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Platelet-Rich and Platelet-Poor Plasma Treatment Pre- and Post-Breeding Differentially Regulates Gene Expression in Early Equine Embryos from Mares Susceptible to Persistent Breeding-Induced Endometritis

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Abstract

Endometritis is a major cause of subfertility in mares, and platelet-rich plasma (PRP) and platelet-poor plasma (PPP) are used to modulate uterine inflammation and improve fertility. However, their effects on embryonic gene expression remain unknown. This study evaluated the transcriptomic profiles of embryos recovered from mares susceptible to persistent breeding-induced endometritis (PBIE) treated with PRP or PPP and compared them with controls. Treatments were administered by intrauterine

infusion during estrus. Each mare received four 40-mL intrauterine infusions of the assigned treatment at 48 and 24 h before breeding, and 6 and 24 h after breeding. Embryos underwent RNA sequencing and differential gene expression analysis. A total of 1,489 differentially expressed genes (DEGs) were identified in the PPP group (385 upregulated and 1,104 downregulated) compared to the controls, whereas 1,564 DEGs were identified in the PRP group (479 upregulated and 1,085 downregulated). Most DEGs were shared between the treatments, and no genes showed opposite expression patterns. However, PRP-treated mares produced embryos with increased expression of genes associated with cellular homeostasis and epithelial integrity and reduced expression of genes involved in metabolism and early development. In contrast, PPP-derived embryos showed increased expression of genes associated with developmental regulation and energy metabolism. These findings indicate that plasma-derived therapies modulate the embryonic transcriptome through their effect on the endometrium and that their effects are not solely dependent on platelet concentration.

In-brief: Platelet-rich and platelet-poor plasma induced distinct embryonic transcriptomic profiles in mares with endometritis. These findings provide new insights into the molecular basis of platelet-based therapies in equine reproduction.

Key-words: subfertility; RNA sequencing; equine embryo; platelet products.

1 Introduction

Endometritis is the leading cause of subfertility in mares and represents a major source of reproductive inefficiency in the equine industry. The condition may arise from infectious etiologies, including bacterial and fungal pathogens, or from a persistent and exaggerated post-breeding inflammatory response that fails to resolve following breeding. Persistent breeding-induced endometritis is characterized by prolonged uterine inflammation, impaired uterine clearance, and an altered endometrial environment that can negatively affect embryo survival, pregnancy establishment, and overall fertility. [1–3]. Conventional therapies for equine endometritis include the administration of anti-inflammatory agents, antimicrobials, uterine lavage, and ecbolic drugs to enhance uterine clearance [2]. Although these treatments are effective in many mares, a subset of individuals with chronic or recurrent endometritis fail to respond adequately to standard therapeutic protocols. Consequently, these challenging cases often require adjunctive therapies aimed at modulating the uterine inflammatory response, enhancing endometrial repair, and improving fertility outcomes.

Among these emerging therapies, blood-derived products such as PRP and PPP have gained considerable attention because of their anti-inflammatory, immunomodulatory, and regenerative properties. These products contain numerous bioactive molecules that may improve the uterine environment and enhance reproductive performance in mares susceptible to PBIE. Blood plasma and related products (e.g., platelet-rich-plasma and -hyperimmune plasma) have re-emerged as a prominent alternative therapy to mitigate PBIE via modulation of inflammatory cytokines and local antimicrobial properties in mares [1,4,5].

A healthy uterine environment plays an essential role in early embryonic development by providing nutrients and a variety of bioactive molecules, including growth factors, cytokines, lipids, and extracellular vesicles [6–8]. These molecules mediate the complex interactions between the conceptus and the maternal uterus that are essential for conceptus development and the establishment of pregnancy [9]. Therefore, alterations in the uterine environment caused by inflammation, such as endometritis, may negatively affect embryonic development [5,10]. Furthermore, these changes may influence embryonic gene expression and, consequently, the embryonic transcriptome [6].

RNA sequencing has become a powerful tool for evaluating gene expression under different biological conditions. This approach enables the identification of transcriptomic changes associated with both physiological and pathological processes [11]. In horses, transcriptomic analyses have been used to characterize changes in embryonic gene expression during maternal recognition of pregnancy and embryo fixation [6,9]. In addition, alterations in the platelet transcriptome have been described under various physiological and pathological conditions, suggesting that platelet-derived products possess biological functions beyond their traditional role in hemostasis [12,13].

Whether intrauterine administration of platelet-derived products, such as PRP and PPP, can alter the embryonic transcriptome remains unknown. Given the immunomodulatory and regenerative properties of these blood-derived therapies and their ability to modify the uterine environment, it is plausible that they may influence embryo development at the molecular level. However, the effects of uterine infusion

with PRP or PPP on embryonic gene expression in mares susceptible to persistent breeding-induced endometritis have not been investigated.

Therefore, the objectives of this study were to determine the effects of intrauterine administration of PRP and PPP on the transcriptomic profile of embryos recovered from PBIE-susceptible mares and to investigate whether platelet concentration influences the uterine inflammatory response and subsequent embryonic gene expression.

We hypothesized that intrauterine treatment with autologous blood plasma products, including PRP and PPP, would modulate the uterine environment of mares susceptible to PBIE, resulting in alterations in the embryonic transcriptome of recovered embryos. Furthermore, we hypothesized that differences in platelet concentration between plasma preparations would differentially influence the magnitude of uterine inflammation associated with PBIE and, consequently, embryo development and gene expression profiles.

