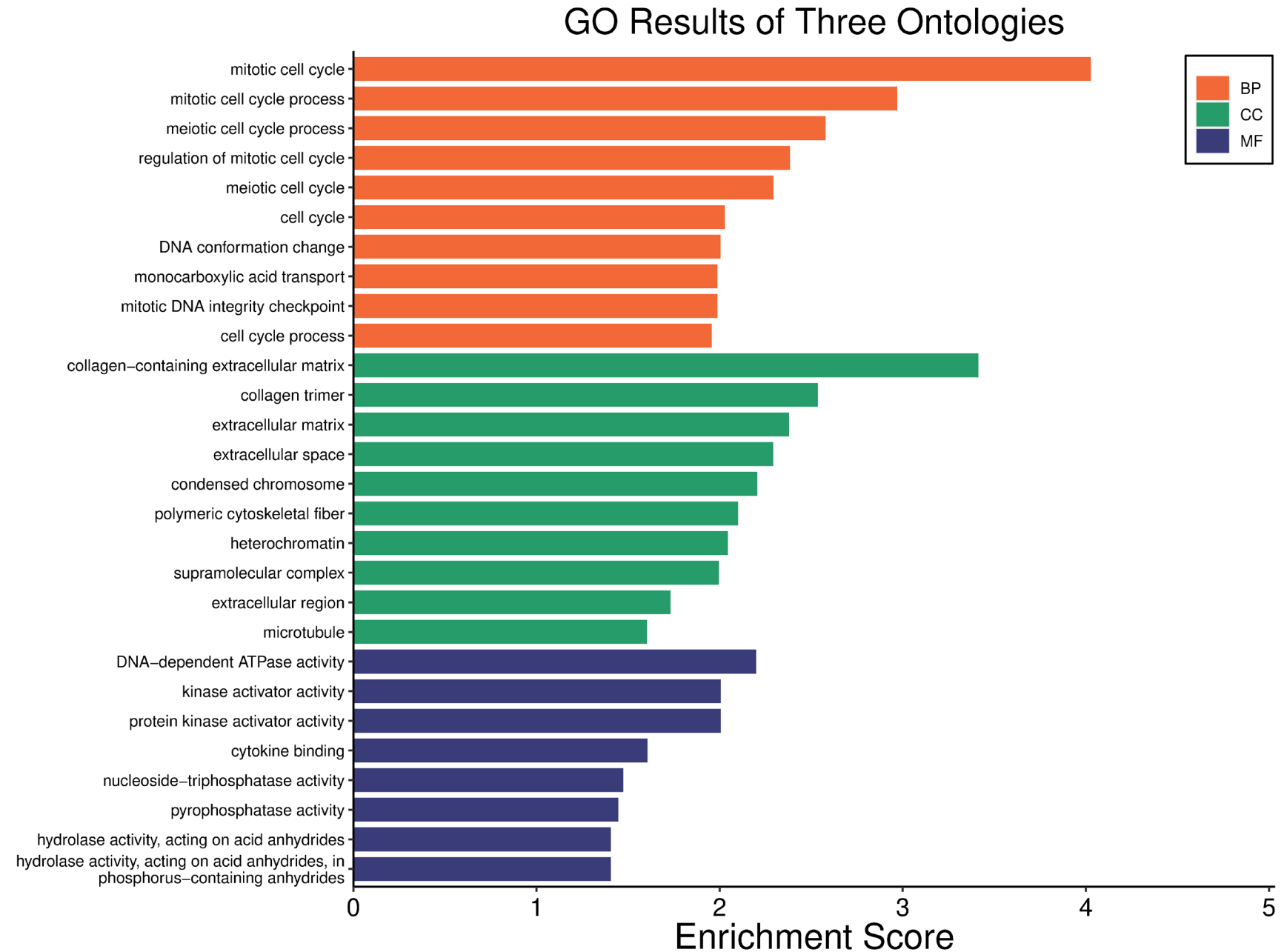
F2 generation

SYNs vs. Control



F2 generation

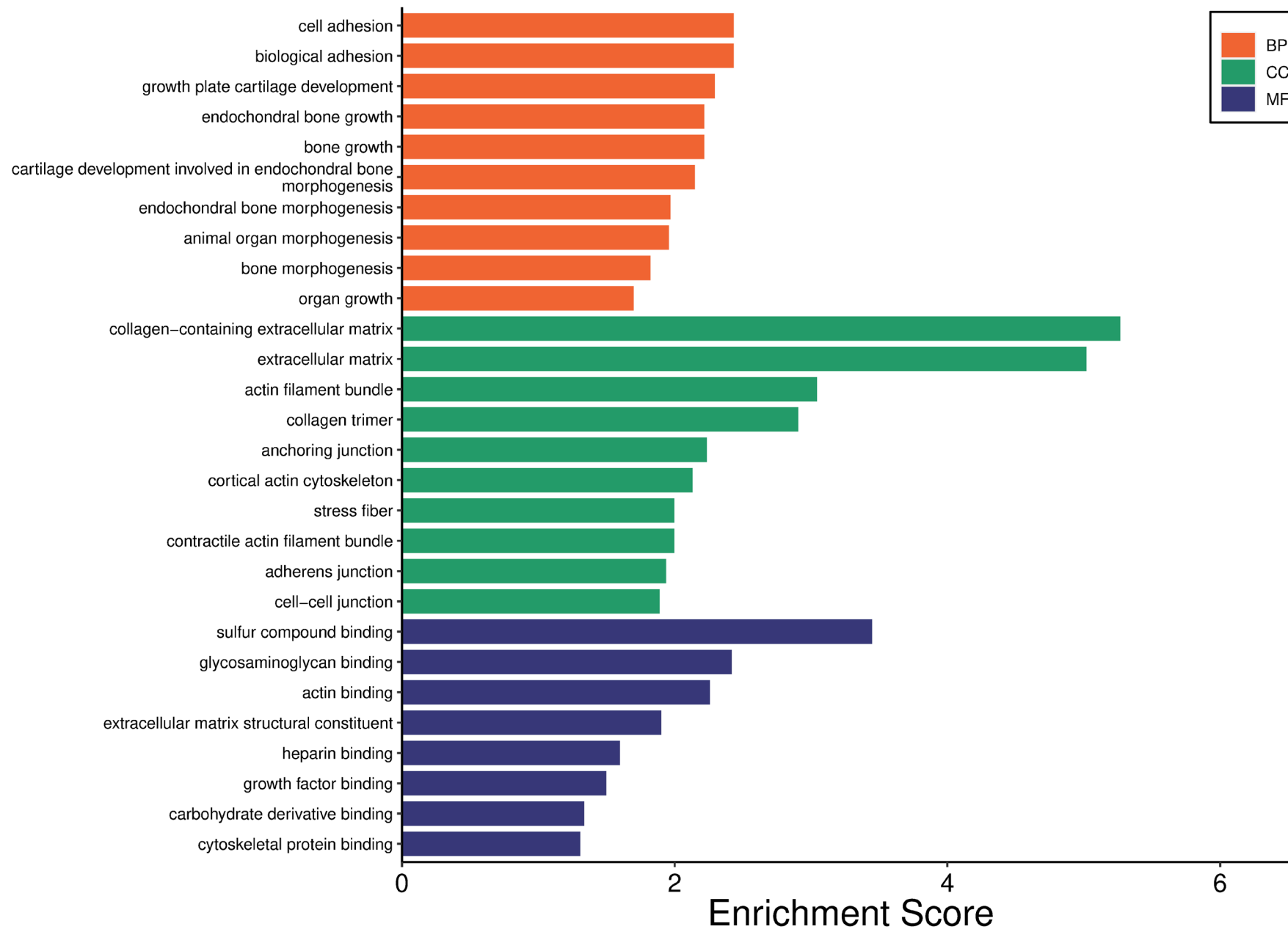
