The Degradation of Apolipoprotein B100 Is Mediated by the Ubiquitin-proteasome Pathway and Involves Heat Shock Protein 70*

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Apolipoprotein B (apoB) is the major protein component of atherogenic lipoproteins of hepatic origin. In HepG2 cells, the standard cell culture model of human hepatic lipoprotein metabolism, there is a limited availability of core lipids in the endoplasmic reticulum for association with nascent apoB. Under these conditions, apoB is partially translocated, interacts with cytosolic Hsp70, and undergoes rapid degradation. We show that increasing the expression of Hsp70 in HepG2 cells promotes apoB degradation. In addition, apoB is polyubiquitinated and its degradation both normally and after Hsp70 induction is blocked by inhibitors of the proteasome. The apoB that accumulates after proteasome inhibition is endoplasmic reticulum-associated and can be assembled into lipoproteins and secreted if new lipid synthesis is stimulated. Thus, apoB is the first example of a wild-type mammalian protein whose secretion is regulated by degradation in the cytosol via the ubiquitin-proteasome pathway. Furthermore, targeting of this secretory protein to the proteasome is regulated by the molecular chaperone Hsp70 and the availability of apoB's lipid-ligands.

Apolipoprotein B100 (apoB)¹ is essential for the assembly and secretion of very low density lipoproteins from the liver, and the binding of apoB to the low density lipoprotein receptor promotes the tissue uptake of low density lipoproteins from the plasma (1, 2). The plasma level of apoB in humans correlates directly with the risk of coronary artery disease (3, 4). ApoB is a 540-kDa protein with a multidomain structure that includes a globular amphipathic domain at the amino terminus and several hydrophobic amphipathic β sheet domains throughout its length (5, 6). Like other secreted proteins, apoB is synthesized as a precursor containing a signal peptide that targets the nascent protein for translocation across the endoplasmic reticulum (ER) membrane (7). Unlike most secretory proteins, however, a significant proportion of newly synthesized apoB is degraded prior to secretion from hepatic cells (8–10).

In HepG2 cells, a human hepatocarcinoma cell line, a large fraction of apoB undergoes rapid degradation, but if oleic acid (OA) is provided to the cells, there is stimulation of the synthesis of lipid components of apoB-containing lipoprotein particles, especially triglycerides, and protection of apoB from rapid degradation (10, 11). The relative increase in degradation when lipid is limiting is associated with an apparent slowing, or even halting, of translocation, resulting in a “bitopic” orientation of apoB in which some domains are exposed to the cytosol and some to the ER lumen (12–14). The protection of apoB by newly synthesized lipid may reflect a direct role in facilitating apoB translocation across the ER membrane or an indirect one by promoting the recently demonstrated (15, 16) association between apoB and microsomal triglyceride transfer protein (MTP), the ER-luminal protein in liver required for the transfer of lipids to nascent apoB (17).

In its bitopic orientation, apoB associates with cytosolic heat shock protein 70 (Hsp70; 18), an abundant molecular chaperone which binds to unfolded proteins and facilitates their achieving native conformations or their translocation across membranes, including those of the ER (for recent reviews, see Refs. 19–22). In addition, recent studies have shown that Hsp70 and its cofactors participate in the degradation of certain proteins by the ubiquitin-proteasome pathway, by ATP-dependent proteases, and by lysosomes (23–25). Therefore, we set out to determine whether this chaperone may play a role in the degradation of nascent apoB. Because the protease inhibitor ALLN (acetyl-leucyl-leucyl-norleucinal; Refs. 26 and 27) protects apoB from degradation in HepG2 cells (e.g. Ref. 11), we also investigated the role of the ubiquitin-proteasome pathway in apoB degradation, given the recent demonstration that ALLN can inhibit protein breakdown by cytosolic proteasomes (28–30).