2 Materials and methods

The study was conducted at the University of Illinois Veterinary Teaching Hospital using experimental mares maintained at the University of Illinois Veterinary Research Farm, University of Illinois Urbana-Champaign (Urbana, IL, USA). The mares were part of the university's research herd. Embryo collections were performed between August and December 2019. All animal procedures were approved by the Institutional Animal Care and Use Committee of the University of Illinois Urbana-

Champaign (Protocol No. 19141), and all methods were performed in accordance with the relevant institutional guidelines and regulations.

2.1 Embryo Collection and Experimental Design

The embryos analyzed in the present study were obtained from mares enrolled in a previously published randomized crossover trial that evaluated the effects of intrauterine platelet-rich plasma (PRP), platelet-poor plasma (PPP), and lactated Ringer's solution (control) in mares susceptible to persistent breeding-induced endometritis (PBIE) [1]. Twelve mares susceptible to PBIE were included in the study and assigned to receive PRP, PPP, and LRS during separate estrous cycles in a randomized crossover design. Platelet-rich plasma and platelet-poor plasma were prepared immediately before uterine infusion using a double-centrifugation technique [14]. The PRP preparation contained approximately $623 \times 10^3 \pm 144$ platelets/μL, corresponding to a 5.2-fold enrichment relative to baseline whole blood values, whereas PPP contained approximately $36 \times 10^3 \pm 25$ platelets/μL. Both plasma products demonstrated marked reductions in red blood cell (RBC) and white blood cell (WBC) concentrations while maintaining high platelet viability (approximately 97%).

During each estrous cycle, mares received four intrauterine infusions of 40 mL of their assigned treatment at 48 and 24 h before breeding and at 6 and 24 h after breeding. Ovulation was induced with 500 μg histrelin acetate after detection of a preovulatory follicle (≥33 mm) with uterine edema. Mares were inseminated with approximately 2×10^9 progressively motile fresh sperm from a fertile stallion, resulting in a total of 30 breeding cycles.

Embryos were recovered on Day 8 after ovulation by nonsurgical uterine lavage using 4 L of lactated Ringer's solution. A total of 23 embryos were recovered. After collection, embryos were examined under a stereomicroscope, measured, and morphologically graded according to the International Embryo Transfer Society (IETS) criteria. Immediately thereafter, each embryo was individually transferred to RNAlater™ (Thermo Fisher Scientific, Cat. No. AM7021) and stored at −80 °C until RNA extraction.

Detailed descriptions of animal selection, experimental design, breeding management, treatment administration, and sample collection procedures have been previously reported [5]. The present study evaluated the transcriptomic profile of embryos recovered from these treatment cycles to determine whether modulation of the uterine environment by PRP or PPP uterine treatment pre- and post-breeding influences gene expression of embryos collected from mares susceptible to PBIE.

2.2 RNA extraction

Embryos recovered from each treatment group were processed for RNA extraction using the Single Cell RNA Purification Kit (Norgen, Cat. No. 51800) according to the manufacturer's instructions. RNA concentration and integrity were subsequently assessed using the High Sensitivity RNA ScreenTape Assay on the TapeStation System® (Agilent Technologies, CA, USA). Of the 23 embryos recovered, 21 met the quality criteria for RNA sequencing, including four from the control group, eight from the PPP group, and nine from the PRP group.

Sequencing libraries were generated using the SMART-Seq® v4 Ultra® Low Input RNA Kit (Clontech, Cat. No. 635025) in combination with the Illumina Nextera

XT DNA Library Preparation Kit (Illumina, Cat. No. FC-131-1096), following the manufacturers' protocols (PARK et al., 2019). The resulting libraries were sequenced on the NovaSeq 6000 platform, generating approximately 9 Gb of 150-bp paired-end raw reads per sample.

2.3 Bioinformatics analysis

Raw sequencing reads were quality filtered and trimmed using Trimmomatic version 0.39 (Bolger, Lohse, and Usadel, 2014) to remove adapter sequences and low-quality bases. Trimmed reads were subsequently aligned to the telomere-to-telomere (TB-T2T) equine reference genome assembly (GCF_041296265.1) using STAR version 2.7.2a [15–17]. Read alignment was performed in two-pass mode using default parameters to improve splice junction detection and mapping accuracy.

Gene-level quantification of mapped reads was performed using FeatureCounts from the Subread package version 2.0.0 [18]. Read counting was conducted using the corresponding gene annotation file for the TB-T2T assembly, with only uniquely mapped reads assigned to genomic features. Strand specificity was specified according to the library preparation protocol.

Differential gene expression analysis was conducted using DESeq2 [19]. Genes with low expression across samples were excluded prior to analysis according to DESeq2 recommendations. Differentially expressed genes (DEGs) were identified using a false discovery rate (FDR)-adjusted P-value < 0.01 and an absolute fold change ≥ 1.5.

198 Functional enrichment analyses of DEGs were performed using the Kyoto
199 Encyclopedia of Genes and Genomes (KEGG) database and the PANTHER
200 Classification System version 13.0 [20]. Enriched pathways and gene ontology terms,
201 including biological processes, molecular functions, and cellular components, were
202 considered significant at an FDR-adjusted P-value < 0.05.

203

204

205 **3 Results**

206

207 Differential expression analysis of embryonic transcriptomes identified 1,489
208 differentially expressed genes (DEGs) in the platelet-poor plasma (PPP) group (385
209 upregulated and 1,104 downregulated) and 1,564 DEGs in the platelet-rich plasma
210 (PRP) group (479 upregulated and 1,085 downregulated), compared with untreated
211 controls. UpSet analysis revealed that 1,136 DEGs were shared between treatments,
212 representing 76.1% of PPP-associated and 72.5% of PRP-associated DEGs (Figure 1A).
213 Among these, 889 genes were commonly downregulated and 244 were commonly
214 upregulated, accounting for 80.5% and 81.9% of the downregulated genes and 63.4%
215 and 50.9% of the upregulated genes identified in the PPP and PRP groups, respectively
216 (Figure 1B). Each treatment also exhibited a subset of uniquely regulated genes (Figure
217 1B).

