The Human Amyloid Precursor Protein Adapter Protein X11α Enhances Amyloidosis in a Mouse Model of Alzheimer's Disease

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Abstract

X11α is a protein containing a phosphotyrosine-binding (PTB) domain that binds to the GYENPTY motif of the cytoplasmic terminal of Amyloid Precursor Protein (APP). Normally, APP secretes amyloid-beta, the primary cause of brain cell death in Alzheimer's disease, through cleavage by the beta- and gamma-secretases. Literature indicates that X11α modulates this processing of APP and secretion of amyloid-beta, Aβ, making it a potential therapeutic target for Alzheimer's research. To investigate the role of X11α in APP processing, the levels of amyloidosis were analyzed using immunohistochemistry. Mice overexpressing both APP and X11α and mice overexpressing only X11α were compared to their respective controls. Mice overexpressing both APP and X11α were found to have significantly more Aβ deposition, both by area and number, than mice overexpressing only APP. Further GFAP immunocytochemistry indicated a significantly increased glioses immune response in mice overexpressing just X11α than in wildtype mice. Observation of doublelabeling of mice overexpressing both APP and X11α under high magnification revealed colocalization, indicating that X11α is indeed acting upon APP. This research suggests that X11α has a negative role in APP processing, contradicting the effect hypothesized by many previous findings. Instead of treating X11α as a potential therapeutic target, future research should instead investigate methods which enable for the hindering of APP- X11α interactions.

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1. Introduction

Alzheimer's disease (AD) is the leading cause of dementia in the elderly, affecting some 25 million people worldwide, a figure expected to rise to 81 million by 2040 (Rogelj et al., 2006; Blennow, et al., 2006). Statistics published by the Aging, Demographics and Memory Study and like studies estimate that less than 1% of the population under the age of 65 have AD, 13% of persons over the age of 65 has AD, and anywhere between 24% to 50% of the population of persons over 85 years of age have AD (Blennow et al., 2006; Alzheimer's Association, 2009). Gender also appears to be a contributing factor, as males aged 71 and older have a lower prevalence of AD than women aged 71 and older: 11% and 16%, respectively (Alzheimer's Association, 2009).

The hallmark pathologies of Alzheimer's disease are extracellular plaques, formed by congregated amyloid-β (Aβ) peptides, neurofibrillary tangles, composed of insoluble helical filaments of tau protein (Sano et al., 2006), and dystrophic neurites. Aβ is a 40- or 42- amino acid peptide derived by proteolytic processing of the much larger Amyloid Precursor Protein (APP), a large membrane-spanning protein with size variations due to alternative splicing (King et al., 2004). The secretion of Aβ from APP is a result of cleavage by the β-secretase at the C-terminus of the peptide and by the γ-secretase at the N-terminus (King et al., 2004).

Several medications are available only to alleviate or delay the progression and exacerbation of symptoms of AD (ADEAR, 2009), but currently, no silver bullet exists to either cure or prevent AD. As a result, research is actively pursuing possible therapeutic substances which are either neuroprotective or have the potential to either remove amyloid plaques after deposition or to prevent the aggregation and deposition of Aβ. Much research is dedicated to the study of

proteins that bind to APP and affect its processing, the X11 adaptor protein family being one such protein.

The X11 adaptor protein family has three members: X11α, also known MINT1 and mLIN10; X11β, also known as MINT2 and X11L (X11-Like); and X11γ, also known as MINT3 and X11L2 (Rogelj et al., 2006). For the purpose of this research, these proteins are referred to as X11α, X11β, and X11γ. All three proteins have one central phosphotyrosine binding domain (PTB) and two PSD-95/discs large/ZO-1 (PDZ) domains. X11α and X11β have a Munc interacting region (MI), whereas X11γ lacks this region; it is MI that gives rise to the alternate name of the X11 family, MINT. Only X11α has the CASK interacting region (CI) (Rogelj et al., 2006) (Fig. 1).

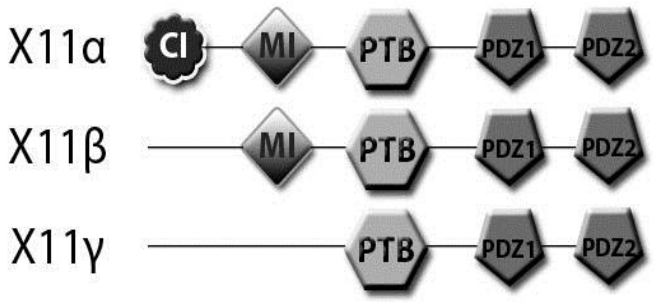

Figure 1. Structure of X11 Family Proteins
A representation of domain structure in X11 family proteins. CI: CASK-interacting region; MI: Munc-interacting region; PB: phosphotyrosine binding doman; PDZ: PSD-95/discs large/ZP-1 domain. (Rogelj, 2006)

The X11 family has been shown to react with APP, by way of their PTB domains, affecting the processing of APP and thus influencing the build-up of Aβ; additionally, studies show that X11*γ* binds to APP by way of its second PDZ domain (Tanahashi et al., 1999). The X11 family's reactivity with APP allows X11s to become a potential target for therapeutic research in Alzheimer's disease. *In vitro* studies have shown that overexpression of X11 results in retention of cellular APP and decreased Aβ secretion (Borg et al., 1998; Sastre et al., 1998). Differentiated

effects were reported for X11α and X11β; X11α can inhibit the generation of both $A\beta_{40}$ and $A\beta_{42}$, whereas X11β can only inhibit $A\beta_{40}$ (Rogelj et al., 2006). Whatever the differences, most *in vivo* studies of the interaction between the X11 and APP reach the consensus that the expression of X11 inhibits the deposition of Aβ plaques (Sano et al., 2006; Lee et al., 2003; Saluja et al., 2009). However, there are two recent studies which contradicts this, claiming that the deletion of X11 proteins decreases amyloid production (Ho et al., 2008; Xie et al, 2005). The exact mechanisms for how these X11 proteins affect APP has not yet come to light (Rogelj, 2006; Miller, 2006)

