Estrogen augments glucose transporter and IGF1 expression in primate cerebral cortex

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ABSTRACT Estrogen has many positive effects on neural tissue in experimental model systems, including stimulation of neurite growth and neurotransmitter synthesis and protection against diverse types of neural injury. In humans, estrogen treatment is reputed to protect against Alzheimer’s disease. To investigate potential mediators of estrogen’s action and determine whether selective estrogen receptor modulators (SERMs) such as tamoxifen have estrogen-like effects in the primate brain, we evaluated the expression of glucose transporters in the primate brain, we evaluated the expression of glucose transporters and insulin-like growth factor 1 (IGF1) and its receptor in the frontal cortex of ovariectomized rhesus monkeys. We treated each group for 3 days with vehicle, another with 17β estradiol (E2), and a third with tamoxifen. The expression of facilitative glucose transporters (Gluts) 1, 3, and 4 was investigated using in situ hybridization, immunohistochemistry, and immunoblot analysis. Gluts 3 and 4 were concentrated in cortical neurons and Glut1 in capillaries and glial cells. E2 treatment induced two- to fourfold increases in Glut3 and Glut4 mRNA levels and lesser but significant increases in Glut3 and 4 protein levels. E2 treatment induced an ~70% increase in parenchymal Glut1 mRNA levels, but did not appreciably affect vascular Glut1 gene expression. IGF1 and IGF1 receptor mRNAs were concentrated in cortical neurons in a distribution similar to Gluts 3 and 4. IGF1 mRNA levels were significantly increased in E2-treated animals but IGF1 receptor mRNA levels were not altered by hormone treatment. Tamoxifen increased cerebral cortical Glut3 and 4 mRNA levels, but did not affect Glut1, IGF1, or IGF1 receptor expression. This study provides novel data showing that Gluts 3 and 4 and IGF1 are coexpressed by primate cerebral cortical neurons, where their expression is enhanced by estrogen. These findings suggest that up-regulation of glucose transporter and IGF1 expression may contribute to estrogen’s salutary effects on neural tissue. Tamoxifen, an antiestrogen at the breast, is shown to have estrogen-like effects on higher brain centers in the monkey, suggesting that some SERMs may share estrogen’s neuroprotective potential for menopausal women.—Cheng, C., Cohen, M., Wang, J., Bondy, C. Estrogen augments glucose transporter and IGF1 expression in primate cerebral cortex. FASEB J. 15, 907–915 (2001)

Epidemiological studies suggest that estrogen treatment may reduce the risk and severity of Alzheimer’s disease in postmenopausal women (1–4). Experimental work has shown that estrogens have many trophic effects on neural tissue (reviewed in refs 4, 5), including promotion of neuronal survival (6, 7) and protection against toxic effects of oxidative stress (8, 9) and beta-amyloid (10–12). Estrogens also stimulate neurite outgrowth (13, 14) and expression of acetylcholine transferase and Trk-A (15). The broad range of estrogen’s salutary effects on neural tissue suggests there may be a fundamental anabolic mechanism underlying estrogen action on brain. Supporting this view, estrogen treatment is associated with increased cerebral metabolic activity in menopausal women (16, 17) and estrogen acutely enhances cerebral glucose utilization in rat brain (18), in which glucose utilization also fluctuates with endogenous estrogen levels during the estrus cycle (19).

The molecular mechanisms of neuronal glucose metabolism have been studied primarily in rodents. There are two major facilitative glucose transporters expressed in the murine brain: Glut 1 and 3. Glut1 is expressed by endothelial and glial cells and Glut3 by neurons (20–25). Glut1 is the major transporter of the blood–brain barrier (BBB; 26), which allows the facilitated diffusion of glucose from the bloodstream into the brain. Brain Glut1 expression is increased by ischemia (25, 27), systemic hypoglycemia (28), and estrogen (29). Experimental data concerning brain glucose transporter expression and regulation for the primate are relatively scarce; however, Glut1 expression is detected at the BBB (30, 31) and Glut3 mRNA has been reported in neurons (32).