218 Volcano plots confirmed the extensive transcriptional changes induced by both
219 plasma treatments compared with controls (Figures 2 and 3). In both comparisons,
220 downregulated transcripts predominated, although PRP treatment resulted in a greater
221 number of upregulated genes than PPP. Several genes exhibited large fold changes and
222 statistical significance, supporting subsequent functional enrichment analyses.

223 Functional enrichment analysis identified both shared and treatment-specific
224 biological processes associated with PRP and PPP treatment (Figures 4 and 5). In the
225 PRP group, enriched Gene Ontology (GO) terms were primarily related to cellular
226 homeostasis, epithelial organization, regulation of biological processes, and intracellular
227 organelles (Figure 4). In contrast, PPP treatment preferentially enriched GO terms
228 associated with developmental processes, cellular metabolism, multicellular organismal
229 processes, and intracellular cellular components (Figure 5).

Among the transcripts differentially expressed following PRP treatment, genes associated with mitochondrial function and metabolism, including **FUOM** and **MTG1**, together with developmental regulators **VENTX** and **NKX1-2**, were downregulated, whereas genes involved in epithelial integrity and cell-supportive signaling pathways, including **PRAP1**, were upregulated (Figure 4). In contrast, PPP-treated embryos exhibited increased expression of **VENTX**, **NKX1-2**, **FUOM**, and **MTG1**, together with a downregulation profile that substantially overlapped with that observed following PRP treatment (Figure 5).

Principal component analysis (PCA) demonstrated close clustering of embryos from the PRP and PPP groups, indicating similar global transcriptomic profiles (Figure 6). The first two principal components explained 83% of the total variance, with PC1 and PC2 accounting for 67% and 16%, respectively. In contrast, embryos from the control group displayed a broader distribution and were primarily separated from treated embryos along PC1.

UpSet analysis further demonstrated that both plasma treatments elicited common and treatment-specific transcriptional responses (Figure 7). Among the downregulated DEGs, 889 genes were shared between the PRP and PPP groups, whereas 215 and 196 genes were uniquely downregulated following PPP and PRP treatment, respectively (Figure 7A). Similarly, 244 genes were commonly upregulated following both treatments, whereas 141 and 235 genes were uniquely upregulated in the PPP and PRP groups, respectively (Figure 7B). Only a single DEG distinguished the PRP and PPP treatment groups, and the combined UpSet analysis confirmed the extensive overlap in transcriptional responses between treatments (Figure 7C).

4 Discussion

Platelet-rich plasma (PRP) has attracted considerable interest in both human and veterinary medicine because of its regenerative, anti-inflammatory, immunomodulatory, angiogenic, and antimicrobial properties [21,22]. Consequently, PRP has been investigated as a therapeutic strategy for numerous reproductive disorders, including persistent breeding-induced endometritis (PBIE) in mares. However, the biological mechanisms underlying its beneficial effects remain incompletely understood, and it has generally been assumed that platelet-derived growth factors are the principal mediators of its therapeutic activity. The present study challenges this paradigm by demonstrating that both PRP and platelet-poor plasma (PPP) induced remarkably similar alterations in the embryonic transcriptome despite their marked differences in platelet concentration.

To our knowledge, this is the first study to demonstrate that modulation of the uterine environment with plasma-derived products during estrus influences global gene expression in Day 8 equine embryos. More than 70% of differentially expressed genes were shared between PRP- and PPP-treated embryos, principal component analysis demonstrated close clustering of both treatment groups, and no genes displayed reciprocal patterns of regulation between therapies. Collectively, these findings indicate that plasma-derived products induce highly concordant molecular responses during early embryonic development and suggest that platelet concentration alone is unlikely to explain their biological activity.

These observations are biologically plausible considering the complex composition of plasma. Although platelets are an important source of growth factors and

cytokines, plasma itself contains hundreds of proteins involved in innate immunity, complement activation, coagulation, oxidative stress, lipid transport, extracellular matrix remodeling, and cellular communication [23,24]. Many of these molecules participate directly in regulation of the uterine immune environment, tissue repair, and maternal-embryonic interactions. Therefore, the substantial overlap observed between PRP and PPP suggests that soluble plasma constituents may contribute as much as, or perhaps more than, platelet-derived factors to the modulation of the uterine environment following treatment.

Functional enrichment analysis further supported this concept by demonstrating that both plasma-derived products influenced similar biological pathways while retaining distinct molecular signatures. PRP preferentially enriched Gene Ontology categories associated with cellular homeostasis, epithelial organization, and intracellular organelles, whereas PPP was more strongly associated with developmental processes, cellular metabolism, and multicellular organismal organization. Importantly, these differences were quantitative rather than qualitative, as the overall transcriptional responses remained highly concordant. This pattern suggests that PRP and PPP may regulate common biological pathways through different degrees of activation rather than through fundamentally different mechanisms of action.

The differential expression of several representative genes further illustrates these treatment-specific responses. PRAP1 was upregulated following PRP treatment and has previously been associated with epithelial homeostasis, p53-mediated DNA damage responses, and negative regulation of apoptosis [25,26]. Although the biological role of PRAP1 during equine embryogenesis remains unknown, increased expression

may reflect alterations in cellular homeostasis and adaptive responses to the uterine environment. Conversely, genes involved in mitochondrial biology and cellular metabolism, including **FUOM** and **MTG1**, together with developmental transcription factors such as **VENTX** and **NKX1-2**, exhibited differential regulation between treatments [27–33]. These observations suggest that PRP and PPP influence metabolic and developmental transcriptional networks; however, transcriptomic analyses alone cannot determine whether these molecular differences translate into altered embryo physiology or developmental potential. Accordingly, these findings should be interpreted as indicators of differential pathway regulation rather than direct evidence of improved embryo quality.