2. Research Question & Hypothesis

This research addresses two objectives. First, it attempts to identify the intracellular localization of X11α in X11α-overexpressing mice. The second purpose of the research is to determine what is the effect of overexpression of X11α on amyloid deposition in a mouse model with cerebral Aβ amyloidosis. The mice brain sections used in the experiments come from the Mendelian progeny of two strains: the transgenic mice expressing human X11α under control sequences of the neuron-specific Thy-1.2 generated in our laboratory and APP overexpressing mice, APP23. This researcher decided to analyze the results of experiments, described in the third section, entitled "Materials and Methodology", to determine the difference in plaque formation between APP23 mice and mice double transgenic for APP and human X11α. From a statistical point of view, the hypothesis that was tested using the presented data was:

Null Hypothesis: There is no difference in levels of amyloid deposition between APP23 mice without X11α overexpression and APP23 mice overexpressing X11α.

Alternate Hypotheses: There is either a decrease or an increase in the levels of amyloid deposition between APP23 mice without X11α overexpression and APP23 mice overexpressing X11α.

Steps dealing with live animals and the procurement of brain sections were completed by a colleague in the laboratory. All subsequent experiments involving immunocytochemistry, histology, and analyses were completed by this researcher.

3. Materials & Methods

Transgenic mice

Transgenic mice expressing human X11α were generated in our laboratory. APP23 mice, overexpressing the Swedish mutation of APP (APPsw), a line of mice utilized in many studies (Sturchler-Pierrat, et al. 1997) were generously provided by Novartis Pharma Inc. Both lines of mice were generated using the same neuron specific Thy-1.2 promoter, resulting in overexpression of both genes in the same neuronal population. Positive lines were identified by PCR analysis of genomic DNA prepared from the tails and crossed further to obtain APP23/X11α mice. APP23/X11α double transgenic mice were compared to singly APP23 or X11α transgenic mice and wild type, non-transgenic controls. Colonies for aging mice were established and mice were sacrificed at 12 months of age. All mice from which brain sections were derived were born between November 2003 and October 2007 and were sacrificed between

December 2005 and October 2008 for the purposes of other X11α research. Brains were divided into two, with one brain hemisphere formalin fixed for histological analyses and the other hemisphere was flash-frozen for biochemical analyses. All interactions and handling of the mice were conducted by colleagues at the laboratory. These mice will be divided into four groups: APP23, X11α, APP23/ X11α, and wild type mice for the purposes of this research (Table 1). Blocks of 25 formalin-fixed mouse brains were prepared by Neuro Science Associates and coronal serial sections cut at 35 μm.

Table 1. Mouse Data

Mouse ID	Gender	Genotype	Date of Birth	Age at Death
FD13	F	APP23	2/9/2005	12.16
FD14	F	WILD TYPE	2/9/2005	12.16
FD15	F	WILD TYPE	2/9/2005	12.16
MD11	M	APP23	2/9/2005	12.16
MD12	M	APP23	2/9/2005	12.16
MD13	M	WILD TYPE	2/9/2005	12.16
MD14	M	WILD TYPE	2/9/2005	12.16
MD3	M	APP23/X11α	12/22/2004	12
MD4	M	APP23/X11α	12/22/2004	12
FD45	F	WILD TYPE	10/15/2007	12.1
MD41	M	WILD TYPE	7/2/2007	12.3
FD39	F	WILD TYPE	7/2/2007	12.1
MD38	M	APP23	8/1/2007	11.97
MD48	M	X11α	10/15/2007	12.1
MD43	M	X11α	7/2/2007	12.3
FD38	F	X11α	7/2/2007	12.1
MD45	M	X11α	8/1/2007	12.03
MD49	M	X11α	10/15/2007	12.1
MD40	M	APP23	7/2/2007	12.3
MD42	M	APP23	7/2/2007	12.3
FD34	F	APP23	8/8/2005	12.1
FD44	F	APP23/X11α	10/15/2007	12.1
FD41	F	APP23	7/2/2007	12.3
MD46	M	APP23	8/1/2007	12.03
FD42	F	APP23/X11α	8/1/2007	12.03
MD44	M	X11α	8/1/2007	12.03
MD29	M	WILD TYPE	6/26/2005	12
FD40	F	X11α	9/14/2005	11.97
MD39	M	X11α	9/14/2005	12.03
MD37	M	X11α	9/14/2005	11.97
MD47	M	APP23	8/1/2007	12.03

Presented here are the mice used and referenced by this research; certain mice were not used in statistical analyses, but all were subjected to immunocytochemical experimentation and microscopy.

HISTOLOGY

Thioflavine S staining

Coronal serial sections cut at 35 μm were mounted on gelatin coated micro slides 75x50 mm and dried before staining. Sections were stained with filtered 1% thioflavine S in ddH_2O for 10 min followed by incubations with 70%, 80%, 95% and 100% ethanol and xylene, each for 5 min. Slides were covered with cover slips using Permount for observation and storage. Staining was viewed through Carl Zeiss Axioscope 200M research microscope. Images were captured with the Mosaic function, a composite figure of individual 10x microscope images. Three to six images of the hemibrain sections were collected per mouse. Aβ pixels were determined using ImageJ imaging software in which we apply the same threshold value (75 to 240) and then the total number of individual plaques and each plaque's individual areas within the cortex and hippocampus were calculated for each section image. Percent plaque area was determined by (Plaque area measured / Total area selected) x 100%.

