The presenilin 1 Protein Is a Component of a High Molecular Weight Intracellular Complex That Contains β-Catenin*

(Gang Yu‡, Fusheng Chen‡, Georges Levesque‡, Masaki Nishimura‡, Dong-Mei Zhang‡, Lyne Levesque‡, Ekatarina Rogaeva‡, Donghong Xu‡, Yan Liang‡, Monika Duthie‡, Peter H. St George-Hyslop‡§, and Paul E. Fraser‡

From the §Centre for Research in Neurodegenerative Diseases, Departments of Medicine and Medical Biophysics, University of Toronto, Toronto, Ontario M5S 3H2, Canada and ¶Department of Medicine (Division of Neurology), the Toronto Hospital, Toronto, Ontario M5S 3H2, Canada

The presenilin (PS) genes associated with Alzheimer disease encode polytopic transmembrane proteins which undergo physiologic endoproteolytic cleavage to generate stable NH₂- and COOH-terminal fragments (NTF or CTF) which co-localize in intracellular membranes, but are tightly regulated in their stoichiometry and abundance. We have used linear glycerol velocity and discontinuous sucrose gradient analysis to investigate the distribution and native conformation of PS1 and PS2 during this regulated processing in cultured cells and in brain. The PS1 NTF and CTF co-localize in the endoplasmic reticulum (ER) and in the Golgi apparatus, where they are components of a ~250-kDa complex. This complex also contains β-catenin but not β-amyloid precursor protein (APP). In contrast, the PS1 holoprotein precursor is predominantly localized to the rough ER and smooth ER, where it is a component of a ~180-kDa native complex. PS2 forms similar but independent complexes. Restricted incorporation of the presenilin NTF and CTF along with a potentially functional ligand (β-catenin) into a multimeric complex in the ER and Golgi apparatus may provide an explanation for the regulated accumulation of the NTF and CTF.

Mutations in the genes encoding the presenilin (PS)1 (PS1) and presenilin 2 (PS2) account for the majority of early-onset familial Alzheimer’s disease (1–3). Both genes encode polytopic transmembrane proteins that are predominantly localized in intracellular membranes, including the nuclear envelope, the endoplasmic reticulum, and the Golgi apparatus (4, 5). Structural studies suggested that these proteins contain either six or seven transmembrane domains, and that the amino and carboxyl termini are located as a large hydrophilic loop following the sixth TM domain are located in the cytoplasm (5–7). The presenilins undergo physiological endoproteolytic processing within the large cytoplasmic loop following TM6 by an unknown protease that produces heterogeneous ~29-kDa amino-terminal and ~18–20-kDa carboxyl-terminal fragments (8, 9). The presenilin holoproteins are maintained at low steady state levels both in brain and in other peripheral tissues, probably by proteasome-mediated degradation (10, 11). As a result, the presenilin species most readily detected are the stable NH₂- and COOH-terminal endoproteolytic fragments, the stoichiometry of which appears to be tightly regulated (8).

The functional role of the presenilins is still unknown, although roles in cellular differentiation, in signal transduction, in apoptosis, or in intracellular protein trafficking have been proposed. To better understand the presenilin proteins and their biological functions, we have investigated both the native state of these proteins and their biochemical subcellular localization in membrane fractions derived from cultured cells and from the human brain. Our data suggest that the presenilins form detergent-sensitive high molecular mass complexes. The PS1 holoprotein is a component of a complex of ~180 kDa, while the endoproteolytic fragments are components of a ~250-kDa complex that contains β-catenin (a member of the armadillo protein family which may have both structural and signal transduction roles) but not βAPP. In addition to forming complexes of differing sizes, the intracellular distribution of the holoprotein and the endoproteolytic fragments differ.

EXPERIMENTAL PROCEDURES

Protein Extraction from Cells—Untransfected HEK293 cells (10-cm dishes) or the transfected HEK293 cell line stably expressing wild type human PS1 and APP₆₉₅ (gift from Drs. D. Selkoe and M. Citron) were grown to confluence. Cells were washed twice with ice-cold phosphate-buffered saline and then lysed at 4 °C for 30 min with 1 ml of lysis buffer containing either 1.0% digitonin or 0.5% Triton X-100 (Boehringer Mannheim), 0.5% Nonidet P-40 (Boehringer Mannheim), or 0.5% SDS (Sigma) plus 25 mM Hepes, pH 7.2, 150 mM NaCl, 2 mM dithiothreitol, 2 mM EDTA, 5 μg/ml each of chymostatin, pepstatin, leupeptin, and antipain. Other protease inhibitor mixtures have been tested but do not alter the results reported here. Insoluble material was removed from the lysates by centrifugation at 17,000 × g for 15 min.

