during development. The improvement coincides with the selective disappearance of the slowest component of synaptic decay. We recorded single channel properties of heterologously expressed zebrafish subunits α_{twi} along with β,δ,ϵ or γ subunits to identify the basis of this slowest component. $\alpha_{twi}\beta\delta\epsilon$ receptors exhibit a mean open time that corresponds to the intermediate time constant of *twister* decay. By contrast, the embryonic receptor isoform $\alpha_{twi}\beta\delta\gamma$ exhibited a greatly prolonged open time that corresponded to the slowest component of synaptic decay in *twister*. This result is unexpected given the similarity in mean open times for wild type αβδϵ (0.5 ms) and αβδγ (1.2 ms) receptors. We propose that behavioral and functional recovery results from the observed developmental decrease in γ subunit mRNA expression, facilitating a switch from $\alpha_{twi}\beta\delta\gamma$ to $\alpha_{twi}\beta\delta\epsilon$ receptors. In support of this idea, morpholino RNA knockdown of γ subunit expression improved swimming performance and reduced the contribution of the slow component to synaptic current decay, presumably through an increase in $\alpha_{twi}\beta\delta\epsilon$ AChRs.

483-Pos Board B283
Kinetic Control of SNARE-Dependent Fusion by Accessory Factors and Calcium
Minjoung Kyoung, Ankita Srivastava, Yunxiang Zhang, Marija Vrljic, Patricia Grob, Eva Nogales, Steven Chu.
Ca^{2+} triggers neurotransmitter release to initiate signal transmission in all synapses. Synaptotagmin 1 is the Ca^{2+}-sensor for fast synchronous release, complexin is a modulator, and SNAREs are essential for synaptic vesicle fusion. Yet, the molecular mechanism of Ca^{2+}-triggering remains unclear. Here we describe the first successful reconstitution of this machinery at a single vesicle level that captures some of the kinetic and cooperative properties observed in vivo. Starting from a defined state of docked vesicles, simultaneous content and lipid mixing indicators differentiate between hemifusion and fusion. Upon Ca^{2+}-injection half of the docked donor vesicles fuse with acceptor vesicles within 0.27 secs when functional SNAREs, synaptotagmin 1, and complexin are reconstituted. In contrast, we observe a much slower response with SNAREs alone, or non-functional mutants of synaptotagmin 1 or complexin. However, fusion occurrence over a long time period is unaffected by these mutants, suggesting that synaptotagmin 1 and complexin are Ca^{2+}-controlled kinetic regulators of SNARE-induced fusion.

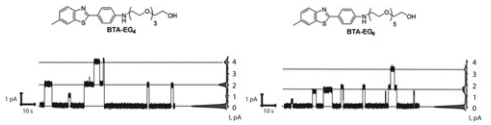
Ion Channels, Other

484-Pos Board B284
Ion Channels, Action Potentials and Ca^{2+} Handling in Human Pancreatic Beta-Cells. a Computational Approach
Leonid E. Fridlyand, Louis H. Philipson.
In this study, we examined the ionic mechanisms mediating depolarization-induced spike activity in human pancreatic beta-cells. We formulated a Hodgkin-Huxley-type ionic model for the action potential (AP) in these cells based on experimental voltage- and current-clamp results from our laboratory and literature. The model contains the equations for the currents: inward L-, P/Q and T-type Ca^{2+}, a ''rapid'' delayed rectifier K^+, the voltage-gated and Ca^{2+}-activated K^+ (BK-type), a voltage-independent Ca^{2+}-activated K^+ (SK-type), an ATP-sensitive K^+, a plasma membrane calcium pump, a voltage-gated Na^+ and a Na^+ background. Ionic model is coupled to an equation describing intracellular Ca^{2+} homeostasis. The model simulates the behavior of human beta-cell APs under a wide range of experimental conditions, including changes in the period and amplitude of AP in response to glucose challenge and changes in islet electrical activity due to K^+, Na^+ and Ca^+ channel blockade. The model was used to study the role of specific ionic currents in human pancreatic beta-cell firing. Particularly, modeling supports the importance of constitutively active tetrodotoxin-sensitive Na^+ and voltage-gated Ca^{2+}-activated K^+ channels (BK-type) in maintaining spontaneous spikes. This model provides acceptable fits to voltage-clamp, action potential and Ca^{2+} concentration data and can be used to seek biophysically based explanations of the electrophysiological activity and Ca^{2+} influx in human beta-cells for *in silico* analysis of physiological conditions and channel modulator actions.