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A list of antibodies used in this research can be found in Table 2.

Immunofluorescence Staining

For immunohistochemistry, coronal floating sections were incubated in 0.1 M Tris pH 7.4, 150 mM NaCl, 0.25% Triton X-100 for 15 min at room temperature, and sections were treated with 100% formic acid for 5 min to enhance amyloid staining. The endogenous peroxidase activity was quenched with 0.3% H_2O_2 in ddH_2O for 30 min, and non-specific binding was blocked with 0.05 M Tris-HCl pH 7.6, 0.9% NaCl, 0.25% Triton X-100, buffer containing 20% goat serum and 0.2% bovine serum albumin for 30 min. The sections were then

incubated overnight at 4°C with the primary antibody. See Table 2 for a complete list of antibodies used in this research. The immunoreactions were revealed by secondary antibody after 30 min. Sections were mounted on gelatin coated slides and using Fluoromount and coverslipped for observation and storage. Negative controls included substitution of the primary antibody with mouse serum.

Immunocytochemical staining of paraffin sections

Xylene and ethanol series were applied to deparaffinized and hydrated sections, followed by microwave treatment of the sections in 10 mM citrate buffer and cooling down for at least 30 minutes. Sections were rinsed with TBS, treated with blocking buffer (TBS/2%DHSorFBS/0.1%TritonX-100) for 1 hour, incubated in primary antibody (antibody diluents: TBS/1%DHS or FBS/0.01%TritonX-100) in humidified chamber either overnight at 4°C or for 1-3 hours at room temperature. Sections were rinsed, incubated with secondary antibody in humidified chamber for 1 hour, and then incubated with the tertiary ABC solution for another hour. DAB solution was applied and sections were prepared for observation and storage with permount.

Aβ plaque quantification

Images were obtained by using Carlzeiss Axioscopes model (city, state) (Figure6 & 7). Two forms of analysis were used: the number of distinct plaques and the percent area of plaques in the selected area of experimentation, in the hippocampus and cortex of the midbrain. Both forms used the Image J software

Table 2. Utilized Antibodies

All primary antibodies are presented here alongside their respective secondary antibodies, the targeted objects, any relevant notes for immunocytochemistry, and company and location of origin. Thioflavine S is a stain, not an antibody, and has been excluded from this table as such.

Primary antibody & dilution	**Stained target**	**Secondary antibody &dilution**	**Additional Notes**	**Manufacturer; City**
X11α 1:150	X11α proteins	Alexafluor 488, anti-Rabbit 1:750		Santa Cruz Biotech Inc.; Santa Cruz, CA
4G8 1:200	Aβ	Alexafluor 594 anti-Rabbit 1:750	Pretreat with 70% formic acid in ddH_2O	Signet; Deadham, MA
GFAP 1:200	Gliosis-protein glial fibrillary acidic protein	Alexafluor 594 anti-Mouse 1:750		Dako Canada Inc.; Mississauga, Canada

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4. Results

4.1 X11α expression

X11α overexpressing transgenic mice were created using the gene promoter Thy1.2 to drive expression solely in neuronal cells. Expression was detected using anti-X11α antibody, and. X11α was found to be expressed throughout the brain, but primarily in the sensory motor cortex, and the CA3 region of the hippocampus in both wild type and transgenic X11α mice. High magnification microscopy showed X11α expression to be predominantly cytoplasmic in the cell body and axons, with negligible synaptic localization. Punctate intracellular staining in neuronal cells was also observed (Fig. 3).

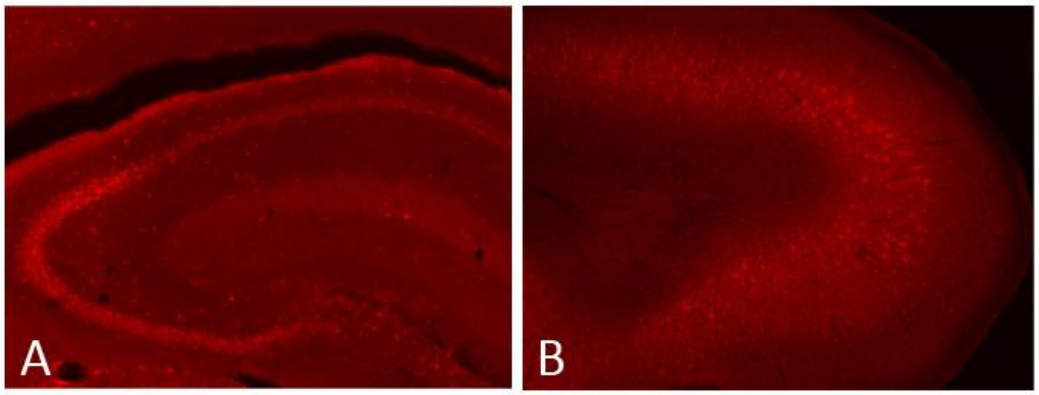

Figure 2. Elevated expression of X11α in the hippocampus and cortex
The CA3 region of hippocampus shows heightened X11α expression, as does the cortex, with a greater staining in the sensory motor cortex