To prepare membrane extracts, cells were washed twice with ice-cold phosphate-buffered saline and resuspended in 10 mM Hepes, pH 7.2, containing protease inhibitors. After being swollen for 60 min, cells were subjected to a freeze-thaw cycle. The homogenate was clarified by centrifugation at 1000 × g for 15 min at 4 °C; the resulting supernatant was then centrifuged at 107,000 × g for 1 h at 4 °C, and the pellets were washed once with 10 mM Hepes, pH 7.2, containing protease inhibitors, followed by extraction with lysis buffer as above.

Human brain extracts were prepared by homogenizing 1.0 g of normal cerebral cortex in a total volume of 5.0 ml of 20 mM Hepes, pH 7.2, at 4 °C with protease inhibitors using a Polytron tissue shredder and a Teflon homogenizer followed by rehomogenization in the presence of 1.0% digitonin or 0.5% Nonidet P-40. Samples were centrifuged at 100,000 × g for 1 h at 4 °C.

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¶ To whom correspondence should be addressed: Centre for Research in Neurodegenerative Disease, Tanz Neuroscience Bldg., University of Toronto, Toronto, Ontario M5S 3H2, Canada.

1The abbreviations used are: PS, presenilin; TM, transmembrane; APP, amyloid precursor protein; CTF, COOH-terminal fragment; NTF, NH₂-terminal fragment; ER, endoplasmic reticulum, SER and dER, smooth and rough endoplasmic reticulum.
11,000 × g for 30 min, and the supernatant was used for gynecol
gradient centrifugation.

Glycerol Velocity Gradient Centrifugation—The gynecol gradient
centrifugation procedure was as described previously (12, 13). Briefly,
0.5 ml of total protein extracts or membrane extracts was applied to the
top of a 11.5-ml 10–40% (w/v) linear glycerol gradient containing 25 mM
Hepes, pH 7.2, 150 mM NaCl, and 0.4% appropriate detergent. Gradi-
ents were centrifuged for 15 h at 35,000 rpm and 4 °C using an SW41
rotor and collected by upward displacement into 1.0-ml fractions using
an Iuco model 640 density gradient fractionator. To quantitatively re-
cover proteins, 10 μg of bovine serum albumin were added to 150-μl
aliquots of each fraction, followed by precipitation with 5 volumes of
–20 °C acetone.

Immuno precipitation and Immunoblotting—Antibodies to PS1
COOH-terminal fragment (CTF) (rabbit polyclonal antibodies 520 and
1143 (4, 14, 15), to PS1 NH₂-terminal fragment (NTF) (antibody 14) (8),
and control antibodies (preimmune) were cross-linked to protein A-
Sepharose (Pharmacia Biotech Inc.) with dimethylimidate (Sigma).
Mouse anti-β-catenin and anti-γ-catenin (C-19220 and C-7207, respec-

tively, Transduction Laboratories) were coupled to protein G-agarose
(Life Technologies, Inc.). The 1.0% digitonin- or 0.5% Triton X-100-
extracted proteins from lysates (2 mg/ml) or gradient fractions were
preabsorbed with preimmune serum linked to beads, then combined
with the appropriate antibody linked to beads for 2 h at 4 °C. Beads
were washed four times for 15 min each with 1.0% digitonin or 0.2%
Nonidet P-40 in 50 mM Tris-HCl, pH 7.4, 2 mM EDTA, and 150 mM
NaCl. Immunoprecipitated proteins were eluted with 0.1 M glycine, pH
2.5, neutralized with 1.0 M Tris, separated on Tris-glycine polyacryl-
amide gels (Novex), and transferred to nitrocellulose or polyvinylidene

difluoride (Millipore). Western blots were probed with the appropriate
antibodies and detected using ECL (Amersham Corp.).

RESULTS

Identification of a Stable 250-kDa Complex of PS1 Endopro-
teolytic Fragments—To determine the native state of PS1 protein,
untransfected HEK293 cells were solubilized in nonen-
daturing digitonin buffers, and the extracted proteins were
fractionated on a linear glycerol velocity gradient. Both NTF
and CTF of PS1 were found in identical fractions (fractions 3–7)
with a peak in fractions 5 and 6 corresponding to an apparent
molecular mass of 250 kDa (note that because the apparent
molecular mass is also dependent upon relative buoyancy, it is
likely that the size of a multimeric complex estimated from the
glycerol gradient will be underestimated) (Fig. 1A). There was
virtually no PS1 immunoreactivity in the fractions correspond-
ing to molecular masses lower than 60 kDa even after pro-
longed exposure, implying that very little PS1 exists as stable
monomers in the digitonin lysate. The NTF and CTF of PS1
may therefore be components of the same high molecular mass
oligomeric complex in vivo.