SYNr vs. Control



F2 generation

SYNCHs vs. Control

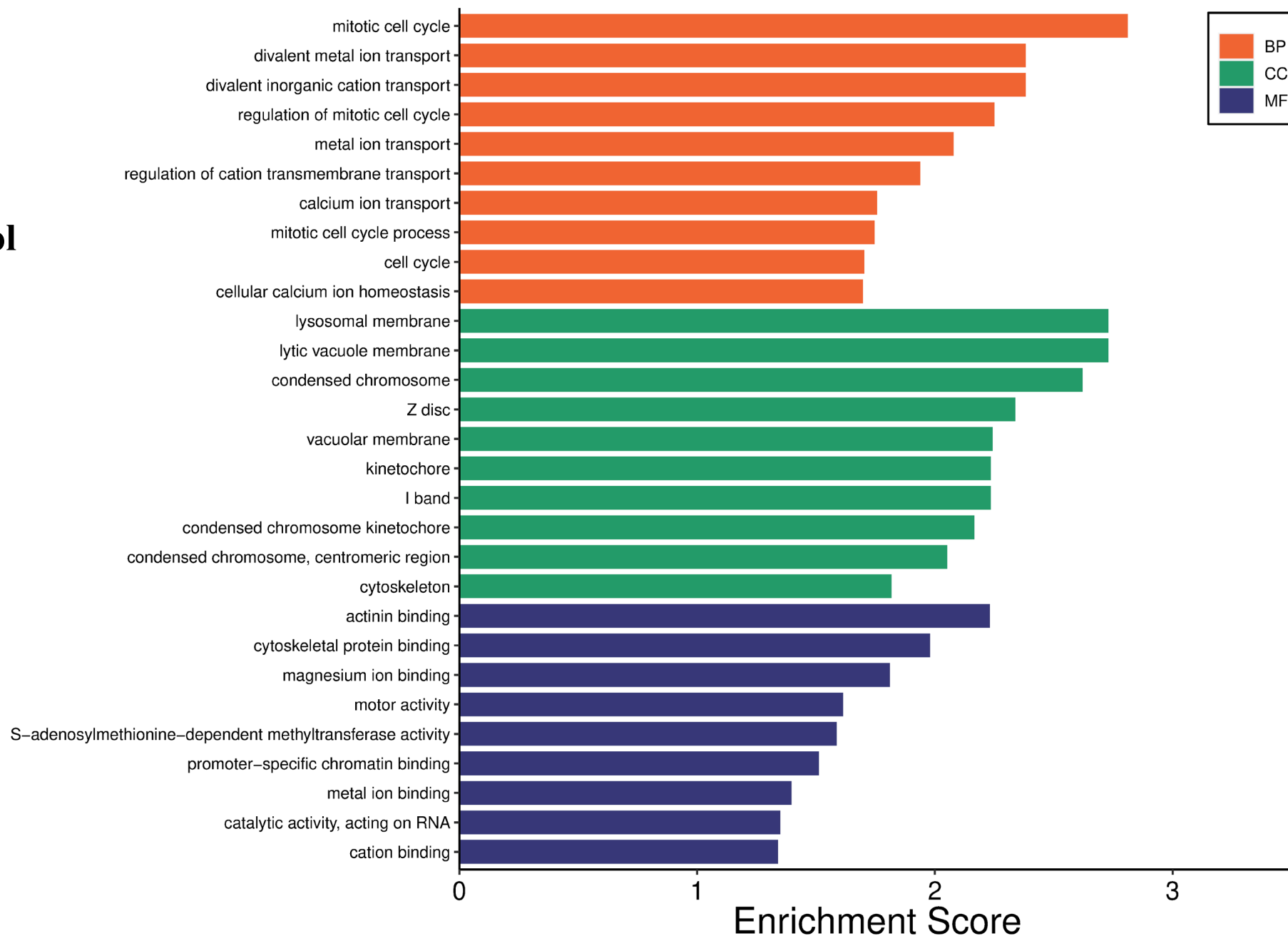