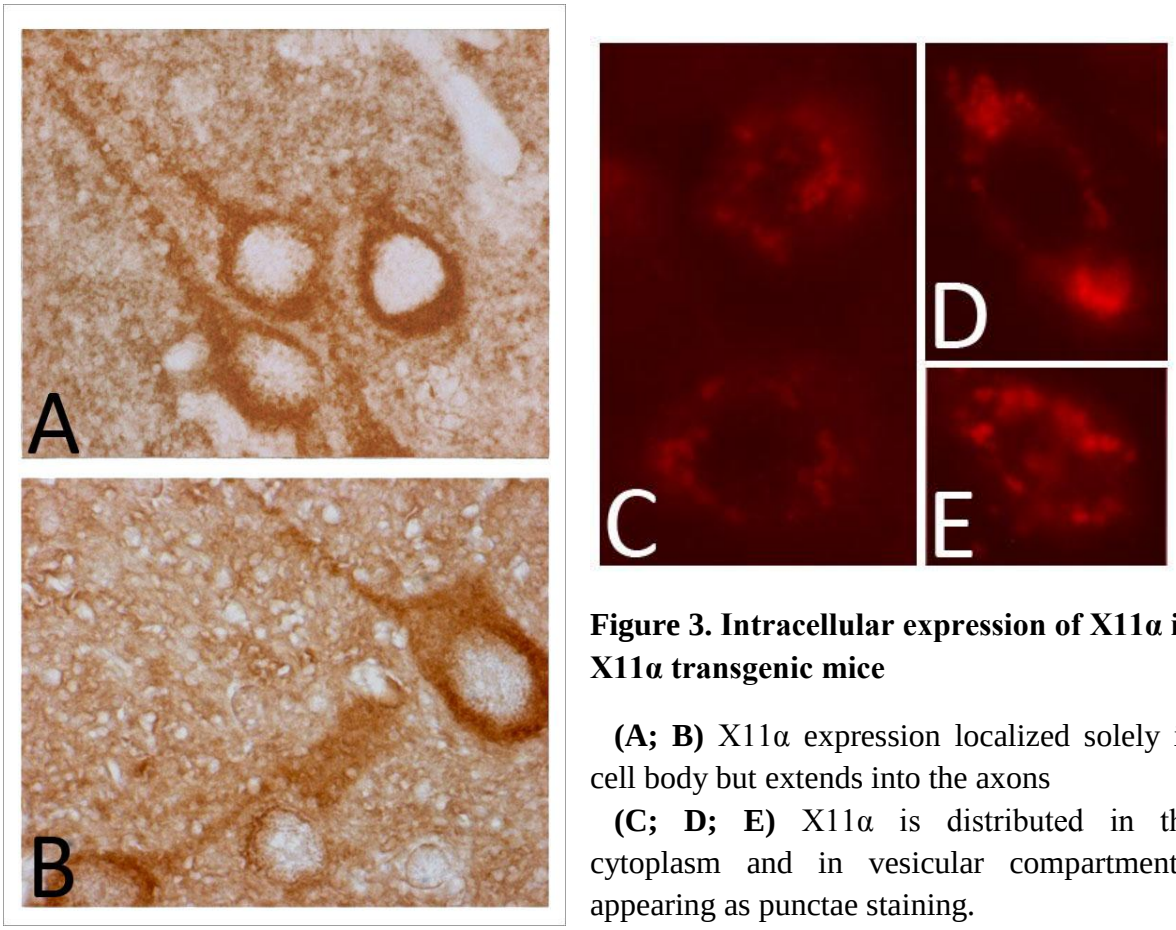

Figure 3. Intracellular expression of X11α in X11α transgenic mice

(A; B) X11α expression localized solely in cell body but extends into the axons

(C; D; E) X11α is distributed in the cytoplasm and in vesicular compartments, appearing as punctae staining.

Photographs were taken at 100x magnification from 12-months old X11α mice immunolabeled with anti-X11α antibody.

4.2 APP/Aβ expression with *X11α*

Coronal mid-sections of mice brains were analyzed using the described histological procedures. Aβ fragments, like X11α, are expressed cytoplasmically, but with vaguely more uniform distribution in the cell body and less evident expression in the axons (Fig. 6). Analysis of APP23/X11α mice brain sections double-labeled with 4G8 and anti-X11α antibody revealed colocalization of X11α and Aβ under 100x magnification (Fig. 4).

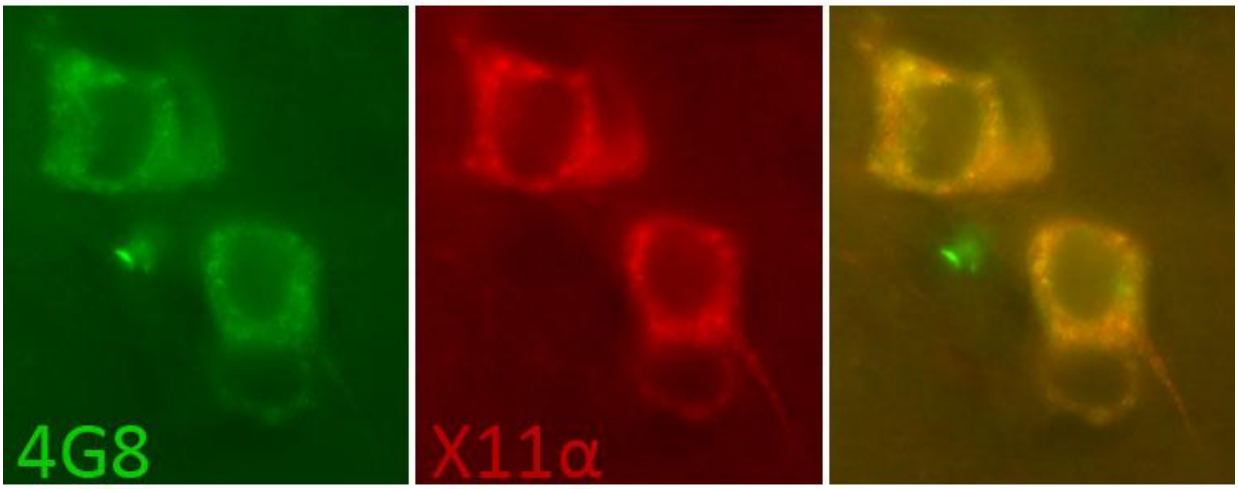

Figure 4. Colocalization of APP and X11α

An APP23/X11α mouse dually stained with 4G8 and anti-X11α antibodies shows the colocalization of APP23 and X11α in the cytoplasm and vesicular compartments within neuronal cells. Pictures were taken at 100x magnification in a 12 month APP23/X11α mouse; overlay of 4G8 (green) and X11α (red) show colocalization of the two proteins (yellow).