One of the most clinically relevant findings of this study is that PPP produced embryonic transcriptional responses that closely resembled those induced by PRP. This observation challenges the widespread assumption that platelet enrichment is the primary determinant of therapeutic efficacy. If confirmed by future reproductive outcome studies, these findings suggest that PPP may represent a practical alternative to PRP for the management of mares susceptible to PBIE. Because PPP is simpler, less expensive, and more rapidly prepared than PRP, comparable biological activity would have important practical implications for clinical reproductive medicine and could facilitate broader implementation of plasma-derived therapies in equine practice.

The mechanisms responsible for these transcriptomic alterations are likely indirect. Neither PRP nor PPP was administered directly to the embryo; instead, both products were infused into the uterine lumen before and after breeding. Consequently, the observed embryonic transcriptional changes most likely reflect modification of the

maternal uterine environment rather than direct exposure of the embryo to plasma-derived molecules. Persistent breeding-induced endometritis is characterized by dysregulated post-breeding inflammation and delayed uterine clearance, resulting in an environment that is less favorable for embryo development. By modulating the inflammatory and reparative milieu of the endometrium, plasma-derived products may alter the composition of uterine secretions encountered by the embryo during its passage through the reproductive tract. The present findings therefore provide molecular evidence that therapeutic modulation of the maternal uterus can influence embryonic gene expression during the earliest stages of pregnancy.

Several limitations should be considered when interpreting these findings. First, RNA sequencing provides information regarding transcriptional activity but does not directly measure protein abundance, metabolic function, or embryo viability. Second, embryos were evaluated at a single developmental stage, preventing assessment of dynamic changes in gene expression throughout early pregnancy. Third, although transcriptomic alterations were clearly demonstrated, the relationship between these molecular signatures and subsequent reproductive outcomes, including pregnancy establishment, fetal development, and live foal production, remains unknown. Future studies integrating transcriptomic, proteomic, metabolomic, and reproductive outcome data will be essential to determine the biological significance of these molecular changes.

Overall, the present study demonstrates that intrauterine plasma-derived therapies modify embryonic molecular programming in mares susceptible to persistent breeding-induced endometritis. Rather than supporting a model in which platelet-

derived growth factors alone account for the biological activity of PRP, the results suggest that the therapeutic effects of plasma preparations arise from the coordinated action of multiple plasma-derived bioactive molecules acting on the uterine environment. This expanded perspective provides a framework for future investigations aimed at identifying the specific maternal pathways responsible for improving uterine health and supporting early embryonic development.

5. Conclusion

In conclusion, intrauterine administration of platelet-rich plasma and platelet-poor plasma during estrus induced highly similar alterations in the embryonic transcriptome of Day 8 equine embryos recovered from mares susceptible to persistent breeding-induced endometritis. Although PRP and PPP differed in the expression of selected genes involved in cellular homeostasis, metabolism, mitochondrial function, and developmental regulation, the majority of transcriptional responses were shared, indicating that modulation of the uterine environment by plasma-derived therapies is largely independent of platelet concentration. Because these treatments were administered exclusively within the uterine lumen, the observed embryonic transcriptomic changes most likely reflect indirect effects mediated through the maternal reproductive tract rather than direct actions on the embryo. These findings provide novel evidence that therapeutic modification of the uterine environment can influence early embryonic molecular programming and identify plasma-derived products as promising tools for improving uterine health in mares susceptible to persistent breeding-induced endometritis. Future studies should determine whether these molecular alterations translate into improvements in embryo quality, pregnancy establishment, and foal outcomes.

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Data availability: The RNA sequencing data generated in this study have been deposited in the National Center for Biotechnology Information (NCBI) Sequence Read Archive (SRA) under BioProject accession PRJNA1504058. The data are currently not publicly accessible and will be made publicly available upon publication of this article at: https://www.ncbi.nlm.nih.gov/bioproject/PRJNA1504058.