There have been several recent reports that Glut4 is also expressed in the murine brain (33–36). Glut4 is the ‘insulin-sensitive’ transporter responsible for augmentation of glucose uptake by muscle and adipose tissue in response to insulin action (37). Little circulating insulin crosses the BBB (38, 39) and insulin synthesis is detected in only a few hypothalamic neurons (40), so finding the insulin responsive transporter in brain was somewhat puzzling. Insulin-like growth factor 1 (IGF1), however, is expressed in brain and has many...
insulin-like effects, including regulation of glucose transport (41–44). Moreover, the IGF1 and insulin receptors are homologous, with ~85% identity in the intracellular, signal-transducing portion of the receptors (45, 46). Since IGF1 is concentrated in the most metabolically active neurons (47), we have proposed that IGF1 functions as the brain’s endogenous insulin (48) and, similar to insulin, may regulate glucose transporter expression.

The present study aimed to elucidate the cellular patterns of expression for glucose transporters and IGF1 and its cognate receptor in the nonhuman primate brain, which is an important model for the human. Furthermore, we evaluated the effects of estradiol and a selective estrogen receptor modulator, tamoxifen, on glucose transporter and IGF1 expression, in order to evaluate the hypothesis that estrogen may stimulate fundamental anabolic processes of neural cells.

MATERIALS AND METHODS

Animals

Female rhesus monkeys (Macaca mulatta) 6–13 years of age from the NIH Poolesville colony were used in accordance with a protocol approved by the NICHID Animal Care and Use Committee. Ovariectomies were performed under ketamine anesthesia via a midventral laparotomy in the late follicular phases of their menstrual cycles. Three weeks after surgery, animals were randomly assigned to groups receiving vehicle (n=4), tamoxifen (n=5), or 17β estradiol (E2) containing pellets (n=4) inserted subcutaneously between their shoulder blades under ketamine anesthesia. The pellets contained 5 mg E2 and 50 mg tamoxifen in sustained release preparations (Innovative Research, Sarasota, Fla.). After 3 days, animals were killed with pentobarbital. Brains were removed and frontal cortices were dissected; the left was immersed in 10% formalin for 24 h, embedded in paraffin, and cut into 10 μm thick coronal sections. The right frontal cortex was frozen in dry ice and stored at ~70°C. A portion of this tissue was used for protein preparation and immunoblot analysis; the remainder was cut into 10 μm thickness at ~15°C and thaw-mounted onto poly-l-lysine coated slides for histochemical analysis.

In situ hybridization

The in situ hybridization protocol has been described (47) and the Glut1, 3, and 4, IGF1 receptor, and IGF1 cDNAs used for RNA probe synthesis have been described elsewhere (49, 50). After hybridization, sections were dipped in Kodak NTB2 emulsion; exposed at 4°C for 20 days, developed, fixed, and counterstained with hematoxylin. Parallel sections were hybridized to sense probes and exposed together with antisense hybridized sections. Quantitation of the hybridization signal was carried out in a blinded fashion. Hybridization signal was captured at 200× using a monochrome video camera. Signal was captured in cortical layers II and V using NIH image v1.57 software. The signal for Glut1 in capillary endothelial cells was counted at 400× under oil. Two to four sections from each brain were scored for each animal and two to three measurements were made in each section; thus, at least 4 and usually closer to 10 measurements were obtained and the mean determined for each animal. The values were compared among groups using ANOVA. Significant differences among means were determined by Fischer’s least significant difference test.

Immunohistochemistry

Immunohistochemistry for Glut1, 3, and 4 was performed by the avidin-biotin-immunoperoxidase technique, as described previously (51). Paraffin-embedded brain sections were deparaffinized in xylene, rehydrated, and permeated by trypsin digestion at 37°C for 10 min (Zymed, San Francisco, Calif.). Frozen brain sections were fixed in 4% formaldehyde for 10 min. After quenching in 3% H2O2 for 10 min, tissues were blocked in 10% normal serum for 30 min, followed by incubation with primary antibodies at 4°C overnight. Anti-Glut1, anti-Glut3, and anti-Glut4 were all used in 1:200 dilution and purchased from Alpha-Diagnostic (San Antonio, Tex.), Chemicon (Temecula, Calif.) and Biogenesis Inc. (Sandown, N.H.), respectively. After washing, sections were incubated with biotinylated secondary antibodies (1:400) for 30 min. The signal was detected and amplified using the ABC peroxidase method (Vector, Burlingame, Calif.) and visualized with 3,3’-diaminobenzidine. Paraffin-fixed sections were counterstained with hematoxylin and frozen sections were counterstained with methyl green. Controls for the immunohistochemistry procedure were performed by omitting primary antibodies in the incubation and substituting with blocking solution, then processing parallel with the experimental groups.