Brain sections were stained with thioflavine S to detect amyloid deposits (Fig. 5). Comparison of Aβ deposition levels in APP23 and APP23/X11α transgenic mice led to conclusions contradictory to those largely described in the literature. APP23 mice showed significantly less Aβ deposition than APP23/X11α double transgenic mice. The mean number of plaques for APP23 (n = 8) and APP23/X11α mice (n = 4) was 22.2 and 44.6, respectively, and standard

deviations of 7.5 and 18.1, respectively; $p < 0.011$ (Fig. 6). The mean percent plaque area for APP23 and APP23/X11α mice was 0.0418% and 0.167%, respectively, and standard deviations of 0.023 and 0.056, respectively, with $p < 0.00022$ (Fig. 7). 4G8 staining confirmed the aforementioned results (Fig. 8), with p-values for plaque number and percent plaque area being less than 0.033 and 0.017, respectively.

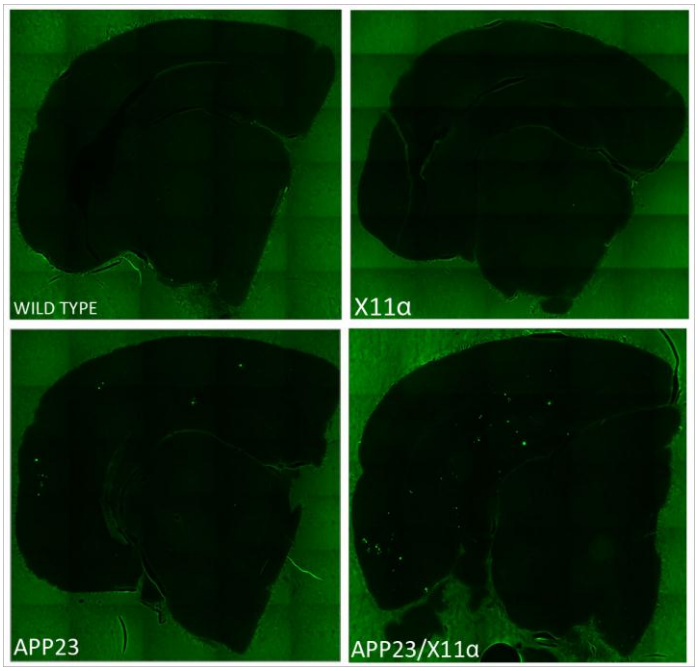

Figure 5. Thioflavine S staining of wild type, X11α, APP23, and APP23/X11α mouse sections
Sections stained with 1% thioflavine S showed no plaque formation, as expected, in the brains of wild type and X11α mice, some plaque formation in the brains of APP23 mice and the greatest in APP23/X11α mice.

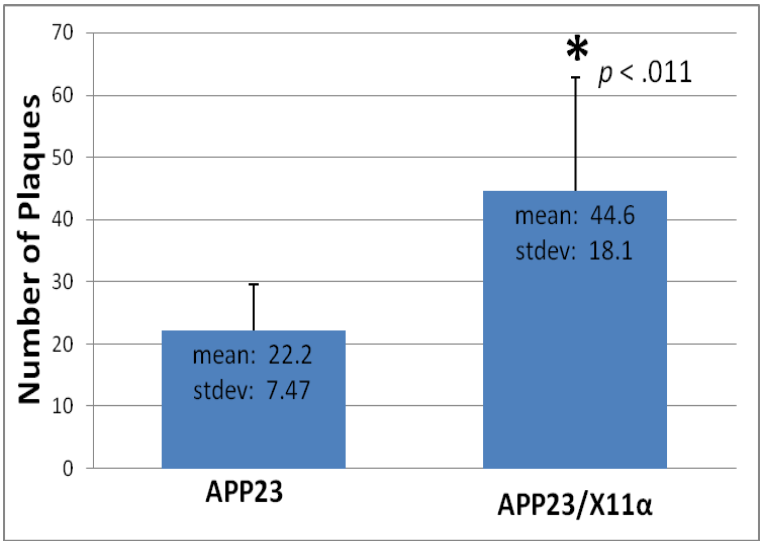

Figure 6. Number of Aβ Plaque in Hippocampal and Cortex Regions

Entire APP23 and APP23/X11α sections stained with 1% thioflavine S were captured by Mosaic photographs at 10x magnification, from which analyses revealed significantly higher number of plaques per region measured in brains of APP23/X11α mice than in brains of APP23 mice.