485-Pos Board B285
Theoretical Biophysics of Ion Concentration Effects on the Affinity of Nanopore OmpF Lumen to Nucleotides
Saeid Hadi Alijanvand, Hamid Mobasheri.
The OmpF porin from the Escherichia coli outer membrane folds into a trimer of ß -barrels, each forming an aqueous pore allowing the passage of ions and small solutes. One of external loops, long L3 carrying multiple acidic residues folds into the ß -barrel pore to form a narrow ''constriction zone'' in the mid height of the channel. The intrinsic charge distribution in the conducting path may be affected by many factors. Nucleotides are known as one of the most important metabolites in the cell that can pass through membrane when they are needed. The experimental findings on translocation of such molecules through channels have provided useful information. Considering instrumental limitations , theoretical studies , modelings and simulations are applied. In this study we calculated the changes in the affinity of OmpF nanochannel lumen to pyrimidin and purine bases at molecular level by Delphi 4. A non-linear Poisson-Boltzmann calculator with definite element integrator found to be appropriate choice to calculate the net energy of OmpF-nucleotide interactions at different ion concentration. Our results indicate that decreasing in the ion concentration led to increase in the OmpF affinity to purine and pyrimidine bases. The calculation outputs indicated that OmpF- affinity for pyrimidine is higher than that of purine. These observations may shed light to understand the selective molecular translocation means implemented by OmpF.

486-Pos Board B286
A novel Gene Required for Male Fertility and Functional Catsper Channel Formation in Spermatozoa
Jean-Ju L. Chung, Betsy Navarro, Grigory Krapivinsky, Luba Krapivinsky, David E. Clapham.
Calcium (Ca2+) signaling is critical for successful fertilization. In spermatozoa, capacitation, hyperactivation of motility, and the acrosome reaction are mediated by changes in the intracellular Ca2+. Cation channels of sperm (CatSpers1-4) are alkalinization-activated Ca2+-selective ion channels controlling hyperactivated motility of spermatozoa and male fertility. CatSpers form a heteromeric complex with each of the CatSpers1-4 surrounding a Ca2+-selective pore, in analogy with other six-transmembrane (6TM) ion channel alpha subunits. In addition to the pore-forming proteins, the sperm Ca2+ channel contains auxiliary subunits, CatSper beta and CatSper gamma. Here, we identify the Tmem146 gene product as another novel CatSper accessory subunit, CatSper delta. We find that mice lacking the sperm tail-specific CatSper delta are infertile and their spermatozoa lack both CatSper current and hyperactivated motility. We show that CatSper delta has a central role in CatSper channel complex stability during spermatogenesis, providing new insight into the function of auxiliary subunits and the mechanism of CatSper channel complex assembly.

487-Pos Board B287
Self-Assembly of a Small Synthetic Molecule forms Well-Defined Ion Channels
Panchika Prangkio, Divya Rao, Mark Rubinshtein, Jerry Yang, Michael Mayer.
Synthetic ion channels are interesting for pharmacological and biomedical applications, including the development of antimicrobial agents, drug delivery vehicles, and biosensors. Previous work demonstrated that the attachment of polyethyleneglycol (PEG) groups to a molecular template such as cyclodextrin can lead to the formation of pore-forming molecules in lipid membranes. Here, we introduce small synthetic molecules based on oligo(ethylene glycol) and benzothiazol aniline (BTA) derivatives, which self-assemble in lipid bilayer to form ion channels with very-well defined ion conductances. Remarkably, these self-assembled ion pores were selective for monovalent cations and could be ''gated'' by changes in pH.



488-Pos Board B288
Energetics and Molecular Mechanisms of Permeation and Selectivity of Transport in the Urea Transporter
Giray Enkavi, Elena J. Levin, Ming Zhou, Emad Tajkhorshid.
Urea, ubiquitously used as a nitrogen source by bacteria and a safe end product of protein catabolism, depends on a specialized facilitator, urea transporter (UT) for its selective transport across the plasma membrane. Despite the name ''transporter'', UT has been suggested to operate by a channel like mechanism owing to its high transport rate. The crystal structure of a bacterial UT was reported recently as a homotrimer in the apo and substrate (dimethylurea; DMU)-bound states. However, important transport characteristics, such as urea binding sites and water permeability, have not been identified. To understand these transport properties of UT, we have performed extended equilibrium molecular dynamics simulations of the UT trimer with each monomer in one of the apo, DMU-bound, and urea-bound (modeled by Ming Zhou group) states, as well as umbrella sampling simulations of urea. These simulations allowed us to characterize expulsion