Immunoblotting

Immunoblotting was performed as described (51) with minor modifications. A small portion of frontal cortex (~1 g) adjacent to the region used for histochemical analysis was homogenized with a solution containing 0.32 M sucrose, 20 mM Tris (pH 7.4), 1 mM EDTA, 1 mM phenyl-methylsulfonal fluoride, 1 μg pepstatin A, 1 μg leupeptin, and 1 μg aprotinin in a ratio of 20 μl/g tissue weight. The membrane fractions were then prepared by centrifugation of brain protein homogenates at 100,000 g for 1 h. Pellets were washed in a Tris-EDTA-based solution as above, except no sucrose was added, and centrifuged again at 100,000 g for another hour. The resulting pellets were resuspended in a solution containing 0.1% Triton X-100, 20 mM Tris (pH 7.4), 1 mM EDTA, 1 mM phenyl-methylsulfonal fluoride, 1 μg/ml pepstatin A, leupeptin, and aprotinin. Thirty milligrams of cortex membrane proteins were loaded on precoated 10% SDS-polyacrylamide gels (Novex, San Diego, Calif.) and transferred to nitrocellulose membranes using an electrophoretic transfer cell (Bio-Rad, Hercules, Calif.). Anti-Glut1, anti-Glut3, and anti-Glut4 antibodies were purchased from companies as described above and used in 1:250, 1:1000, 1:200 dilution, respectively. Glut1 and 3 positive controls were total membrane protein homogenates at 100,000 g for 1 h. Pellets were washed in a Tris-EDTA-based solution as above, except no sucrose was added, and centrifuged again at 100,000 g for another hour. The resulting pellets were resuspended in a solution containing 0.1% Triton X-100, 20 mM Tris (pH 7.4), 1 mM EDTA, 1 mM phenyl-methylsulfonal fluoride, 1 μg/ml pepstatin A, leupeptin, and aprotinin. Thirty milligrams of cortex membrane proteins were loaded on precoated 10% SDS-polyacrylamide gels (Novex, San Diego, Calif.) and transferred to nitrocellulose membranes using an electrophoretic transfer cell (Bio-Rad, Hercules, Calif.). Anti-Glut1, anti-Glut3, and anti-Glut4 antibodies were purchased from companies as described above and used in 1:250, 1:1000, 1:200 dilution, respectively. Glut1 and 3 positive controls were total membrane protein fractions prepared from rat brains, kindly provided by Dr. Ian Simpson. The Glut4 protein standard was a recombinant protein synthesized from Glut4-HA clone, kindly provided by Dr. H. Chen (52). After incubation with horseradish peroxidase-linked secondary antibodies, protein bands were visualized on Kodak XAR film by ECL detection (Amersham, Cleveland, Ohio). A digital image of the immunoblots was made and the relative amounts of Glut-specific proteins in vehicle- and E2-treated group were compared using the NIH image program (Image 1.57, NIH).
RESULTS

Glut3 mRNA is widely expressed in primate cortical neurons but is not abundant in vehicle-treated ovariectomized monkeys (Fig. 1A, B). Estrogen treatment, however, results in marked increases in cortical Glut3 levels (Fig. 1C, D), which increased by almost threefold in cortical layer II and slightly less in layer V (Fig. 1E). Tamoxifen-treated animals demonstrate increased Glut3 expression that is less than that of E2 but still statistically significant (Fig. 1E). Glut4 mRNA is also concentrated in cortical neurons and, like Glut3, is quite low in ovariectomized monkey cortex (Fig. 2A, B). Glut4 mRNA expression is increased by four- to fivefold in E2-treated animals (Fig. 2C, D). Tamoxifen treatment also increased Glut4 expression, but the effects were of marginal statistical significance (Fig. 2E). Examination of adjacent sections from each brain hybridized alternately to Glut3 and Glut4 suggests that many cortical neurons express both transporters. The similar pattern of Glut3 and 4 expression in the cerebral cortex is not attributable to cross-hybridization of the RNA probes, which produce distinct hybridization patterns in other tissues (49, 50). Moreover, the Glut4 cDNA template used for probe synthesis (corresponding to nucleotides 1575–1939 of rat cDNA) in these studies has less than 40% homology to the Glut3 cDNA.