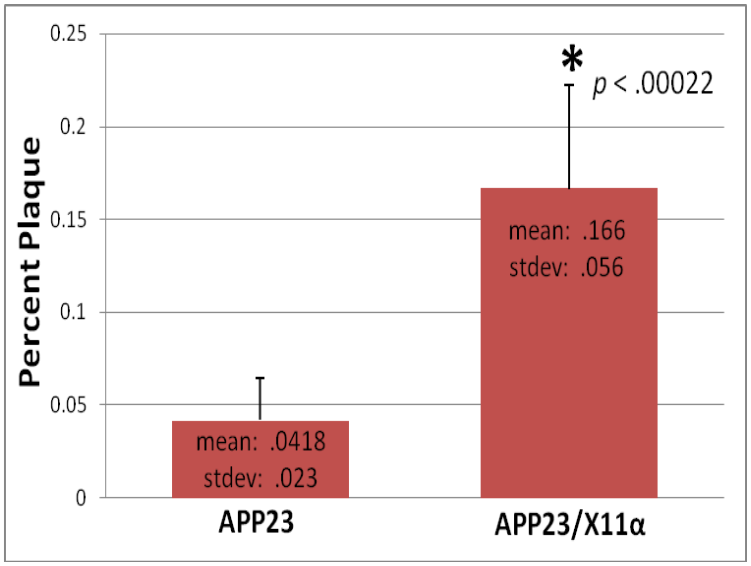

Figure 7. Percent Area of Hippocampal and Cortex Regions occupied by Aβ Plaque

Entire APP23 and APP23/X11α sections stained with 1% thioflavine S were captured by Mosaic photographs at 10x magnification, from which analyses revealed significantly greater percentage of plaque area per area measured in the brains of APP23/X11α mice as compared to APP23 mice

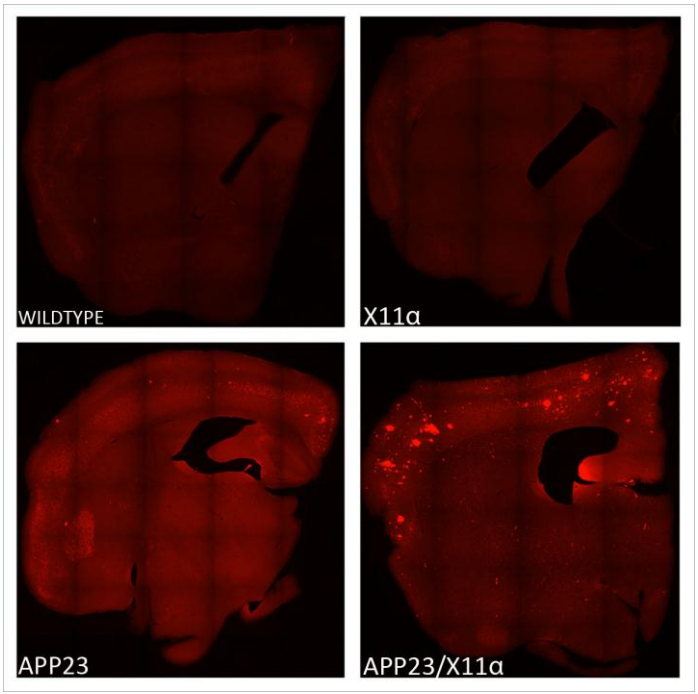

Figure 8. 4G8 staining of wild type, X11α, APP23, and APP23/X11α mouse sections

Sections stained with 4G8 showed no plaques , as expected, in wild type and X11α mice, some plaque deposits in the brains of APP23 mice and the greatest in APP23/X11α mice.

4.3 Gliosis

Histological staining was done on brain sections from the same group of mice described in section 4.2. These sections were subjected to GFAP staining (glial fibrillary acidic protein), which tests the glioses reaction of glial cells in the brain (Fig. 9).

