the disappearance of tensin from the cell adhesion sites of Chicken embryonic fibroblasts (CEFs) exposed to etoposide, coindicent with the disruption of antin cytoskeleton and morphological change during apoptosis. Furthermore, tensin was identified as a new substrate for caspase-3. Tensin cleavage is occurred by caspase-3 at DYPD1226G sequence during etoposide-induced apoptosis in vivo and after caspase-3 treatment in vitro and removes the actin binding domains and SH2 domain of the protein. The resulting tensin product is unable to bind phosphoinositide 3-kinase (PI 3-kinase) that is responsible for cell survival signaling. We also showed that expression of amino-terminal tensin fragment containing actin-binding regions induced disruption of actin cytoskeleton in CEFs. Thus, these results suggest that caspase-mediated cleavage of tensin contributes to the disruption of actin organization and interrupts survival signals through PI 3-kinase from extracellular matrix.

D104 Expression of Epithin in Mouse Preimplantation Development: Functional Role in Compaction

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The present study was performed to examine the expression and function of a mouse type II membrane serine protease, epithin during mouse preimplantation development. RT-PCR analysis showed that epithin mRNA was expressed throughout the cleavage stages from 1 cell zygote to blastocyst. Immunocytochemical study revealed that epithin protein was expressed at the blastomere junction. Inhibition of epithin synthesis by double strand RNA injection into 2 pronucleus stage embryo resulted in the block of the preimplantation development at 8 cell stage just before the compaction. Epithin was colocalized with E-cadherin at the membrane junction of the compacted morula stage embryo as revealed by double staining immunocytochemistry and confocal microscopy. Taken together, we demonstrate for the first time that epithin is expressed in mouse preimplantation embryo and might play an important role in the compaction process that precede the first differentiation into trophoectoderm and inner cell mass.

D105 Effect of Calcium Ion on mammalian period 1(mPer1) mRNA Expression in NIH-3T3 Cell Line

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Circadian rhythms have a periodicity of approximately 24 hours and are entrained by environmental cues among which light is the dominant regulator. In mammals, light/dark information is transmitted to suprachiasmatic nucleus *via* glutamate receptors which mediate Ca^{2+} influx. In the present study, calcium ionophore (A23187) was used as an environmental cue to examine the molecular mechanism of *mammalian period1* (*mPer1*) mRNA induction in NIH-3T 3cells in vitro. A23187 (1M) significantly increased *mPer1* mRNA levels and induction of *mPer1* mRNA expression by A23187 was blocked by actinomycin D, a transcriptional inhibitor, but not by puromycin, a protein synthesis inhibitor. These results suggest that *mPer1* mRNA is synthesized *de novo* and such *de novo* synthesis does not require any newly synthesized proteins. To examine the downstream signal mediators of Ca*2*+ effects, several specific inhibitors of protein kinase A (PKA), protein kinase C (PKC) and extracellular signal-regulated kinase (ERK) were used. However, no notable changes in the A23187-induced *mPer1* mRNA transcription were observed by pretreatment of these inhibitors. These

results suggest that intracellular Ca^{2+} may play an important role in induction of *mPer1* transcription in the mouse embryonic fibroblast NIH-3T3 cells.

D106 Organ Induction by Combined dose of bFGF and HGF on Xenopus Early Embryo

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Fibroblast growth factors(FGFs) are known to have multiple functions in early development of vertebratesincluing mesodermal formation, gastrulation and anteroposterial patterning. Hepatocyte growth factor(HGF) also functions in development and hepatic repair, but have no effect on organ induction from animal cap assay. In this study, the pattern of organ induction from *Xenopus* presumptive ectoderm by combined dose of bFGF and HGF were investigated. Explants were cultured in combined solutions for 3days, examined histologically, immunohistochemically, and were analysed with PT-PCR. As a result, the synergistic effect was seen in the combined dose of bFGF and HGF rather than in the single dose of bFGF. Eyes were developed at high rates in each concentration, 1-10ng/ml of bFGF and HGF respectively. RPE65 was detected both in induced eye and normal embryonic eye with monoclonal antibodies, 40A11 and 25F5. The expression of retinal specific opsin and muscle actin were detected by RT-PCR in explants.

D107 Modulation of Thr-Phosphorylation of Integrin 1 during Muscle Differentiation

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We have previously demonstrated that the engagement of the integrin 71 in E63 skeletal muscle cells by anti-7 antibodies and laminin triggered transient elevations in the intracellular free Ca^{2+} concentration ($[Ca^{2+}]_i$) (Kwon et al., 2000). In this study, we extend these observations in an attempt to determine both the function of integrin phosphorylation and how phosphorylation levels are regulated. Okadaic acid treatment of E63 muscle cells induced increased phosphorylation of integrin b1A, inhibition of integrin activation, loss of focal adhesions and disruption of the stress fibers. In the presence of okadaic acid, PP2A association with integrin 1A was reduced while 1D remained bound with PP2A. Both coimmunoprecipitation and in vitro phosphatase assays revealed that dephosphorylation of residues TT788-789 in the 1A cytoplasmic domain was dependent upon PP2A activity. These results suggest that PP2A may be a primary regulator of integrin 1A Thr-phosphorylation and subsequent integrin activation. Taken together, we propose that dephosphorylation or deletion of residues TT788-789 in the cytoplasmic domain of integrin 1A may contribute to the linkage of integrins to focal adhesion proteins and induce integrin activation.

D108 The effects of melatonin on the in vitro immature mouse oocytes maturation and preimplantional embryos development

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Melatonin, secreted by the pineal gland, is known to regulate the ovary function and reproduction in mammalians. Several reports have demonstrated that melatonin is an efficient free radical scavenger and general antioxidant. The present study was done to clarify the effects of melatonin on mouse oocytes muturation and embryos development in vitro. Melatonin (10^{-10}, 10^{-8},