To explore this hypothesis further, we examined the sensi-
tivity of the complexes to different detergents. When total
proteins from the untransfected HEK293 cells were extracted
in 0.5% Triton X-100 and then subjected to the same gradient
centrifugation, the majority of the NTFs and a portion of the
CTFs were detected in the low molecular mass range (less than
69 kDa) (Fig. 1A). However, the bulk of the CTFs still
appeared in the high molecular mass fractions at 150–200 kDa
(Fig. 1B) indicating either a stronger CTF self-association or
high affinity interactions with other proteins. In the presence
of 0.5% SDS, the high molecular mass complex was completely
disassembled, so that both the NTF and CTF were found within
the low molecular mass range (less than 69 kDa) (Fig. 1C).

In agreement with the observations from HEK293 cells, the
majority of PS1 NTFs and CTFs from digitonin extracts of
human brain were present in fractions 3 through 7, with a peak
at an apparent molecular mass of approximately 250–300 kDa
(Fig. 2A). As with the HEK293 cells, the NTF and CTF com-
plexes from human brain were detergent-sensitive. Thus, when
extracted in 0.5% Nonidet P-40, the brain NTFs were present
in fractions 1–5, while the majority of the CTF were found in
fractions 5 and 6 (Fig. 2B). When brain tissue was extracted
with Triton X-100 (data not shown), there was slightly more
disassembly of the complex than was observed in the Nonidet
P-40 treated brain extracts. Extraction of brain tissue with
SDS resulted in complete dissociation of the PS1 complex (Fig.
2C). The differential sensitivity of the complex to different
detergents is therefore similar in brain and in cultured
HEK293 cells.

To investigate the distribution of the low abundance PS1
holoprotein, we examined gynecol velocity gradient fractions
derived from digitonin extracts of an HEK293 cell line stably
transfected with human PS1 and βAPP695, which expresses
moderate levels of full-length PS1. As with the untransfected
HEK293 cells, the NTF and CTF were components of a ~250
kDa complex (fractions 5 and 6; Fig. 3). The PS1 holoprotein
appeared in fractions 2–7 (molecular mass of 69–250 kDa),
which partially overlapped with the fractions containing the
NTF and CTF (Fig. 3). However, the strongest holoprotein
signal was obtained in lower molecular mass fractions (Fig. 3,
fractions 4 and 5). This suggests that PS1 holoprotein is either
a digitoxin-sensitive part of the high molecular mass fragment
complex or, more likely, that it forms a distinct complex.

The PS1 NH₂- and COOH-terminal Fragments Associate
within the 250-kDa Complex—The presence of both the NTF
and CTF of PS1 in the same density gradient fractions does not
preclude the possibility that they form independent complexes
after endoproteolytic cleavage. To address this, fractions con-
taining the 250-kDa complex were pooled (fractions 3–8) and used for reciprocal immunoprecipitation studies with anti-CTF antibody (Fig. 4), or with anti-NTF antibody (not shown). As with co-immunoprecipitation experiments performed on whole cell lysates (Fig. 5 C and D; lanes 2 and 3), the CTF and NTF co-precipitate from glycerol gradient fractions containing the 250-kDa complex (Fig. 4). The specificity of the NTF-CTF association was confirmed by the absence of co-immunoprecipitation of holoprotein, NTF or CTF with: 1) preimmune serum (Fig. 4; lanes 4–6), or 2) with antibodies to irrelevant ER-resident proteins such as calnexin (data not shown). However, as would be expected from the detergent sensitivity of the 250-kDa complex, the NTF and CTF did not co-precipitate when the HEK293 cells were lysed with Triton X-100 (Fig. 5, C and D; lanes 5 and 6). Taken together, these results indicate that the NTF and CTF of PS1 are components of the same 250-kDa complex. The NH2- and COOH-terminal fragments of PS2 are also present in a similar high molecular mass complex in digitonin lysates of HEK293 cells (data not shown). However, PS2 NTF does not co-precipitate with either the NTF or the CTF of PS1 (Fig. 5 B). This suggests that the two presenilins form distinct protein complexes of similar size.