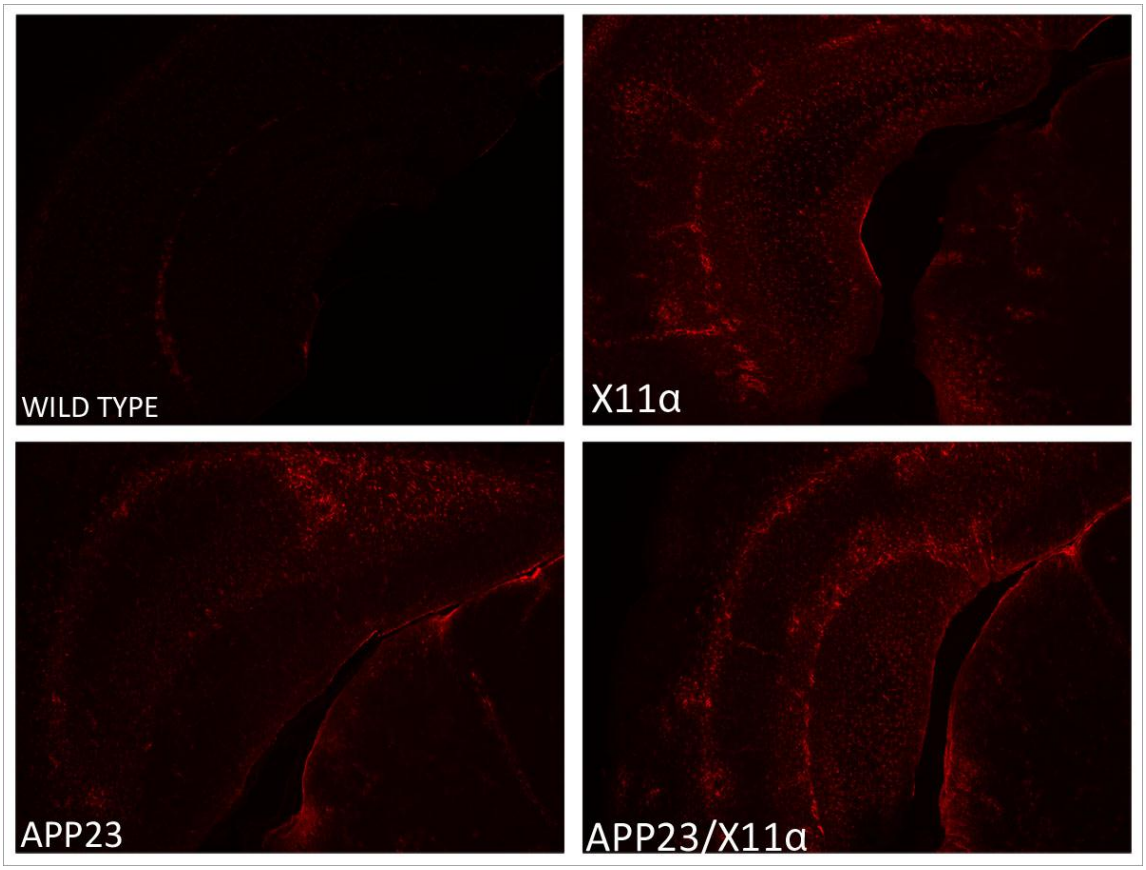

Figure 9. GFAP staining of wild type, X11α, APP23, and APP23/X11α mouse hippocampi.
Sections stained with an antibody to glial fibrillary acidic protein (GFAP) reveal significantly more glioses reactions in APP23/ X11α mice than in APP23 mice and significantly more glioses in X11α mice than in wild type. Photographs taken at 5x magnification, focusing on the hippocampal region, which revealed the greater GFAP staining than other regions, which consistently revealed negligible GFAP staining/gliosis.

ImageJ measured the intensity and density of each section, and analyses revealed that there was significantly more glioses in APP23/X11α mice than in APP23 mice, $p < 0.010$, which is to be expected as there are more plaques and a greater area of plaques in APP23/X11α mice, as reported in section 4.2 (Fig. 10). GFAP staining in wild type and X11α mice revealed significantly more glioses in X11α mice, with $p < 0.039$, thus indicating that overexpression of X11α is exacerbating the stress response in the brain (Fig. 10).

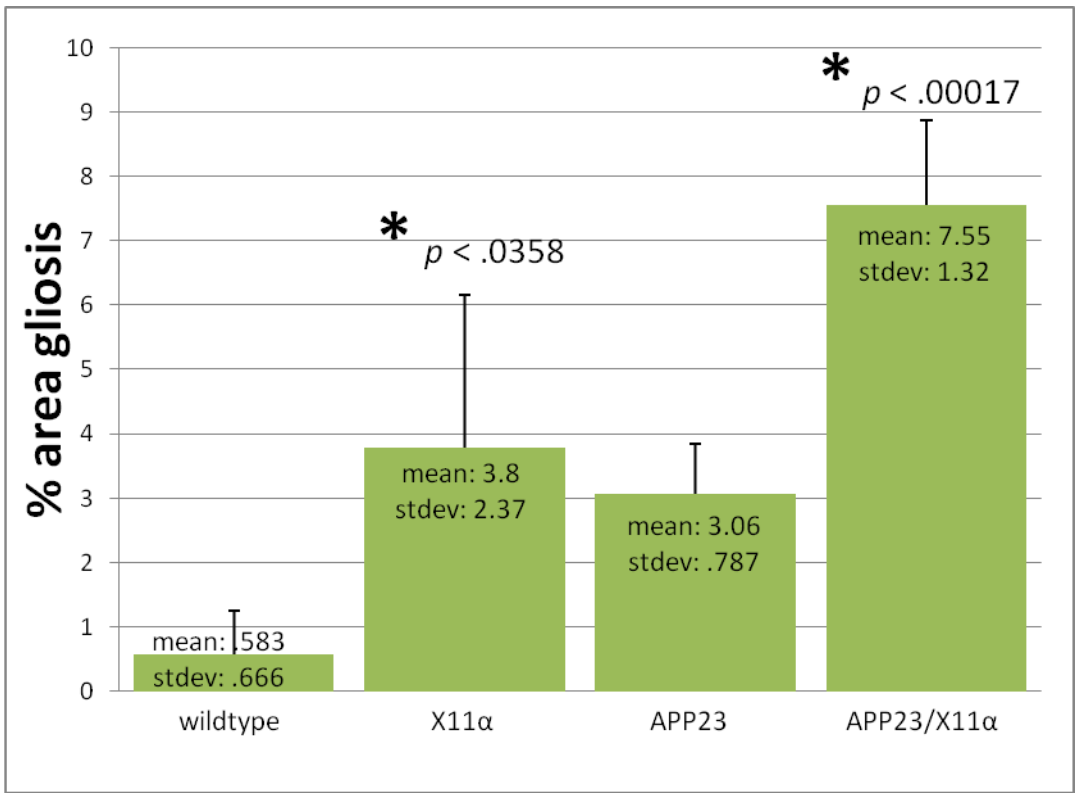

Figure 10. Variation of GFAP staining across wild type, X11α, APP23, and APP23/X11α mice

The difference in the mean percent area of hippocampus between wild type and X11α mice is significant, as is the difference between APP23 and APP23/X11α mice.

5. Discussion

The X11α *in vivo* mouse model of Alzheimer's disease is imperative as it provides insight into the potential effect of X11α on amyloid deposition in APP overexpressing, Aβ depositing APP23 transgenic mice. The ability of the X11 family members to bind to the Glycine-Tyrosine-Glutamic Acid-Asparagine-Proline-Threonine-Tyrosine (GYENPTY) motif of the intracellular cytoplasmic terminal of APP through the PTB domain yields their potential as possible therapeutic targets in Alzheimer's disease (Borg et al, 1996). Immunocytochemistry

conducted on 12 months old X11α and wild type mouse brain sections under high magnification showed that expression of X11α is cytoplasmic and axonal (Fig. 3), consistent with findings that indicate that X11α localizes near and within the cell membrane (Borg et al, 1998). The punctate staining that was observed within the cytoplasm of neuronal cells suggests containment of the protein in vesicular compartments. Furthermore, observation under low magnification showed that X11α in wild type and X11α overexpressing transgenic mice is expressed throughout the brain, but is with high levels of expression in the CA3 region of the hippocampus (Fig. 2), in addition to strong X11α labeling in the sensory motor cortex (Fig. 2). These observations are consistent with previous findings suggesting that X11α is highly expressed in hippocampal inhibitory interneurons in the CA3 region and in the neocortex (Ho et al, 2008; Nakajima et al, 2001).

