hindlimb elevation bone is resistant to the anabolic effects of IGF-I. In this context we wanted to know whether IGF-I could promote bone recovery during 2 weeks of reloading after a 2 week period hindlimb unloading.
Thirty six, 12 week old rats were divided into six groups; loaded (4 weeks), unloaded (4 weeks) and unloaded/reloaded (2/2 weeks), and treated with IGF-1 infusion (2.5 mg/kg/d) or vehicle during the final two weeks. Cortical bone volume (tibia) was assessed at the time of reloading and before sacrifice. Tibial fat free weight, cortical BFR, and bone histomorphometry of the tibial metaphysis were assessed at the end of the experiment.
Cortical bone volume did not change significantly during the four week experiment, a finding not surprising in such a short amount of time. However, tibial fat free weight was less in unloaded vehicle treated animals than in the loaded animals. Periosteal BFR decreased during unloading. During the 2 week recovery period in which skeletal loading was restored to normal, BFR in both the vehicle and IGF-I treated animals increased but the effect was greatly exaggerated in the animals treated with IGF-I. In IGF-I treated animals bone mass was restored more rapidly than in vehicle treated animals. These data show that unloading induces resistance to IGF-I, but that reloading after a period of skeletal unloading increases bone sensitivity to IGF-I.

CELL VOLUME, OSMOREGULATION AND WATER TRANSPORT (496.1-496.14)

496.1

PLA2 – a major regulator of volume-sensitive taurine release in NIH3T3 fibroblasts

Ian Henry Lambert. , Institute of Molecular Biology and Physiology, August Krogh Building, Universitetsparken 13, Copenhagen, DK2100, Denmark

Release of the organic osmolyte taurine efflux from NIH3T3 cells is increased by osmotic cell swelling in hypotonic medium and decreased by osmotic cell shrinkage in hypertonic medium. Release of arachidonic acid is increased under hypotonic conditions if oxidation of the fatty acid via the 5-lipoxygenase (5-LO) system is prevented by the 5-LO inhibitor ETH 615-139 and is reduced under hypertonic conditions. Exposure to the amphiphilic bee venom peptide melittin, which has no effect on the kinetic properties of PLA2 but promotes substrate replenishment, induces release of arachidonic acid and taurine in NIH3T3 cells under isotonic conditions. The potentiating effect of melittin on arachidonic release and taurine efflux is substantially increased in osmotically swollen cells and almost abolished in osmotic shrunken cells. H2O2 potentiates the melittin-induced taurine efflux under isotonic conditions but has only a minor effect on the melittin-induced taurine efflux under hypertonic conditions. Bromoenol lactone and manoalide, known inhibitors of Ca2+-independent phospholipase A2 (iPLA2) and secretory phospholipase A2 (sPLA2), respectively, reduce arachidonic acid and taurine release from NIH3T3 cells under hypotonic conditions and following addition of melittin. It is suggested that iPLA2/sPLA2 activity is responsible for the volume-sensitivity of taurine release in NIH3T3 mouse fibroblasts.

496.2

Guinea-pig Tracheal Epithelial Cell (EC) Shrinkage Induced by Hyperosmolar Challenge

Y. Jing[1], J. A. Dowdy[2], U. B. Ismailoglu[2], M. R. Van Scott[3], J. S. Fedan[1,2]. [1]Dept. Biochem. Molec. Pharmacol., WV Univ., 1 Med. Ctr. Dr., Morgantown, WV, 26506, [2]PPRB, NIOSH, 1095 Willowdale Road, Morgantown, WV, 26505, [3]Dept. Physiol., E. Carolina Univ., 600 Moyle Blvd., Greenville, NC, 27834

Hyperventilation during exercise is thought to increase the osmolarity of airway surface liquid. Hyperosmolar challenge of EC with mucosal D-mannitol (D-M) in methacholine (MCh)-contracted guinea-pig isolated, perfused trachea elicits smooth muscle relaxation mediated by epithelium-derived relaxing factor (EpDRF). To understand the relationship between EpDRF release and hyperosmolarity-induced changes in cell volume (CV), we examined CV changes in response to challenge of EC with 120 mosM D-M. EC were isolated from tracheas by protease digestion (2%; 1 h) and suspended in Krebs-Henseleit solution. Suspended EC remained viable for more than 5 h as indicated by active beating of cilia and exclusion of trypan blue (90 to 95%). CV was measured using a cell sizer. D-M induced a rapid decrease in CV (~9% shrinkage) during the first minute. The peak CV decrease occurred between 3 to 5 min (~22% shrinkage). No increase in CV was observed during the next 5 h. The results indicate that hyperosmolar conditions which cause EpDRF-mediated relaxation also evoke EC shrinkage. The rapid onset of the relaxation and CV responses could suggest that signaling pathways are shared. The findings and conclusions in this abstract have not been formally disseminated by the National Institute for Occupational Safety and Health and should not be construed to represent any agency determination of policy. Funded by NIOSH.