GO Results of Three Ontologies



F2 generation

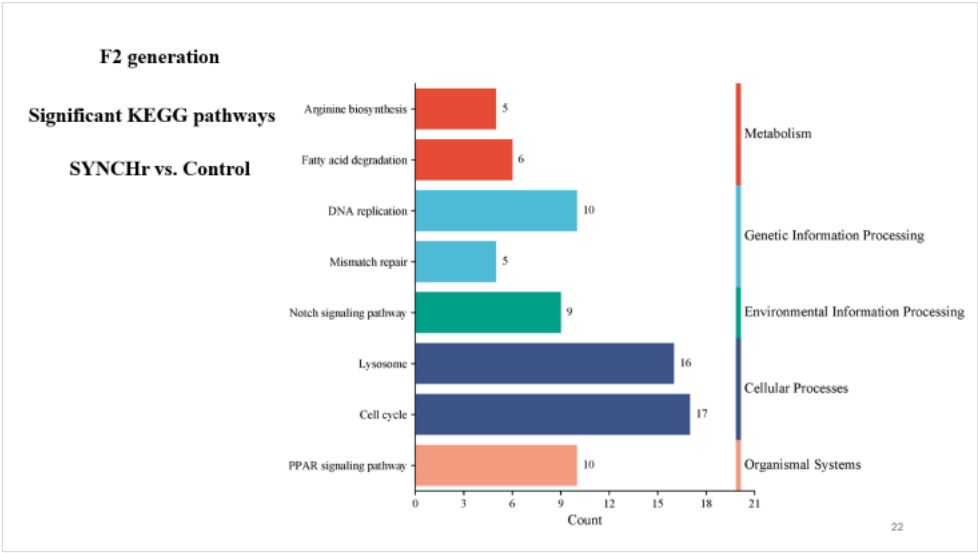
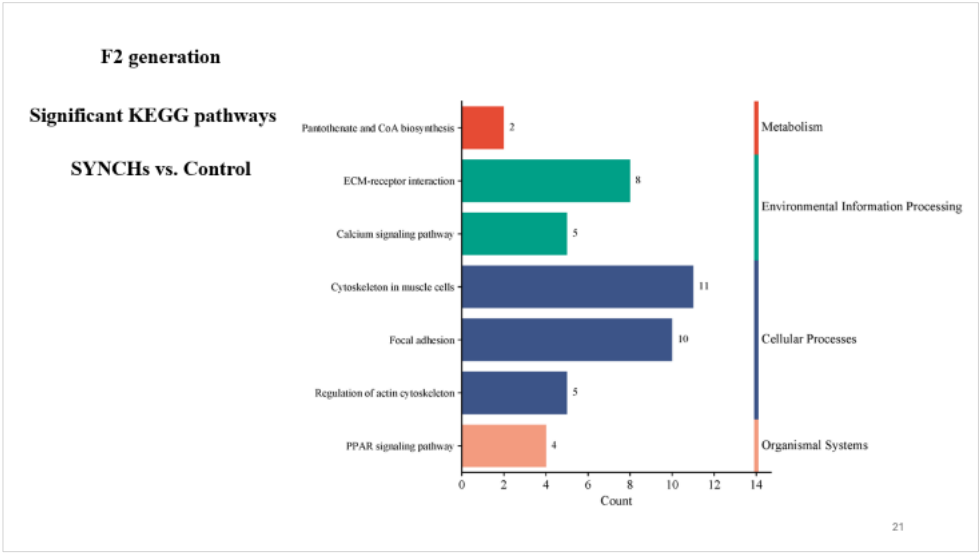