**Fig. 2.** PS1 endoproteolytic fragments exist in a detergent-sensitive 250-kDa complex in human brain. Immunoblots of the PS1 NH2 terminus and COOH terminus in sequential fractions from a linear 10–40% glycerol velocity gradient of human brain proteins extracted in 1% Digitonin (A), 0.5% Nonidet P-40 (B), or 0.5% SDS (C). Arrows at the top indicate the mobilities of molecular mass markers. Numbered fractions collected from the glycerol gradient are indicated at the bottom.

**Fig. 3.** The PS1 holoprotein and its endoproteolytic fragments are present in distinct complexes in a transfected HEK293 cell line stably overexpressing wild type human PS1 and APP. Immunoblot of full-length PS1 (PS1-FL), the NH2-terminal (PS1-NTF), and COOH-terminal (PS1-CTF) fragments in sequential fractions from a PS1/APP stable HEK293 cell line. PS1 holoprotein and NTF were detected with antibody Ab14. The COOH-terminal fragment of PS1 was detected with anti-loop antibody 1143. Arrows at the top indicate the mobilities of protein molecular mass markers. Numbered fractions collected from the glycerol gradient are indicated at the bottom.

**Fig. 4.** The NH2- and COOH-terminal fragments of PS1 co-immunoprecipitate from digitonin solubilized and gradient fractionated native HEK293 cell extracts. Fractions containing the PS1 fragments were pooled pairwise (fractions 3/4, 5/6, and 7/8) and immunoprecipitated with antibodies against PS1-CTF (indicated as Anti-PS1-C; lanes 1–3), preimmune serum (lanes 4–6), and PS1-NTF (indicated as Anti-PS1-N; lane 7). Immunoprecipitation products were probed with the anti-NTF antibody 14.

**Fig. 5.** The PS1 fragments and β-catenin are members of a ~250-kDa complex and interact in a detergent-sensitive manner. Untransfected HEK293 cells were solubilized in either 1% digitonin (lanes 2–4) or 0.5% Triton X-100 (lanes 5–7) and immunoprecipitated using the antibodies as indicated at the top; PS1 NH2-terminal-specific antisera (Anti-PS1-N), COOH-terminal-specific antisera (Anti-PS1-C), and preimmune sera. The immunoprecipitation products resolved on SDS-polyacrylamide gel electrophoresis and the immunoblots investigated with antibodies to: β-catenin (A); the presenilin-2 NH2-terminal fragment (B), PS1-NTF (C), or the PS1-CTF (D). The column labeled Lysate (lane 1) represents about 30% of the starting digitonin lysate utilized for immunoprecipitation, which was used as a positive control.
FIG. 6. The PS1 proteolytic fragments co-immunoprecipitate with β-catenin but not γ-catenin. HEK293 crude cell membrane extracts were prepared and solubilized in 1% digitonin and used for immunoprecipitation studies with anti-β-catenin (lane 2) or anti-γ-catenin (lane 3) antibodies. The immunoprecipitation products were separated by SDS-polyacrylamide gel electrophoresis and the immunoblots probed with a mixture of PS1 NH2- (Ab14) and COOH-terminal-specific (antibody 1143) antisera. This indicated that the NH2-terminal of NTF is contained in lower molecular mass fractions (<100 kDa). This suggests that under slightly stronger detergent conditions, the CTF and NTF dissociate leaving the CTF still associated with partners other than the NTF. Using yeast two hybrid screens, we and others have found that members of the armadillo family such as β-catenin and hNPRAP can interact with presenilins (22). Considering this, we examined the 250-kDa PS1 complex from HEK293 cells to determine whether it also contained endogenous β-catenin.

Antibodies against either the NTF or the CTF of PS1 both co-precipitated β-catenin from digitonin lysates of native HEK293 cells (Fig. 5A, lanes 2–4). Reciprocal co-immunoprecipitation experiments confirm that anti-β-catenin antibodies co-precipitate the PS1 NTF and CTF (Fig. 6, lane 2). We have shown that the armadillo-protein binding site of PS1 is located near residues 372–399 within the CTF.2 As would be expected from this, dissociation of the NTF and CTF by solubilization in Triton X-100 resulted in co-precipitation of β-catenin with the PS1 CTF but not with the NTF (Fig. 5A, lanes 5–7). These interactions are specific because the NTF and the CTF did not co-immunoprecipitate with either γ-catenin (Fig. 6, lane 3) or calnexin (data not shown).