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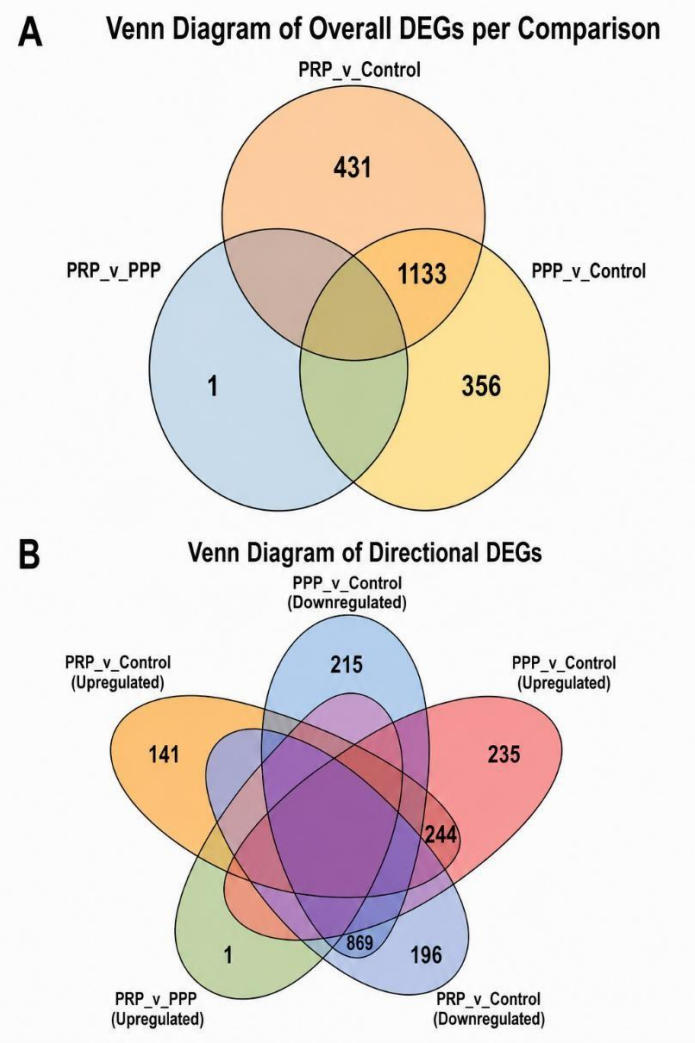
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558 **Figure 1. Overlap analysis of differentially expressed genes (DEGs).** (A)
559 UpSet plot showing the overall overlap of DEGs identified in embryos recovered
560 from mares treated with platelet-rich plasma (PRP) or platelet-poor plasma (PPP)
561 during estrus. (B) UpSet plots depicting the overlap of upregulated and
562 downregulated DEGs between treatments, illustrating shared and treatment-
563 specific transcriptional responses.

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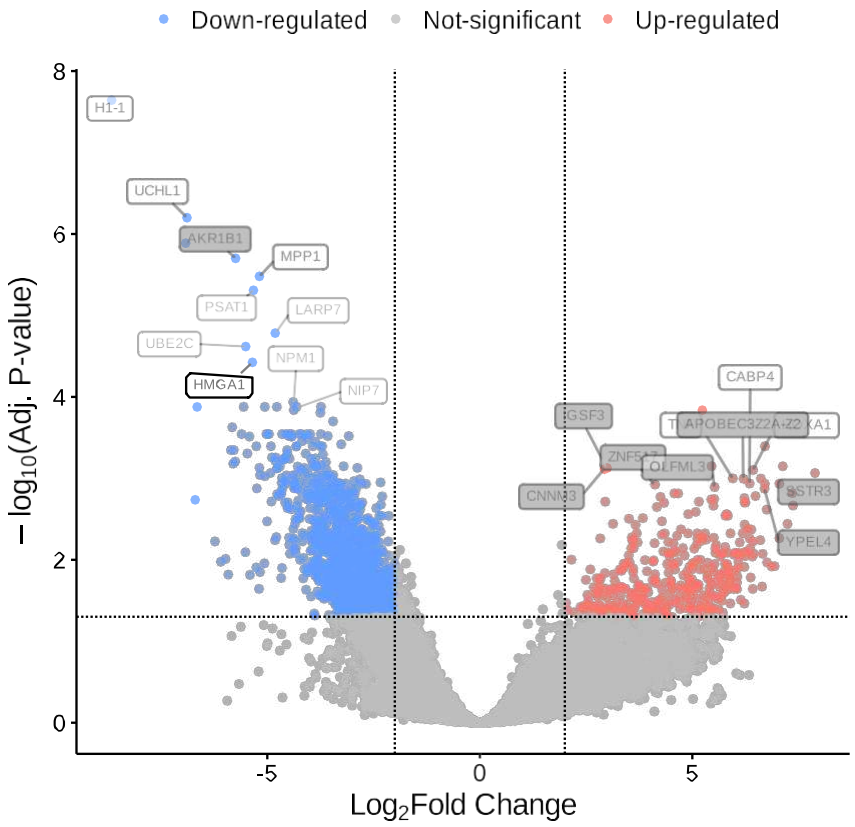


Figure 2. **Volcano plot showing differential gene expression** in Day 8 embryos recovered from mares treated with platelet-poor plasma (PPP) compared with control mares treated with lactated Ringer's solution. The x-axis represents the log2 fold change in gene expression, and the y-axis represents the −log10 adjusted P-value. Red dots indicate significantly upregulated genes, blue dots indicate significantly downregulated genes, and gray dots represent genes that were not differentially expressed. Differentially expressed genes were defined as those with an FDR-adjusted P-value < 0.01 and an absolute fold change ≥ 1.5. Selected genes with the greatest magnitude of differential expression are labeled.