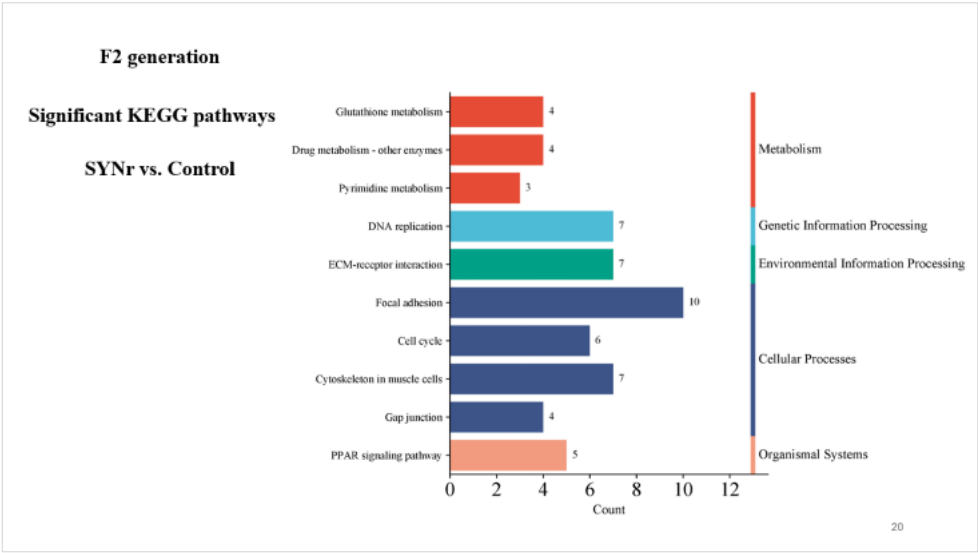
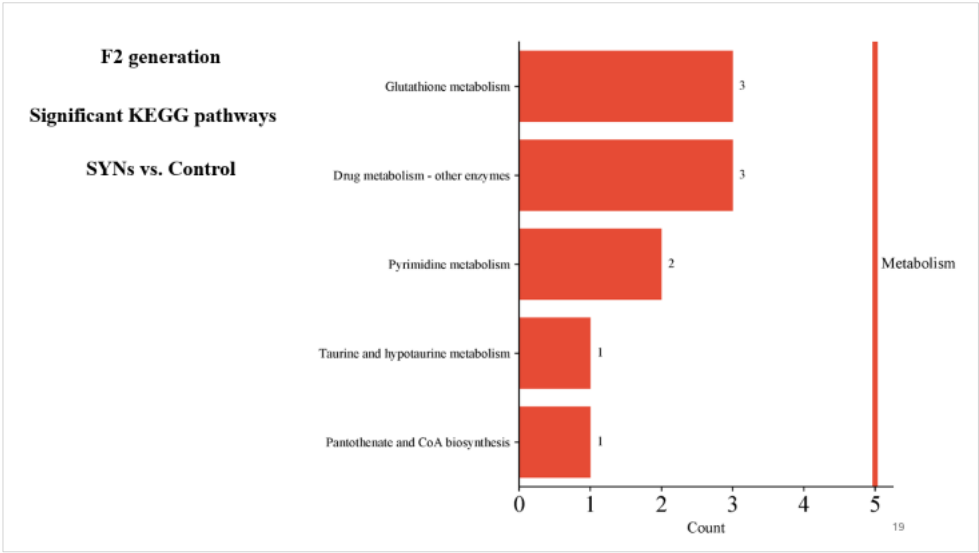
SYNCHr vs. Control

