

**Identification of uterine ionic transport proteins
involved in providing the mineral precursors for eggshell
formation in hens**

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- **Thème de recherche** (indiquer le n° choisi sur la liste) : 2
1 - Biologie cellulaire et Moléculaire. 2 - Biominéralisation.
3 - Développement et différenciation. 4 – Génétique et Physiologie.
5 - Ostéoimmunologie 6 – Biomatériaux

- **Le travail concourt au prix des jeunes chercheurs (Master/Thèse):** **OUI**

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- **Présentation** : **Etudiant (Master/Doctorant)** **Post-Doctorant**
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- **Proposer 4 mots clés:**
1- chicken
2- eggshell
3- uterus
4- ionic transport

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Identification of uterine ionic transport proteins involved in providing the mineral precursors for eggshell formation in hens.

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The bird eggshell is a complex bioceramic formed in the uterus segment of the chicken oviduct. The shell is made of 95% calcium carbonate (calcite) and 3.5% organic matrix made of proteins, glycoproteins and proteoglycans. The prerequisite for shell mineralization is the supply of large amounts of calcium and bicarbonate in the uterine fluid where calcification takes place. Both ions, are supplied by the blood via trans-epithelial transport in the uterus, and require ion channels, ion pumps and ion exchangers. The present study describes and updates the general model for ion transfers across the uterine tubular gland cells during eggshell formation. Our study used transcriptomic approach to reveal genes highly expressed in laying hen uterus during the active phase of mineralization, compared to the same period and tissue but following suppression of shell calcification by premature expulsion of the egg. This suppression abolishes the uterine egg mechanical stimulation and the calcium demand for shell formation. From the total list of genes over-expressed in uterus during eggshell calcification, we have identified 16 proteins related to ion transport updating the earlier model of ionic transfer.

Calcium is not stored in uterus but is continuously supplied from the blood. Hen uterine glandular cells transfer large amounts of calcium into the uterine lumen against the concentration gradient while preserving a low level of intracellular calcium. We observed that calbindin D28K was 17 times over-expressed during calcification. Low free calcium levels in the cell are maintained by calcium uptake by the endoplasmic reticulum via ATP depending calcium pumps as suggested by a 3 fold over-expression of ATP2A3 when the egg is being calcified. Finally, the calcium pumps ATP2B1 and ATP2B2, which can extrude calcium from the cytosol to an extracellular fluid against a strong electrochemical gradient, were 2 times over-expressed in our study, suggesting their involvement in the uterine Ca secretion. Carbonate of the eggshell is not derived from the blood HCO_3^- but rather from the plasma CO_2 , which is hydrated in the uterus to produce bicarbonate. Carbonic anhydrase 2 is over-expressed at a very high level during calcification and should play a pivotal role for conversion of intracellular CO_2 to HCO_3^- in hen uterus.

The transfer of calcium and bicarbonate from the blood to the uterine fluid involves additional ion transfers to maintain cell homeostasis. We reported here transporters involved in Na^+ , K^+ , Mg^{2+} , H^+ , and Cl^- exchanges to maintain physiological ionic concentrations in the cell.

This experimental approach has been used to reveal new candidate proteins and enrich the model describing ion transfers through the glandular cells of the uterus. Additional studies are still needed to quantify the respective role of ionic transporters which are controlling shell formation.