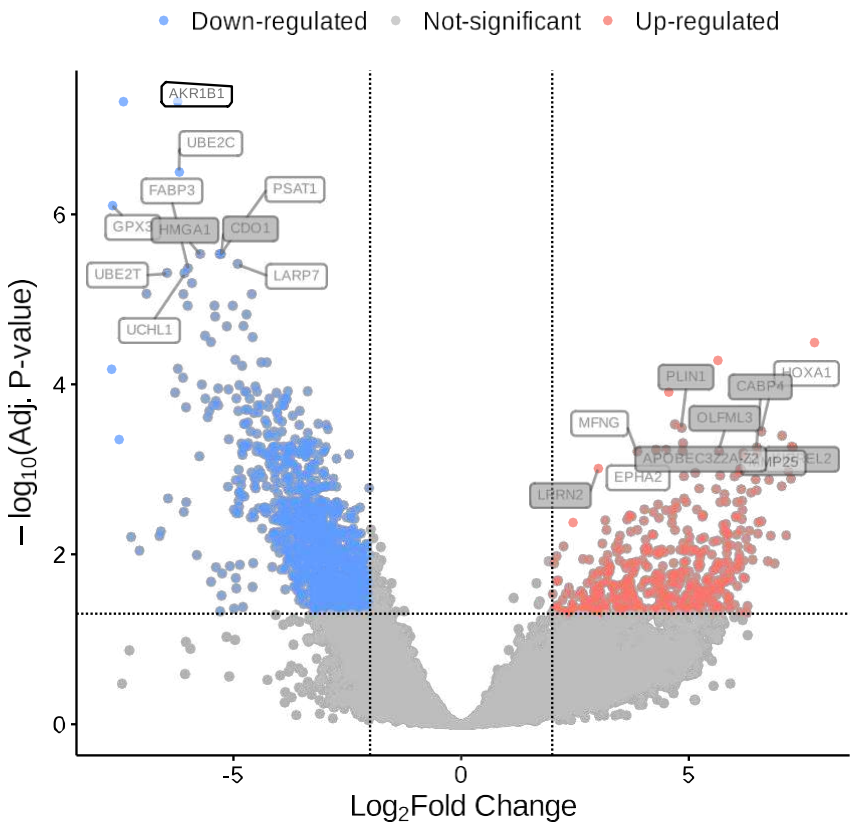


Figure 3. **Volcano plot showing differential gene expression** in Day 8 embryos recovered from mares treated with platelet-rich plasma (PRP) compared with control mares treated with lactated Ringer's solution. The x-axis represents the log2 fold change in gene expression, and the y-axis represents the −log10 adjusted P-value. Red dots indicate significantly upregulated genes, blue dots indicate significantly downregulated genes, and gray dots represent genes that were not differentially expressed. Differentially expressed genes were defined as those with an FDR-adjusted P-value < 0.01 and an absolute fold change ≥ 1.5. Selected genes with the greatest magnitude of differential expression are labeled.

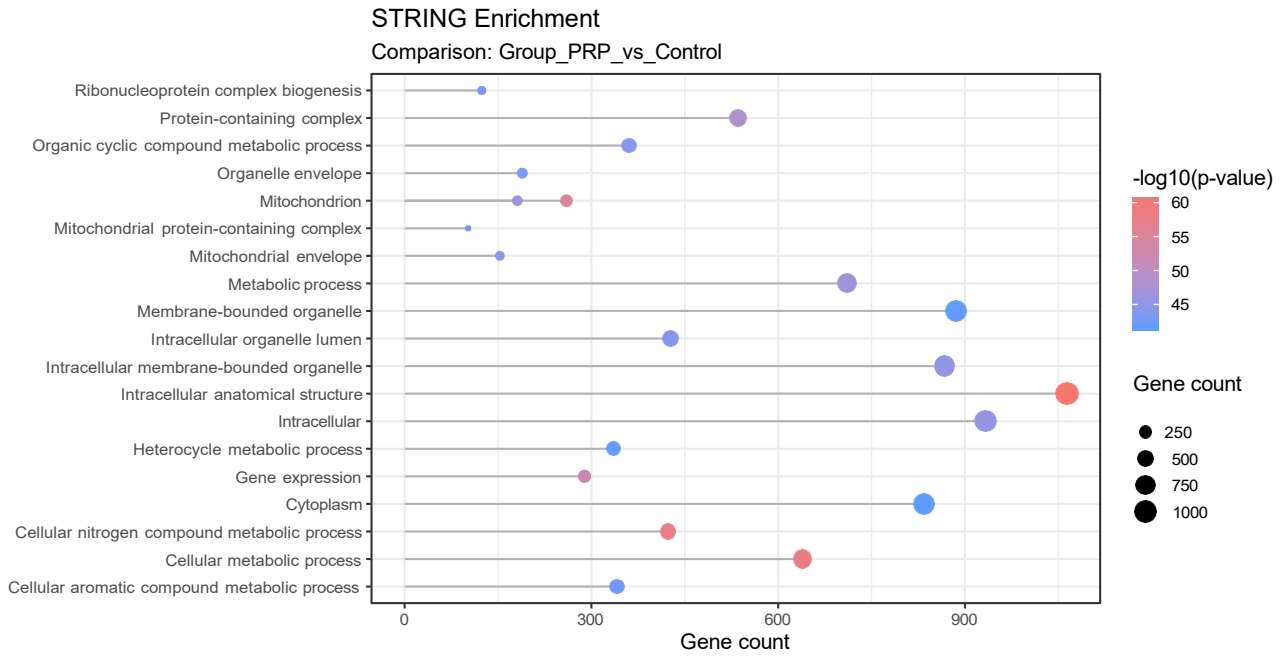


Figure 4. STRING functional enrichment analysis of differentially expressed genes (DEGs) identified in embryos recovered from mares treated with platelet-rich plasma (PRP) during estrus compared with untreated control mares. Functional enrichment was performed using the STRING database to identify significantly enriched Gene Ontology (GO) biological process and cellular component terms. Bubble size is proportional to the number of DEGs associated with each GO term, whereas bubble color represents the enrichment significance (−log10 *P* value), with warmer colors indicating greater statistical significance.