GO Results of Three Ontologies



KEGG Pathways

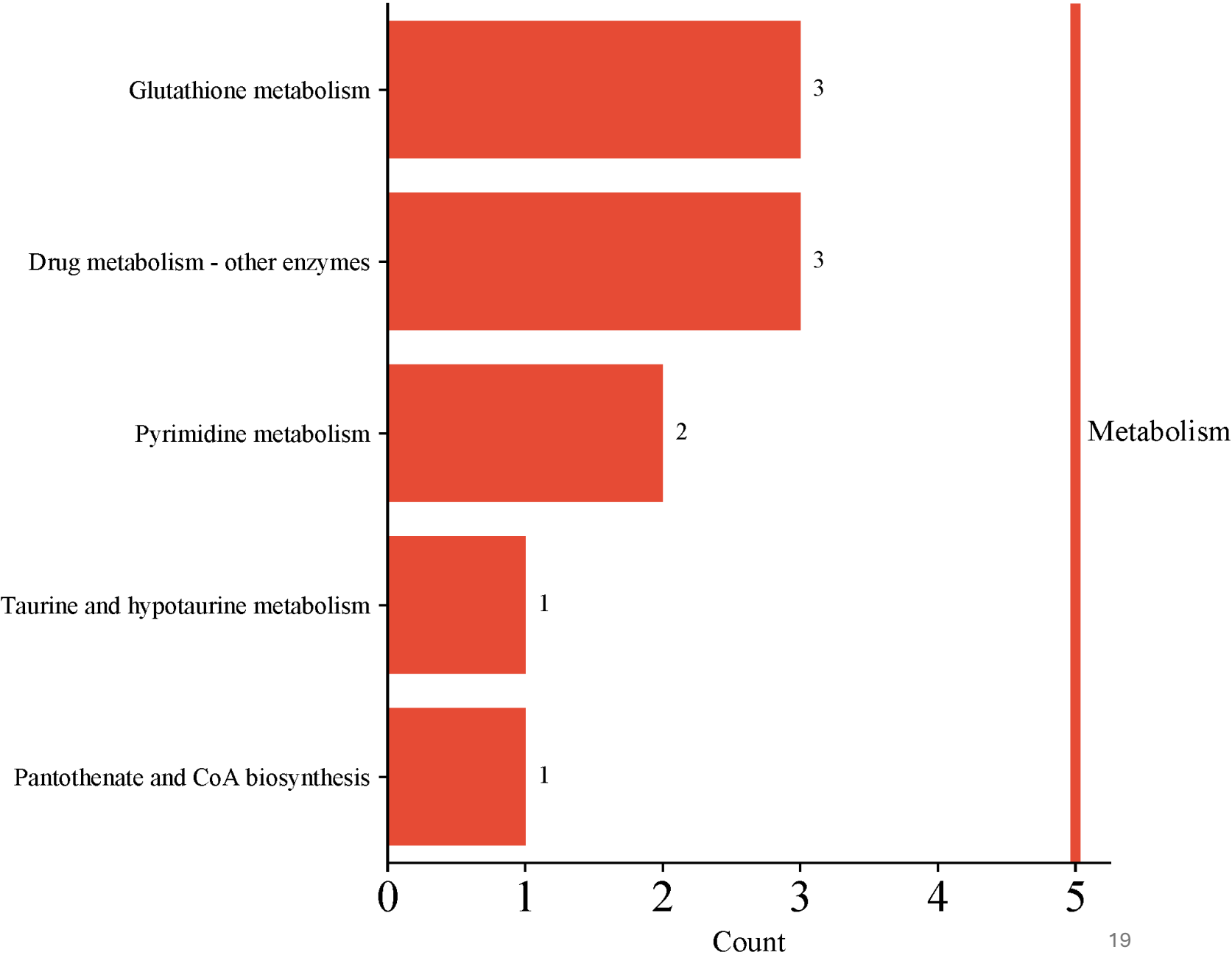
F2 generation



F2 generation

Significant KEGG pathways

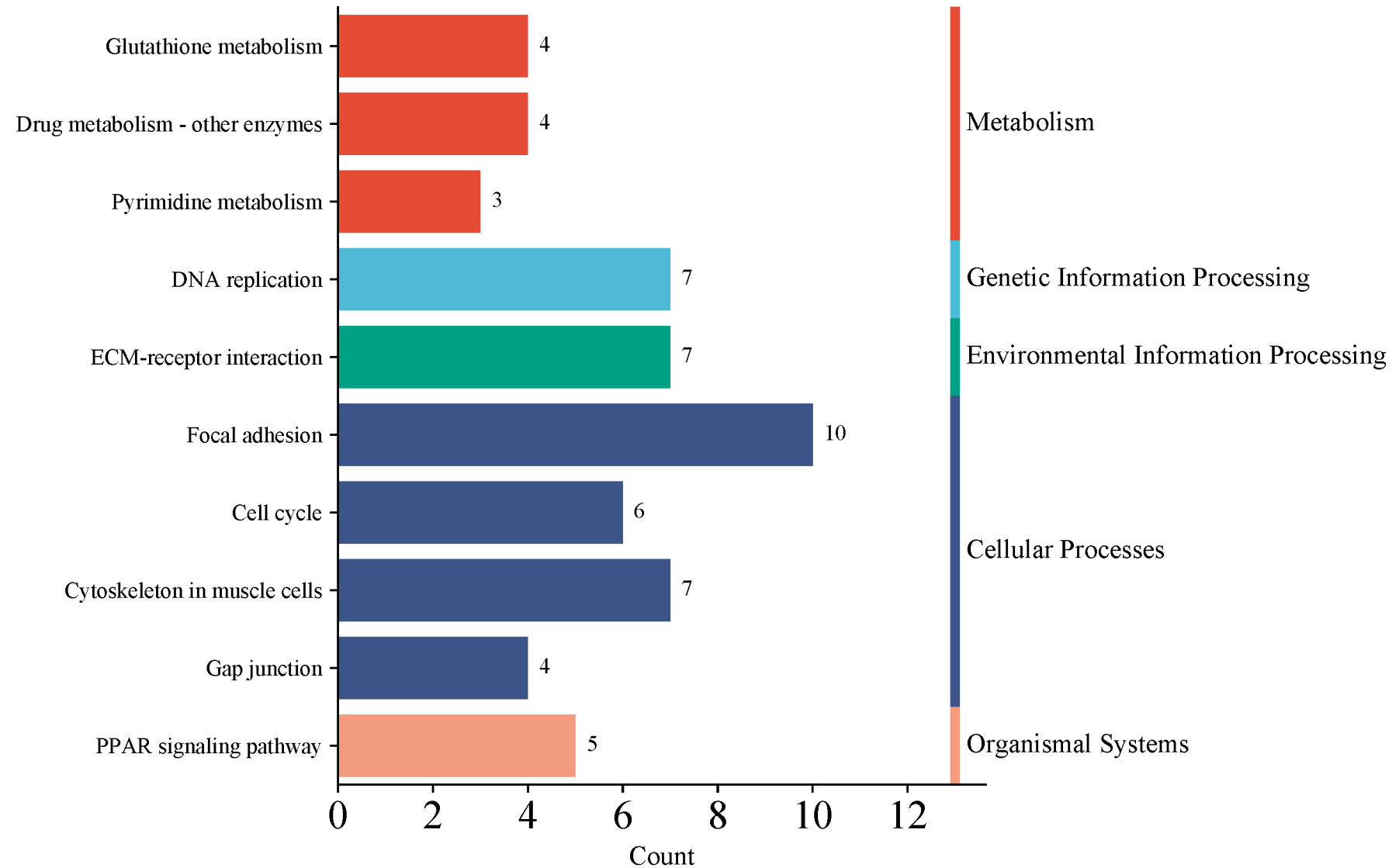
SYNs vs. Control



F2 generation

Significant KEGG pathways

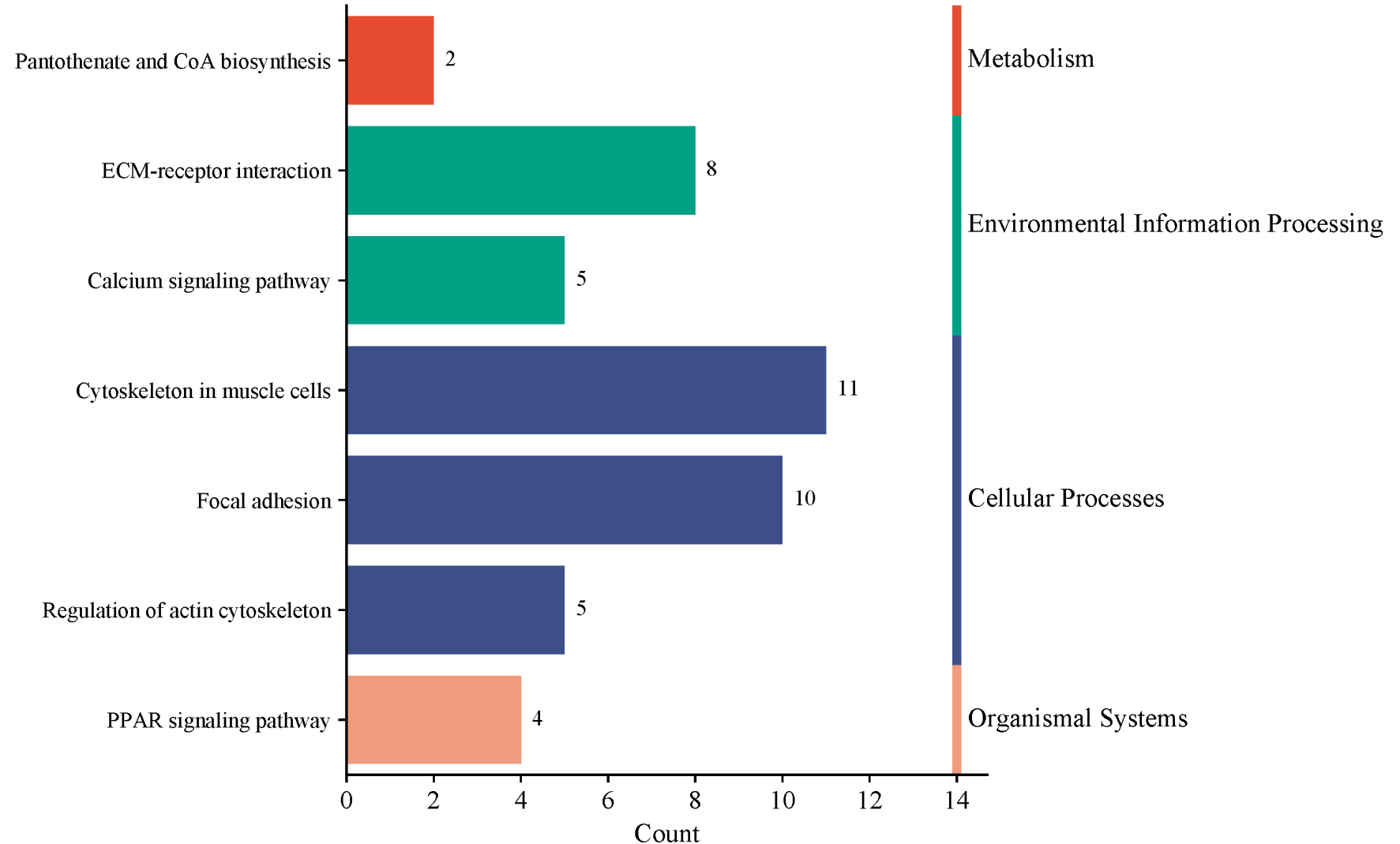
SYNr vs. Control



F2 generation

Significant KEGG pathways

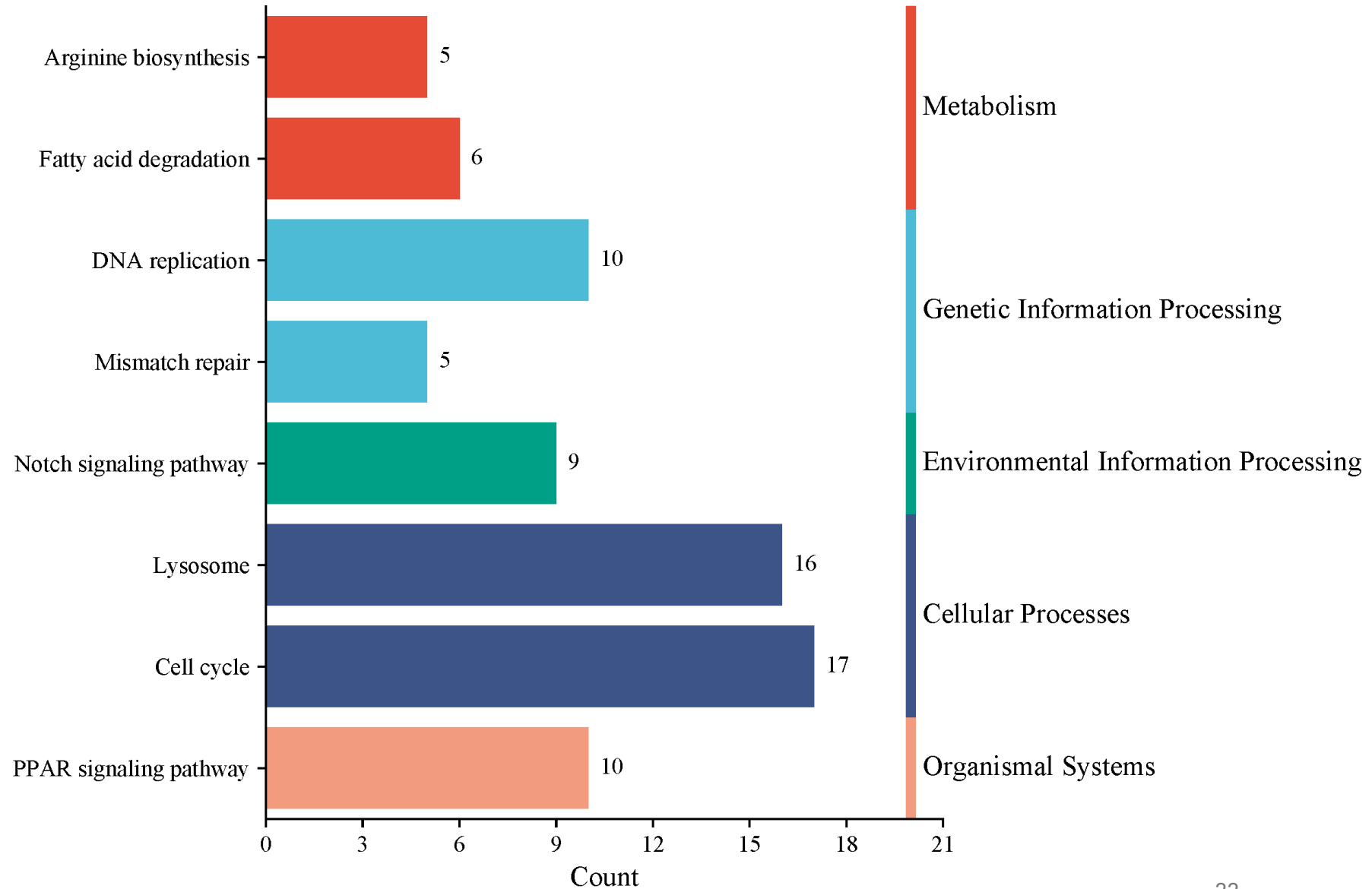
SYNCHs vs. Control



F2 generation

Significant KEGG pathways

SYNCHr vs. Control