**EXPERIMENTAL PROCEDURES**

Reagents—Herbimycin A (HA) and acetyl-leucyl-leucyl-norleucinal (ALLN) were purchased from Sigma. ALLN was used at a concentration of 100 μM. MG132 was provided by ProScript, Inc. and used at a concentration of 10 μM. Lactacystin was either synthesized (31), provided by ProScript, Inc., or purchased from the laboratory of Dr. E. J. Corey, Department of Chemistry, Harvard University, and used at a concentration of 0.1 μM. The proteolytic inhibitors were dissolved in dimethyl sulfoxide; in control cell cultures, an equal volume of dimethyl sulfoxide (“buffer control”) was added.

³⁵[HI]Leucine was used at a concentration of 150 μCi/ml and was supplied as i-[4,5-³⁵H]leucine from Amersham with a specific activity of...
159 Ci/mmol. For the experiments in Fig. 3, [35S]methionine/cysteine was used at a concentration of 100 μCi/ml and was purchased from NEF Life Science Products as EXPRESS™ Protein Labeling Mix (specific activity >1000 Ci/mmol).

**Cell Culture**—HepG2 cells were grown as described in Ref. 10. Briefly, after seeding into collagen-coated plates, cultures were maintained at 37 °C/5% CO2 in minimal essential medium, 10% fetal bovine serum (with penicillin and streptomycin to inhibit bacterial contamination). The medium was changed every three days and experiments were started after cells were 70–90% confluent. During the experiments, cells were maintained at 37 °C/5% CO2 in serum-free medium with the indicated additions or treatments. In Figs. 3 and 5, OA was provided as a complex with BSA (prepared as in Ref. 10). The molar ratio of O/A:BSA was 2:1 and the concentration specified for the OA-BSA complex refers to the OA moiety.

In experiments in which HepG2 cells were transfected, the cells were treated with either Transfectam (Promega) alone (mock transfection) or Transfectam plus rat hsp72 cDNA. 48 h later, the cells were labeled for 2 h with [3H]leucine in the absence or presence of the indicated inhibitor (ALLN or lactacystin). Cell lysates and conditioned media were analyzed by immunoprecipitation with an anti-apoB antibody as below.

**Immunoprecipitation and Immunofluorescence Procedures**—The immunoprecipitations were performed under either non-denaturing (Fig. 1, panel D; Fig. 2, panel A; Fig. 5, panel A) or denaturing conditions (all others) and the immunoprecipitates resolved by SDS-PAGE as described previously (18, 32). The antibodies used were mouse anti-human Hsp 72/73 (Boehringer Mannheim), rabbit anti-human apoA-I (Calbiochem), rabbit anti-human apoB made by us (15) or obtained from Calbiochem, goat anti-human albumin (Boehringer Mannheim), and rabbit anti-ubiquitin (StressGen). For immuno-fluorescence studies, cells were fixed with 3% paraformaldehyde, permeabilized with 0.1% Triton X-100, and blocked with 1% BSA. The primary antibodies used were rabbit anti-human apoB (Calbiochem), mouse anti-human apoB (Caltag Laboratories), rabbit anti-human apoA-I (Calbiochem), mouse anti-58K protein (Sigma), and rabbit anti-calnexin (StressGen). The secondary antibodies were either Texas Red-conjugated goat anti-rabbit IgG or fluorescein isothiocyanate-conjugated goat anti-mouse IgG (Jackson ImmunoResearch Laboratories).

Fluorescent images were collected by a Nikon Optiphot-2 microscope connected to a Sony Digital Photo camera. The digitized images were printed using the Adobe Photoshop program.