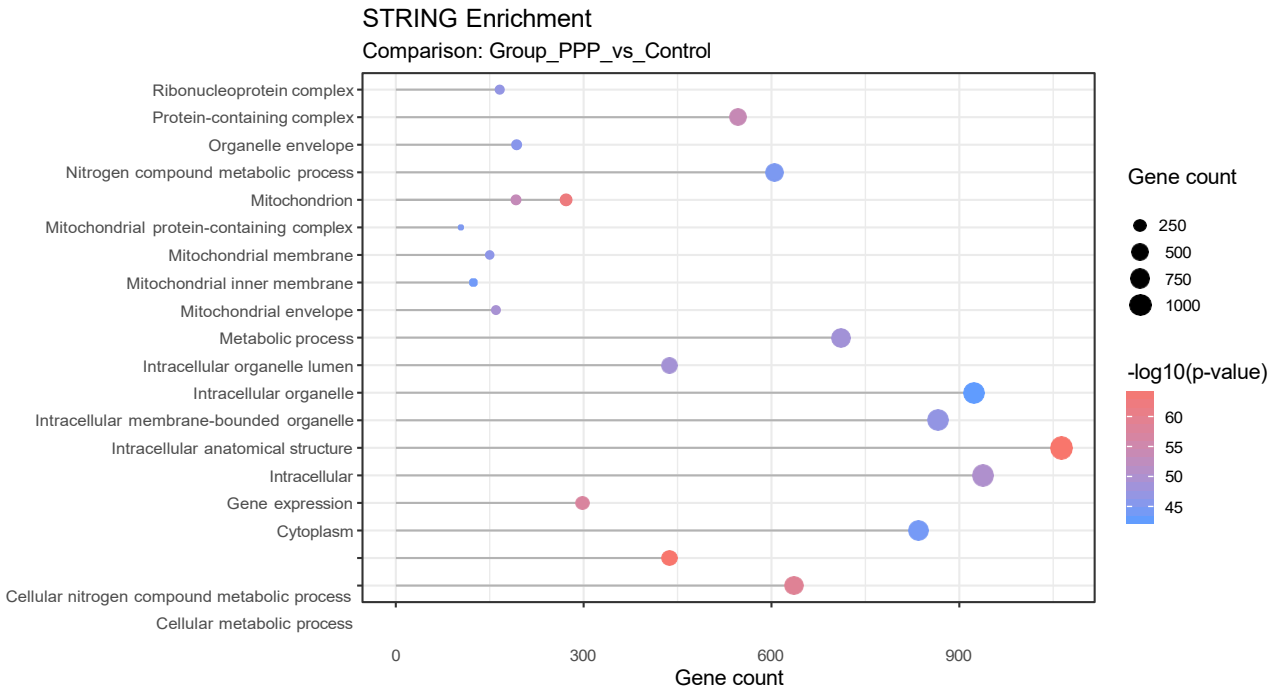


Figure 5. STRING functional enrichment analysis of differentially expressed genes (DEGs) identified in embryos recovered from mares treated with platelet-poor plasma (PPP) during estrus compared with untreated control mares. Functional enrichment was performed using the STRING database to identify significantly enriched Gene Ontology (GO) biological process and cellular component terms. Bubble size is proportional to the number of DEGs associated with each GO term, whereas bubble color represents the enrichment significance (−log10 *P* value), with warmer colors indicating greater statistical significance.