5. Conclusion



Gene Expression Impact

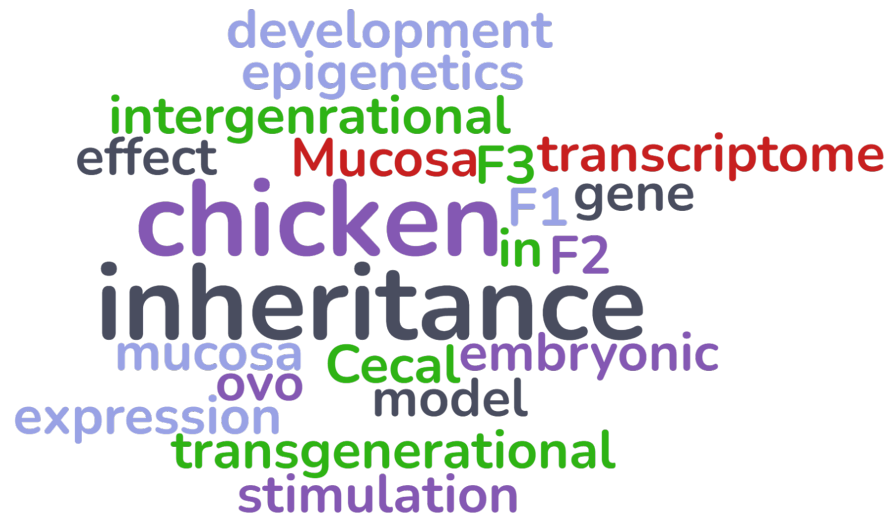
In ovo synbiotic and choline injections significantly alter gene expression across generations.

Intergenerational & Transgenerational Effects

Observed changes suggest both intergenerational and potential transgenerational effect on gene expression.

Cumulative Exposure Effects

Repeated injections across generations intensify gene expression changes, particularly in F2, but may stabilize by F3.