Finally, endogenous β-catenin was present in fractions 2–10 of digitonin-extracted HEK293 cells (Fig. 7A). While the size distribution of β-catenin complexes overlaps that of PS1, it is obviously much broader than that of PS1. The broad size distribution of β-catenin complexes reflects the fact that β-catenin exists as free cytoplasmic molecules, and as complexes with proteins such as α-catenin, γ-catenin, cadherin, adenomatous polyposis coli, and glycogen-synthase-kinase-3β (23–25). A direct interaction between PS1 and β-catenin within the same ~250-kDa complex was confirmed by showing that antibodies to PS1-CTF could co-immunoprecipitate β-catenin and PS1-CTF only from the subset of fractions containing both proteins, i.e. fractions 3–7 (Fig. 7B). The profile of PS1 CTF-precipitated β-catenin is very similar to the migration profile of the fragments of PS1 in the glycerol gradient (compare Figs. 1A and 7). β-Catenin, NTF, and CTF are therefore tightly associated members of a ~250-kDa complex.

βAPP has also been proposed to interact with PS1 and PS2 (26, 27). However, in the double stable PS1/βAPP HEK293 cell line, βAPP immunoreactivity was found predominantly at ~150 kDa, close to its expected migration for a monomeric molecular mass of ~110 kDa (Fig. 7C). There was partial overlap with the migration of PS1 holoprotein although the peak immunoreactivities for each occur in different regions of the gradient (βAPP in fraction 3, Fig. 7C; PS1 holoprotein in fraction 4, Fig. 3). In contrast to the co-immunoprecipitation of PS1 and β-catenin, βAPP and PS1 could not be co-immunoprecipitated from fractions containing either the ~180-kDa or the ~250-kDa complexes (data not shown).

Subcellular Localization of PS1 Fragments and Holoprotein—The apparent difference between the sizes of complexes containing PS1 holoprotein and those containing the NTF/CTF implies that they may exist in different intracellular compartments. We therefore examined the distribution of PS1 holoprotein and NTF/CTF complexes by subcellular discontinuous sucrose density fractionation of homogenates from human brain, untransfected HEK293 cells, and HEK293 cells overexpressing both wild-type human PS1 and βAPP. The organellar distribution within the gradient was confirmed using the Golgi-specific marker protein, p58 (19, 20), and the ER-resident protein, calnexin (12, 21). In brain, as well as in transfected and untransfected HEK293 cells, PS1-NTF and PS1-CTF were both present in fractions from the Golgi apparatus and sER (Fig. 8). Slightly lower amounts were present in the rER (Fig. 8). This topographic co-localization of the NTF and CTF is in good agreement with the results described above showing that the NTF and CTF of PS1 form stable high molecular mass complexes.

In fractions from the PS1/βAPP stable HEK293 cell line, PS1 holoprotein partially overlapped the distribution of PS1 NTF and CTF (Fig. 8D). Thus, the majority of the holoprotein immunoreactivity was present in fractions corresponding to rER and sER, while the NTF and CTF predominated in fractions corresponding to Golgi as well as both the rER and the sER. Overexpression of PS1 may have caused a wider distribution of

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**Intracellular Presenilin Complexes**

**Fig. 8.** Biochemical subcellular localization of PS1 holoprotein and endoproteolytic fragments within the rER and sER and Golgi apparatus. Immunoblots are sequential fractions from discontinuous sucrose gradients containing cellular proteins with the organelle distribution shown by the localization of ER-resident calnexin and Golgi-specific p58 (A). Distribution within these compartments was investigated for full-length PS1 (PS1-FL as well as NH-terminal and COOH-terminal fragments from: human brain (B), native HEK293 cells expressing endogenous PS1 levels (C), and HEK293 cells stably over-expressing PS1 and APP (D). The location of β-catenin immunoreactivity relative to the various PS1 proteins is also indicated (E).

The sucrose gradient blots from the PS1/βAPP HEK293 stable cells and from human brain were investigated with anti-β-catenin antibodies (Fig. 8E). Although β-catenin is a soluble protein, it co-purifies with PS1 holoprotein and PS1 CTF in the rER, sER, and Golgi membrane fractions. This would be expected from its interaction with a domain in the large cytoplasmic loop of PS1.