Confirming the in situ hybridization data, Glut3 and 4 proteins are concentrated in cortical neurons as shown immunohistochemically (Fig. 3). Glut4 immunoreactivity is concentrated in granular deposits in neuronal perikarya and processes and is distinctly more abundant in tissue from E2-treated animals. Glut3 immunoreactivity is also granular, with dense deposits in pyramidal perikarya but little detected in processes (Fig. 3C, D). Glut3 immunostaining is also markedly increased in E2-treated monkeys. Immunoblot analysis confirms significant increases in Glut3 and 4 protein levels in E2-treated animals (Fig. 4). IGF binding protein 5 (IGFBP5) levels were evaluated as an internal

Figure 1. Glut3 mRNA in vehicle- (A, B) and E2-treated (C, D) monkey frontal cerebral cortex. These are representative, paired bright and dark field micrographs with the cortical surface on the left of each panel. The Glut3 mRNA hybrid signal is concentrated in neurons, which have relatively large cell bodies (arrows), and is significantly increased in E2-treated animals (C). Quantitative analysis of the mRNA levels in cortical layers II and V from control, E2-, and tamoxifen-treated (TAM) animals is shown in graph (E). mRNA values represent grains per 10,000 μm². n=4–5 per group. Scale bar: 200 μ.

Figure 2. Glut4 mRNA levels in vehicle-treated control (A, B) and E2-treated (C, D) monkey cerebral cortex. A–D) Representative paired bright and dark field micrographs, with the cortical surface on the left of each panel. Insert dark field image in panel B shows background signal produced by sense probe hybridization. The Glut4 mRNA hybrid signal in control animals (A) is significantly above background, but not by much and is markedly increased in E2-treated animals (C), particularly concentrated in neurons (arrows). Quantitation of Glut4 mRNA levels in cortical layers II and V is shown graphically in panel E. Data for the Glut4 mRNA levels in tamoxifen-treated (Tam) animals are also shown. n=4–5 per group; mRNA values represent grains per 10,000 μ². Scale bar: 200 μ.
control to show the specificity of estrogen’s effects on cortical protein expression. IGFBP5 protein levels showed no change resulting from estrogen treatment. IGFBP5 mRNA levels were also equal in the different treatment groups (not shown).

Glut1 mRNA is highly abundant in vascular endothelium in the cerebral cortex (Fig. 5). There is, in addition, a diffuse Glut1 hybridization signal in the cortical parenchyma. This signal is associated with cells with small, dark-staining, oblong nuclei typical of glia. These cells are uniformly distributed throughout the cortex, including the molecular layer. This pattern is similar to that we have described for Glut1 expression in the rat brain (20, 27). In the latter study (27), colocalization of Glut1 mRNA and GFAP immunostaining showed that Glut1 is expressed by astrocytes. Glut1 immunoreactivity is abundant in cortical capillaries, but is not detected in brain parenchyma (Fig. 3E). Failure to detect Glut1 immunoreactivity in glial cells has been noted previously (30) and may be explained by altered antigenic presentation associated with glycosylation patterns of the glial transporter (see below). Glut1 mRNA levels were quantified by grain counting in areas of the cortex excluding capillaries, revealing a significant increase in parenchymal Glut1 gene expression in the E2-treated group. Grain density in capillary endothelial cells, however, was not altered in the different treatment groups (Fig. 5E). Immunoblot analysis of cortical Glut1 protein is complex because several isoforms are expressed in brain (Fig. 6). The higher molecular mass forms represent more heavily glycosylated transporters, which are concentrated in blood vessels, whereas the ~45 kDa form is expressed in glial cells (53). All Glut1 isoforms showed an increase in E2-treated animals, but the differences did not attain statistical significance (Fig. 6).

IGF1 and IGF1 receptor mRNAs are localized in cortical neurons in a pattern similar to Gluts 3 and 4.
E2 treatment was associated with a 60% increase in cortical IGF1 mRNA levels (vehicle, 65±11 grains/10,000 μ²; E2, 104±8; tamoxifen, 74±5, n=4–5 per group, P<0.01 for E2 vs. vehicle). IGF1 receptor mRNA levels were not altered by E2 treatment (not shown). Systemic E2 levels at the time brains were collected for these studies were as follows, in pg/ml: vehicle: 6.3±0.6; E2-treated = 400.2±121; tamoxifen = 7.8±1.4. These E2 levels are similar to mid-cycle values during the luteinizing hormone surge in cycling monkeys.