To ascertain the modulation of APP accumulation and Aβ deposition by X11α, this research explored the results of X11α overexpression in APP23 transgenic mice at 12 months. Double-labeling of sections with anti-X11α and 4G8 antibodies revealed that X11α and APP/Aβ colocalized within cells (Fig. 4). Colocalization of the two proteins was expected given that the expression of the two genes was controlled by the Thy-1.2 promoter. Expression in the same cells accommodated interactions between X11α and APP and its fragments.

While wild type mice have shown no amyloid deposition, APP23 transgenic mice showed significantly amyloidosis at 12 months, consistent with previous studies (King et al, 2004; Simón et al, 2009). The means by which X11α modulates the metabolism of APP remain unclear (Rogelj et al. 2006; Miller et al, 2006), but theories are that: first, X11α alters the cleavage of APP by the γ-secretase protein complex through binding to the presenilin-1 domain

via both PDZ domains (Miller et al, 2006); and second, X11α binds to the GYENPTY motif within the cytoplasmic domain of APP through PTB domain, inhibiting or exacerbating amyloidosis; these two mechanisms, may not be mutually exclusive (Miller et al, 2006). Previous findings show conflicting support for two different effects of X11α on modulation of levels of amyloidosis: there is an inverse correlation between X11α expression and plaque deposition (Saluja et al, 2009; Sastre et al, 1998; Lee et al, 2003) or there is a direct relationship between X11α and plaque deposition (Ho et al, 2008; Xie et al, 2005). This incongruity can be dependent upon the promoters used, the model on which the study was based, strain of APP gene utilized, the use of X11α transgenic mice as compared to X11α knockout mice, alongside any number of unknown variables. The discrepancy in results makes the function of X11α in modulating levels of amyloidosis and in the pathogenesis of Alzheimer's disease ambiguous.

In the presented research, coronal brain sections were stained with thioflavine S, which stains the Aβ plaques; images were taken and analyzed using ImageJ software (Fig. 5). No amyloid deposition was observed in the brains of X11α mice. For both the number of plaques and the percent area of cortical and hippocampal regions that is occupied by plaques, APP23/X11α had significantly more plaque deposition than APP23 mice. Immunocytochemistry and analyses by 4G8 staining confirmed the aforementioned results (Fig. 8). The Null Hypothesis for this research has hence been rejected, and the Alternate Hypothesis, which states that there is a change in the levels of amyloid deposition between APP23 mice without X11α overexpression and APP23 mice overexpressing X11α.

This study concludes that there is an increase in the levels of plaque deposition in APP23 transgenic mice with X11α overexpression, a conclusion consistent with the studies conducted

by Ho et al., 2008, which found that the deletion of X11 family members decreased and delayed amyloid production in transgenic mouse models of Alzheimer's disease, and Xie et al, 2005, which found that silencing of X11α and X11β attenuated Aβ levels. The study conducted by Ho et al. shows that up to 9 months of age, mice expressing X11β and X11γ but with X11α deletion (to be referred to as $X11\alpha^{-/-}/X11\beta^{+/+}/X11\gamma^{+/+}$ henceforth) had more gradual and less plaque accumulation of Aβ compared to mice which expressed all three members of the X11 family. At 12 months, the study shows that the percent area of plaque deposition in X11α knockout mice surpasses that of X11α–, X11β–, and X11γ–expressing mice in cortex regions, but not hippocampal. This discrepancy can be accounted by the presence of X11β and X11γ; Ho et al stated that the deletion of X11β showed the greatest decrease in amyloidosis (Ho et al, 2008). Should this study have included the study of mice that expressed APP but none of the X11 family members, as well as other combinations of X11 knockout mice that expressed APP, a clearer comparison of the effects of the X11 family members could have been reached. In the *in vitro* study by Xie et al., the genes encoding X11α and X11β were silenced by RNA interference, resulting in lowered Aβ levels in both cell strains.

GFAP immunocytochemistry illustrates the activation of astrocytes in damaged areas as response to central nervous system injury and stress, a reaction known as gliosis. GFAP immunolabeling conducted in this research showed significantly more gliosis in the hippocampus, especially the CA3 region, of APP23/X11α mice than in APP23 mice (Fig. 9), a confounded result that can be attributed to either the overexpression of X11α or to the aforementioned increased plaque deposition. Analysis of GFAP immunolabeled brain sections from wild type and X11α mice showed that X11α had statistically significant greater glioses (Fig. 10). The finding that higher level of gliosis was observed in the brains of APP23/X11α

compared to either APP23 or X11α conveys that X11α overexpression, not directly related to amyloidosis, in addition to augmenting amyloidosis, causes a greater stress response in mouse model of Alzheimer's disease.

The effects of X11α overexpression on amyloidosis may have significant implications for the progression of Alzheimer's disease. As such, increased X11α expression may be considered a risk factor for the development of AD; future research should delve into the mechanisms by which X11α modulates APP and possible methods of inhibiting these mechanisms as a potential therapeutic strategy.

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8. Bibliography

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