**DISCUSSION**

We have identified a ~250-kDa native protein complex which contains at least β-catenin, PS1-NTF and PS1-CTF. This complex is present in human brain, in native HEK293 cells, and in PS1 transfected HEK293 cells. It is apparent that the molecular masses of PS1-NTF (29 kDa), PS1-CTF (18 kDa), and β-catenin (92 kDa) on a 1:1:1 stoichiometry do not account for all of the estimated mass of the ~250-kDa complex. It is unclear whether the additional mass reflects a different ratio of PS1 and β-catenin (e.g. 3:3:1 or 1:1:2) or, more probably, whether other unidentified PS1-associated proteins are present. Further studies will be required to address this question and identify these other components if they exist.

Like many membrane proteins, presenilins have a tendency to aggregate under non-native conditions (28). Several lines of evidence indicate that the ~250-kDa protein complex we have identified is authentic and is not caused by nonspecific aggregation. First, we analyzed the native state of endogenous PS1 by linear glycerol velocity gradients. Second, the same complex exists in both human brain and in the HEK293 cell line. Third, when PS1 is overexpressed the holoprotein complex appears to be smaller than the native fragment complex, an observation which contradicts the intuitive expectation that nonspecific aggregation should be more severe when PS1 is overexpressed. Fourth, the fact that the PS1 complex does not contain the highly homologous PS2 protein also contradicts the expectations for a nonspecific aggregation. Fifth, three components of the complex (PS1 NTF, PS1 CTF, and β-catenin) can be specifically co-immunoprecipitated from both whole cell lysates and from the appropriate glycerol gradient fractions. The identification of β-catenin as a member of the high molecular mass complex, but not other proteins such as γ-catenin, calnexin and βAPP (discussed below), also argues against a nonspecific aggregate. Finally, in agreement with previously published immunocytochemical studies on the intracellular distribution of presenilin proteins (4, 5), we can show that the three identified components of the ~250 kDa presenilin complex (β-catenin and NTF and CTF of PS1) are located in the same biochemically defined subcellular compartment.

βAPP has been reported to be capable of interacting with PS1 and PS2 (26, 27). However, βAPP is unlikely to be a significant component of the ~250-kDa PS1 complex described here for the following reasons. First, the expression level of endogenous βAPP is very low in HEK293 cells. Despite this, the glycerol gradient migration pattern of PS1 fragments in untransfected HEK293 cell lysates was very similar to that in lysates of both PS1/βAPP double-transfected HEK293 cells and of human brain. If βAPP is a major component of the complex, the PS1 complex would be expected to differ in size between the untransfected and the PS1/βAPP-transfected HEK293 cells. Second, the glycerol gradient fractionation profiles of βAPP and PS1 holoprotein only partly overlap even in digitonin extracts, and do not overlap at all with the PS1 fragment complex. Thus, only a small amount of βAPP and PS1 holoprotein would be available to interact. Finally, βAPP and PS1 do not co-precipitate from the glycerol gradient fractions containing PS1 and βAPP in digitonin lysates of βAPP/PS1 stable cells. However, we can not exclude the possibility of a greater detergent sensitivity of βAPP binding or that βAPP is a component of a minor PS1-containing complex. This would be especially true if this complex was the result of a weak or transient interaction with the PS1 holoprotein (27).

Whether the complex functions simply to fold, transport and process PS1, or whether it has other intrinsic functions remains to be determined. However, regulated insertion of the PS1 NTF and CTF into this complex does provide a potential explanation for the observation that the stoichiometry and level of these fragments appears to be tightly controlled in most cells (8). Previous investigations have demonstrated that the majority of the PS1 and PS2 holoproteins are rapidly degraded by the proteasome with half-lives of ~15 min (10, 11). It is likely that this represents a separate pathway for the removal
of full-length presenilins, and is consistent with our observation that most PS1 holoprotein exists as a separate complex which may be more susceptible to proteasome degradation. In contrast, PS1 CTF and NTF are incorporated into a stable ~250-kDa complex which contains at least one other potentially functional ligand (β-catenin). The regulated incorporation into this complex (e.g. by limited availability of one or more other ligands) would provide an explanation for the regulated abundance of the CTF and NTF. Furthermore, inclusion of the CTF and NTF into a stable complex would also account for the long turnover times of the cleavage fragments. It is unclear whether endoproteolysis occurs before or following insertion into the ~250-kDa complex. The transition from a lower molecular mass holoprotein enriched complex to the ~250-kDa fragment enriched complex could be brought about by an enhanced PS1 NTF/CTF oligomerization following endoproteolytic cleavage. Alternatively, the endoproteolytic cleavage might occur beforehand, but could expose previously internalized ligands. Understanding the mechanism and the composition of this complex will likely lead to a better understanding of PS1 function.

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