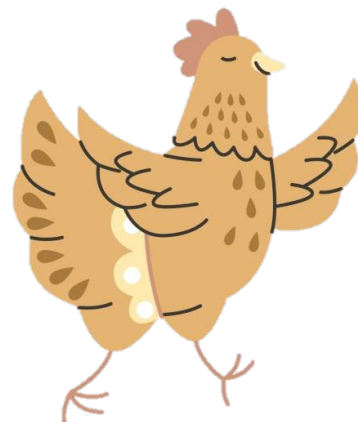


A word cloud visualization of key terms from the text. The words are arranged in a cluster, with 'chicken' and 'inheritance' being the largest and most central. Other prominent words include 'transcriptome', 'development', 'epigenetics', 'intergenerational', 'effect', 'Mucosa', 'F3', 'gene', 'F1', 'F2', 'mucosa', 'Ceca', 'embryonic', 'ovo', 'model', 'transgenerational', 'stimulation', 'expression', and 'in'. The words are colored in various shades of blue, green, red, and purple.

Implications for Supplementation

The in ovo stimulation may affect metabolic, cellular, and structural pathways, suggesting benefits for gut health and overall well-being by enhancing nutrient absorption, reducing inflammation, and improving cellular repair.

Thank you for your attention!



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This project is funded by the National Science Centre (NCN), Poland
NCN OPUS 19 grant no. 2020/37/B/NZ9/00497