**RESULTS**

The initial experiments used herbimycin A (HA), an ansamycin antibiotic that inhibits tyrosine kinases and markedly induces Hsp70 in several cell lines (33–35). Unlike heat shock or other injurious treatments that induce Hsp70, long term exposure to low doses of HA does not have any adverse effects on protein maturation, protein solubility, the integrity of the intermediate filament cytoskeleton, or overall cell viability (34). HepG2 cells were pretreated with HA for 0, 0.5, or 5 h, and ALLN was added to half of the cultures for the final 30 min of the 5-h period. All of the cultures were then incubated with [3H]leucine for 5 min and immediately lysed. HA did not appear to be toxic to the cells, since incorporation of [3H]leucine into albumin or total protein (trichloroacetic acid-insoluble material) was not changed (data not shown). As shown in Fig. 1A, treatment for 5 h, but not 0.5 h, resulted in markedly increased levels of Hsp70, as determined by immunoprecipitation with an anti-Hsp70 antibody. There was no effect of either HA or ALLN on the incorporation of [3H]leucine into immunoprecipitable apoB (Fig. 1, B and C). Since apoB intracellular degradation is negligible within an 8-min labeling period, these results indicated that neither HA nor Hsp70 affected the rate of apoB synthesis.

To test whether these treatments affected apoB degradation, a pulse-chase protocol was used. HepG2 cells, pretreated with HA and/or ALLN, were labeled with [3H]leucine for 15 min and then incubated without isotope for 40 min. For incubations in which ALLN was added in the pretreatment period, it was also present during the labeling and chase periods. Since rates of apoB synthesis were not altered by the treatments, and because minimal amounts of newly synthesized apoB are secreted in 40 min (10), the levels of radiolabeled apoB in the cell lysate at the end of the chase period reflected primarily intracellular degradation. As shown in Fig. 1, D and E, treatment with HA for 5 h, which markedly increased Hsp70 levels, resulted in significantly (p < 0.001) reduced recovery of apoB (lane 3 versus lane 1; bar 3 versus bar 1); thus, degradation of newly synthesized apoB was accelerated. This rapid degradation was inhibited by the addition of ALLN 30 min prior to the pulse labeling (lane 3 versus lane 4; bar 3 versus bar 4). In contrast, pretreatment with HA for only 30 min did not affect Hsp70 levels (Fig. 1A) and did not increase apoB degradation (lane 2 versus lane 1; bar 2 versus bar 1).

These consequences of HA treatment on apoB degradation were likely due to a direct effect of the induced chaperone. For example, after HA treatment, both Hsp70 levels and the association of Hsp70 with apoB were increased, as shown by co-immunoprecipitation of the two proteins with an anti-apoB antibody (Fig. 1D; lanes 3 and 4, lower bands). As noted above, HA appears to specifically induce Hsp70 in some cell types (35), but in others it can induce several heat shock proteins (34). To confirm that the results with HA were specifically due to the elevated level of Hsp70, HepG2 cells were either mock transfected or transfected with the cDNA for rat Hsp72. After a 2-h labeling period with [3H]leucine, apoB was immunoprecipitated from samples of cell lysate and conditioned medium. Hsp70 levels were significantly elevated following transfection and, consistent with the HA results, the recovery of apoB was decreased in the transfected cells, undoubtedly due to in-
creased degradation, since ALLN blocked the effect of increasing the level of Hsp70 on apoB recovery (data not shown).

As noted earlier, when HepG2 cells are provided with OA, apoB translocation is favored and apoB degradation decreases, as does the extent of the association of Hsp70 with apoB (15, 18, 36). We therefore tested whether this protection from degradation would be affected by increased levels of Hsp70. HepG2 cells were treated with either OA alone, or with HA plus OA. After a 15-min incubation with [3H]leucine, cells were maintained in isotope-free medium, and labeled apoB in the cell lysate and medium was then measured at different times. As shown in Fig. 2, OA treatment significantly reduced apoB degradation, and, as a result, both cell and medium apoB levels were greater than in the untreated control wells (panels A and B). However, HA co-treatment, which markedly increased Hsp70 levels, decreased this protective effect of OA; thus, the amounts of both cell and medium-apoB in HA/OA-treated cells were less than those in OA-treated cells, but still more than those in control cells. As observed earlier (Fig. 1D), the greater apoB degradation observed in HA/OA-treated cells (compared with OA alone) was associated with increased apoB-Hsp70 complex formation (Fig. 2A, lower bands). In control experiments, treatment with HA for 4 h did not affect TG synthesis under either basal or OA-stimulated conditions, as measured by incorporation of [3H]glycerol into cellular TG (data not shown).