DISCUSSION

This study has shown that the facilitative glucose transporters Gluts 3 and 4 are both widely expressed in primate cortical neurons and that the expression of both these transporters is augmented by estrogen at the mRNA and protein level. We have shown that IGF1 mRNA is also expressed in adult primate cortical neurons and is significantly augmented by estradiol. Although direct coexpression studies were not attempted, the fact that IGF1 and Gluts 3 and 4 are localized in the same cell populations assessed in serial sections makes it likely that these three factors are coexpressed in many cortical neurons. Tamoxifen, an estrogen receptor antagonist used to treat breast cancer (54), is shown to have estrogen-like effects in the brain in terms of enhancing expression of Gluts 3 and 4 and IGF1. The findings provide novel insight aiding in our understanding of estrogen’s role in neuroprotection.

The current investigation of estrogen’s effects on brain IGF1 and glucose transporter expression was stimulated by prior observations that estrogen increases IGF1 expression (55, 56) and glucose utilization in the uterus (57, 58). These effects are also associated with augmentation of uterine Glut1 expression (59). An estrogen effect has been associated with increased

Figure 5. Glut1 mRNA levels in vehicle-treated control (A, B) and E2-treated (C, D) monkey frontal cerebral cortex. The hybridization signal, seen as white grains in the dark-field view, appears in brain parenchyma and blood vessels (BV, arrows). E) Graph shows the quantitation of mRNA levels in blood vessels and parenchyma. Parenchymal signal was assayed in 2000 μ² regions of frontal cortex, excluding capillaries; blood vessel signal was obtained overlying capillary endothelial cells. Glut1 mRNA is significantly increased in parenchyma but not in the blood vessels of E2 animals. No difference is seen in Tam-treated animals compared to controls; n = 4–5 for each group. Scale bar: 200 μ.

Figure 6. Immunoblot analysis of Glut1 protein levels in vehicle-treated control and E2-treated rhesus monkey frontal cortex. A) Immunoblot was probed with antibodies against Glut1, visualized by chemiluminescence, and exposed briefly to reveal the abundant, higher molecular mass form of Glut1 (55 to 60 kDa) in blood vessels (upper panel). The same blot was exposed longer to reveal the 45 kDa form of Glut1, which is characteristic of glial membranes (lower panel). B) Quantitation of Glut1 protein levels in frontal cortex. Protein bands from blots shown in panel A were quantified using computer-assisted image analysis. Data are expressed as mean ± se of percentage of control values. Although all Glut1 isoforms appear increased with E2 treatment, the effects were not statistically significant due to intergroup variability.
glucose utilization in the rat brain (18, 19). Recent clinical studies have shown that cerebral glucose utilization is increased in estrogen-treated menopausal women (16, 17), in the high-estrogen phase of the menstrual cycle (60), and in menstruating women compared with men (61). Together, these observations suggest the possibility that estrogen’s trophic effects on brain involve local IGF1 and glucose transporter expression.

A previous study has reported Glut3 mRNA expression in monkey cortical neurons (32). The present study confirms this finding and provides immunohistochemical and immunoblot verification that Glut3 is expressed by primate cortical neurons. To our knowledge, no previous study has addressed the role of sex hormones in regulating Glut3 expression. The present data show clearly a strong augmentation of Glut3 gene expression and a significant increase in Glut3 protein levels related to 3 days of *in vivo* estrogen exposure. The present study demonstrates for the first time that the insulin-sensitive transporter Glut4 is expressed in the primate brain. This observation extends recent reports on localization of this transporter in the murine brain (33–35). As noted, our data suggest that Gluts 3 and 4 are coexpressed by cortical neurons, a finding that has been documented in the rat brain (36). The immunohistochemical observations in this study suggest that both transporters are concentrated in cytoplasmic vesicles, with Glut4 but not Glut3 also detected in dense deposits along neuronal processes. These interesting observations require further examination, including the use of a variety of antibodies and fixation methods, before we can make firm conclusions about possible differential intracellular distributions of these transporters. The significance or utility of the expression of two different facilitative transporters by cortical neurons is not known. It seems likely that these two transporters will be found to be differentially responsive to specific regulatory stimuli or differentially associated with intracellular signaling pathways or metabolic compartments.