496.3

Effects of Na^+ and Cl^- Channel Blockers on Guinea-pig (GP) Isolated Tracheal epithelial cell (EC) shrinkage in response to hyperosmolar challenge

U. B. Ismailoglu[1], J. A. Dowdy[1], Y. Jing[2], M. R. Van Scott[3], J. S. Fedan[1,2]. [1]PPRB, NIOSH, 1095 Willowdale Road, Morgantown, WV, 26505, [2]Dept. Biochem. Molec. Pharmacol., WV Univ., 1 Med. Ctr. Dr., Morgantown, WV, 26506, [3]Dept. Physiol., E. Carolina Univ., 600 Moyle Blvd., Greenville, NC, 27834

In GP trachea that are precontracted with methacholine (MCh; $3H10^{-7}$ M), hyperosmolar D-mannitol (D-M) induces changes in epithelial Na^+ and Cl^- transport and smooth muscle relaxation mediated by epithelium-derived relaxing factor (EpDRF). We examined the effects of sequentially-added ion channel blockers, MCh and D-M (30 mosM) on EC cell volume (CV) changes in ECs isolated enzymatically (2% protease, 1 h) from GP-trachea and suspended in Krebs-Henseleit solution, under conditions in which these agents are used in the isolated, perfused trachea preparation to examine D-M-induced EpDRF release. CV was measured with a cell sizer. D-M elicited shrinkage of EC (~15 %) under conditions in which it caused EpDRF-mediated relaxation. The Na^+ channel blocker, amiloride (A; 10^{-4} M), did not affect EC CV. MCh had no effect on CV (±A). Shrinkage stimulated by D-M was unaffected by A. On the other hand, incubation with the Cl^- blocker, DIDS (10^{-4} M), resulted in an immediate and prolonged decrease in CV (~9%). Again, MCh had no effect on CV (±DIDS). However, in the presence of DIDS the CV decrease in response to D-M was diminished. Thus, the inhibitory effect of A on EpDRF release is not due to an effect on EC shrinkage, whereas the inhibitory effect of DIDS on EpDRF release may be linked to the decrease in the CV response. The findings and conclusions in this abstract have not been formally disseminated by the National Institute for Occupational Safety and Health and should not be construed to represent any agency determination of policy. Funded by NIOSH.