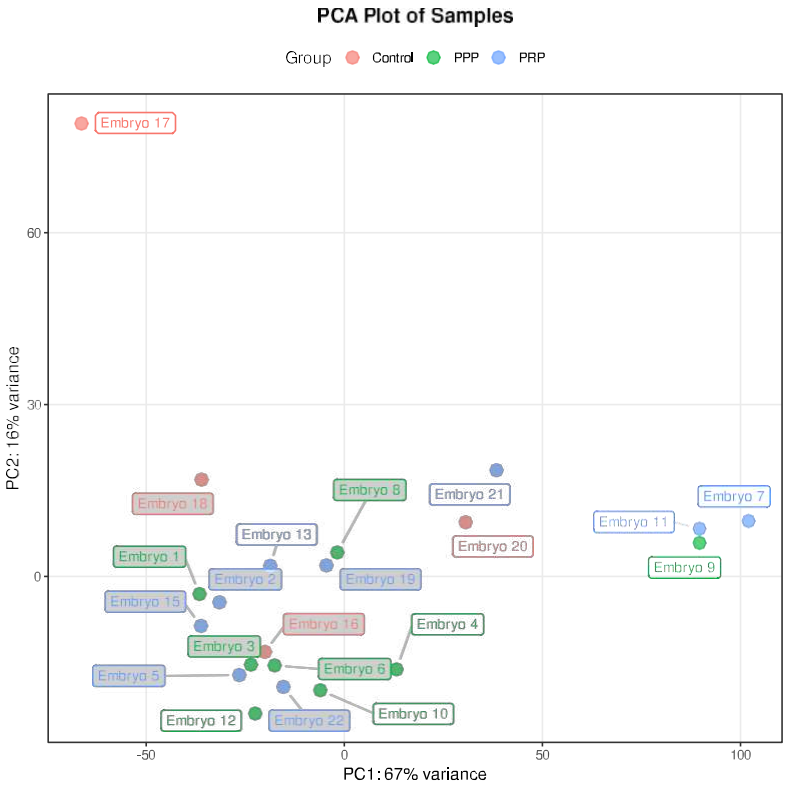
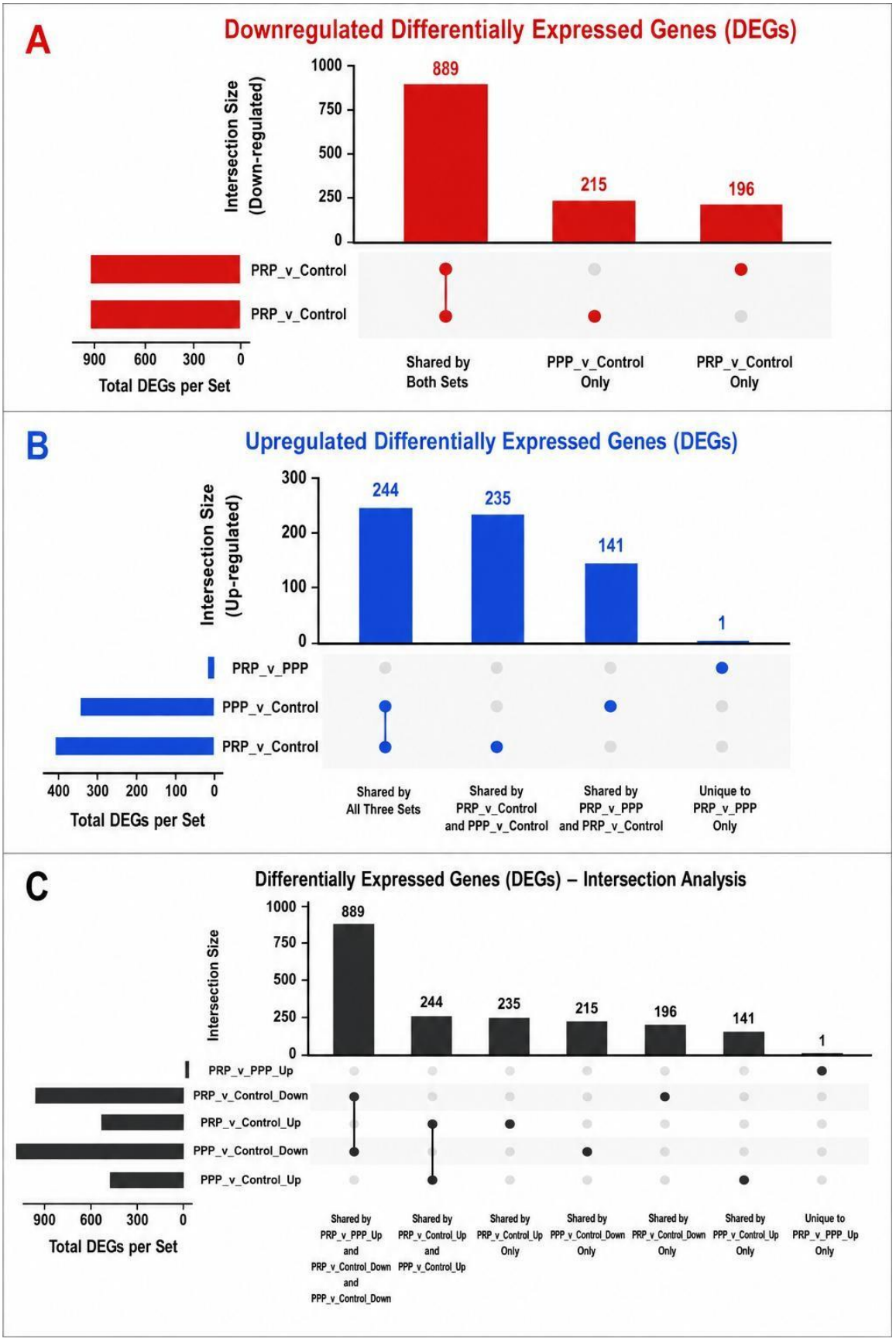


Figure 6. **Principal component analysis (PCA)** of normalized embryonic transcriptomes from embryos recovered from mares treated with platelet-rich plasma (PRP), platelet-poor plasma (PPP), or left untreated (control) during estrus. PCA was performed using normalized RNA-seq gene expression data to visualize overall transcriptomic relationships among samples. Each point represents an individual embryo, with colors indicating treatment group. The percentages shown on the axes correspond to the proportion of total variance explained by PC1 and PC2.


A
Downregulated Differentially Expressed Genes (DEGs)
Intersection Size (Down-regulated)
1000
750
500
250
0
889
215
196
PRP_v_Control
PRP_v_Control
900 600 300 0
Total DEGs per Set
Shared by Both Sets
PPP_v_Control Only
PRP_v_Control Only
B
Upregulated Differentially Expressed Genes (DEGs)
Intersection Size (Up-regulated)
300
200
100
0
244
235
141
1
PRP_v_PPP
PPP_v_Control
PRP_v_Control
400 300 200 100 0
Total DEGs per Set
Shared by All Three Sets
Shared by PRP_v_Control and PPP_v_Control
Shared by PRP_v_PPP and PRP_v_Control
Unique to PRP_v_PPP Only
C
Differentially Expressed Genes (DEGs) – Intersection Analysis
Intersection Size
1000
750
500
250
0
889
244
235
215
196
141
1
PRP_v_PPP_Up
PRP_v_Control_Down
PRP_v_Control_Up
PPP_v_Control_Down
PPP_v_Control_Up
900 600 300 0
Total DEGs per Set
Shared by PRP_v_PPP_Up and PRP_v_Control_Down and PPP_v_Control_Down
Shared by PRP_v_Control_Up and PPP_v_Control_Up
Shared by PRP_v_Control_Up Only
Shared by PPP_v_Control_Down Only
Shared by PRP_v_Control_Down Only
Shared by PPP_v_Control_Up Only
Unique to PRP_v_PPP_Up Only

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Figure 7. UpSet plots illustrating the overlap of differentially expressed genes (DEGs) identified in embryos recovered from mares treated with platelet-rich plasma (PRP) or platelet-poor plasma (PPP) during estrus compared with untreated control mares. (A) Overlap of upregulated DEGs. (B) Overlap of downregulated DEGs. (C) Overall overlap of DEGs. Horizontal bars indicate the total number of DEGs identified in each comparison, while vertical bars represent the number of shared and unique DEGs among comparisons. Connected dots beneath each vertical bar identify the comparisons contributing to each intersection.