We also demonstrate, for the first time we know of, cellular patterns and hormonal regulation of IGF1 expression in the primate brain. IGF1 is highly expressed in the developing murine brain, where it is concentrated in maturing projection neurons (47, 62). It is expressed transiently by cortical pyramidal neurons but is reduced and nearly undetectable in the adult rat and mouse cerebral cortex. The present study shows that IGF1 mRNA is readily detected in adult monkey cortical neurons, suggesting a role for IGF1 in mature brain function/homeostasis in the primate. The observation that brain IGF1 gene expression is increased significantly by estrogen is consistent with estrogen-
induced IGF1 expression in peripheral tissues such as uterus (55, 56) and mammary gland (63). We have recently shown that IGF1 is an essential mediator of estrogen’s action on the uterus (64). Estrogen’s many anabolic effects on neural tissue (4) are identical to those reported for IGF1 [e.g., promoting neurite growth, synaptogenesis, and neuron survival, enhancing cholinergic activity and neuroprotection against a variety of insults (reviewed in ref 65)], and estradiol’s neuroprotective effects are reportedly blocked by an IGF1 receptor antagonist (66). These observations, together with the present data, suggest that many of estrogen’s salutary effects on the brain may be mediated by IGF1. In the present case, for example, the parallel increases in IGF1 and Glut expression in the E2-treated cerebral cortex could be due to an autocrine or paracrine effect by IGF1 to enhance Glut3 and 4 expression. Although parallel changes in IGF1 and Glut expression do not establish cause and effect, the fact that the IGF1 receptor is expressed by these same neurons suggests it as a possibility. Further studies in the IGF1 null brain, for example, should help clarify these issues.

We found Glut1 mRNA and immunoreactivity to be abundant in monkey brain vasculature. Glut1 mRNA was also localized in parenchymal cells uniformly distributed through the gray matter. These are likely to be astrocytes, based on their morphology and distribution. We have demonstrated colocalization of the parenchymal Glut1 mRNA with an astrocyte marker in the rat (27), and others have recently produced convincing demonstrations of glial cell expression of Glut1 immunoreactivity (24, 25), so it seems likely that the Glut1 parenchymal signal in primate brain also originates in glial cells. We found that parenchymal Glut1 mRNA levels were significantly increased by estrogen treatment, but capillary Glut1 mRNA levels were not. One earlier study reported estrogen-induced increases in Glut1 expression in the BBB of the rat (29); however, the investigators examined time points within 24 h after a single E2 injection whereas we examined brains after 3 days of sustained treatment. This earlier study of the rat did not investigate parenchymal Glut1 levels or other glucose transporters. The Glut1 immunoblot data from the present work suggests an increase of all Glut1 isoforms in brains from E2-treated animals, although statistical significance was not attained for these changes. It appears that to obtain a clearer picture of Glut1 regulation by estrogen in the primate brain, more time points and more animals per group may be necessary.

Originally developed as an antiestrogen (67), tamoxifen is now included in the class of drugs known as selective estrogen receptor modulators (SERMs). These agents have effects that vary from anti- to proestrogenic depending on the target cell type and the ambient steroid environment (68). For example, tamoxifen functions mainly as an antiestrogen for mammary tissue in women, whereas it exerts proestrogenic effects on bone and lipids (67). Tamoxifen treatment induces or exacerbates hot flashes typical of estrogen withdrawal, suggesting it has antiestrogen effects at brain vasoregulatory centers. The present data show, however, that tamoxifen treatment of ovariectomized monkeys augments Glut3 and 4 expression in the cerebral cortex, which suggests that in higher brain centers, tamoxifen has an estrogen-like action. If these changes in the cerebral cortex are involved in estrogen’s salutary effects on brain, it appears that tamoxifen and other SERMs may have at least a partial estrogenic benefit for brain. This observation is an important one: because of the concern about breast cancer risk, many women are now being treated with SERMs rather than estrogen (67).

In summary, this study has shown that primate cortical neurons express two different glucose transporters, Gluts 3 and 4 and IGF1, and that the expression of all three factors is significantly increased by estrogen. It is worth noting that brain Glut1 and 3 levels (Glut4 was not evaluated) are significantly reduced in subjects with Alzheimer’s disease (69), suggesting one way in which estrogen may address the Alzheimer’s neuropathology. We hypothesize that these effects may be part of the fundamental mechanism underlying estrogen’s manifold anabolic and neuroprotective functions on neural cells, with IGF1 mediating some of estrogen’s effects in brain just as it does in other tissues.

REFERENCES

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translocation of GLUT4 in transfected rat adipose cells. J. Biol. Chem. 272, 8026–8031

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