496.4

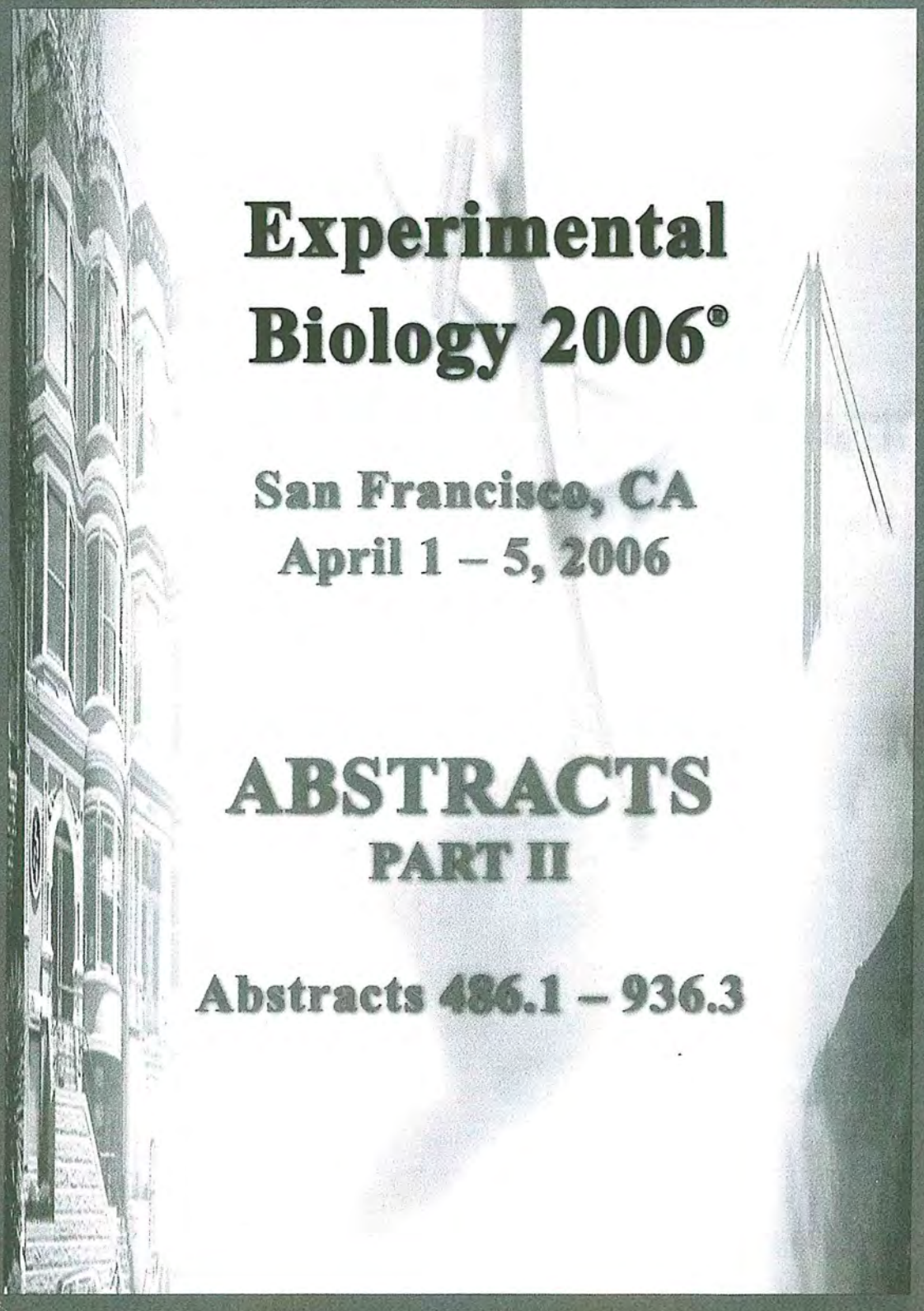
Swelling-activated K efflux and regulatory volume decrease efficiency in human bronchial epithelial cells

Adrian Caplanusi[1,2], Kwang-Jin Kim[3], Els L[redacted] Driessche[1], Danny Jans[4]. [1]K. U. Leuven, Labo[redacted] Campus Gasthuisberg, Leuven, B-3000, Be[redacted] Biochem., Carol Davila Univ. of Med. & Pharm., [redacted], Romania, [3]University of Southern California, Dept. of Medicine and Will Rogers Institute Pulmonary Research Cent[redacted] 90033, [4]Hasselt University, CMK, Laboratory of [redacted] 1D, Diepenbeek, B3590, Belgium

We studied the correlation between the cell swell[redacted] and regulatory volume decrease (RVD) in huma[redacted] cells 16HBE14o-. Cell thickness (as an index for cell volume) and K efflux (using Rb as a substitute for K) were monitored during 30 min hyposmotic shock (by reduction of both apical and basolateral osmolality to 0.17 osmol/kg water), followed by restoration of isosmotic conditions. In control conditions, hyposmotic stress increased cell volume by 35%, with subsequent RVD restoring cell volume to 94% of isosmotic control. Subsequent exposure of cells to the isosmotic solution decreased cell volume by 33%. Basolateral Rb efflux markedly increased during hyposmotic stress (from 0.50/min to a peak 6.32/min), while apical Rb efflux was negligible during these maneuvers. Rb efflux returned to the initial value when cells were re-exposed to isosmotic fluid. Gd, quinine, and 5-nitro-2-(3-phenyl-propylamino) benzoic acid (NPPB) all abolished RVD, while tyrphostin 23 and genistein blocked RVD with variable degrees. Activation of adenylate cyclase by forskolin hastened the recovery of cell volume with an enhancement in K efflux. These data taken together suggest that K extrusion during RVD may be

The FASEB Journal

The New Biology: Reports, Reviews, and FJ Express



Experimental Biology 2006®

San Francisco, CA
April 1 – 5, 2006

ABSTRACTS
PART II

Abstracts 486.1 – 936.3

March 7, 2006
Vol. 20, No. 5