Taken together, these results suggest that elevated levels of Hsp70 increased the susceptibility of apoB to a degradative system, especially when lipid synthesis was not stimulated and apoB translocation was not favored. In addition, the enhanced apoB degradation due to increased Hsp70 levels was inhibited by ALLN, as is degradation under basal conditions (11).

Despite the important influence of degradation in regulating the net secretion of hepatic apoB-containing lipoproteins, the proteolytic machinery involved has not been identified definitively. A prime candidate is the proteasome, which has been shown recently to be the major site of degradation of cytosolic proteins in mammalian cells (28, 30). The proteasome is found in two major forms, a 20 S (700 kDa) and a 26 S (2000 kDa) particle. The 26 S form contains a 20 S particle associated with two 19 S (∼600 kDa) regulatory complexes. The 20 S particle contains multiple peptidase activities and the 19 S complex provides the components necessary for the binding and ATP-dependent degradation of ubiquitinated proteins (30).

Several observations are consistent with a role for the proteasome in apoB degradation. 1) The breakdown of pre-secretory apoB is not reduced by chloroquine or ammonium chloride,2 which inhibit lysosomal proteolysis; 2) ALLN, although originally identified as an inhibitor of cysteine proteases, such as the calpains and cathepsins (26, 27), can also inhibit proteasomes (28–30); 3) apoB degradation requires ATP (37) as does the ubiquitin-proteasome pathway; and 4) recent studies suggest that Hsp70 and its co-factors (such as the DnaJ homologues) can participate in the degradation of other proteins by the ubiquitin-proteasome pathway (23, 25).

The hypothesis that the proteasome degrades apoB in HepG2 cells was tested by using inhibitors of the proteasome, MG132 and lactacystin, which are more potent and specific than ALLN (28, 30, 38–40). Like ALLN, MG132 is a peptidyl aldehyde that competitively inhibits the chymotrypsin-like and peptidyl glutamyl sites on the 20 S proteasome. Lactacystin is a natural product that covalently modifies the active site threonine of the 20 S catalytic site and does not affect any other

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2 H. Wang, E. A. Fisher, X. Wu, J. L. Dixon, and H. N. Ginsberg, unpublished data.

3 N. Sakata and H. N. Ginsberg, unpublished data.

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Fig. 2. HA treatment partially reversed the enhanced secretion and reduced degradation of apoB associated with OA treatment. HepG2 cells were pretreated with either OA (0.8 mM) complexed to BSA (OA/BSA) for 2 h, or with HA (1.0 μg/ml) for 4 h in the presence of OA/BSA during the last 2 h. Cells were then radiolabeled with [3H]leucine for 15 min and chased for different periods of time as indicated. Cell lysate and medium samples were collected for immunoprecipitation with anti-apoB antibody. Panel A, results for cell lysate samples. The 70-kDa bands in the fluorogram (top) were confirmed as apoB and Hsp70 by Western blotting (18). The numerical data (bottom) are expressed as percent of initial apoB synthesized and plotted as the mean ± S.D. (n = 3). Panel B, corresponding results for conditioned medium samples.
another peptidyl aldehyde, MG115, which weakly inhibits the proteasome, also decreased apoB degradation in HepG2 cells (41). Overall, then, the results in Fig. 3, A and B, suggest that apoB can be degraded by proteasomes in HepG2 cells and the reported effects of ALLN on apoB degradation (e.g. Refs. 11, 13, and 14) can be attributed to its inhibition of proteasomes and Hsp70.

**FIG. 3.** Proteasome inhibitors decreased apoB degradation and OA stimulated the secretion of apoB spared from degradation. Panel A, HepG2 cells were labeled for 2 h with [35S]methionine/cysteine (100 μCi/ml) in the absence of proteasome inhibitors (lanes 1–3) and in the presence of MG132 (lanes 4–6), lactacystin (lanes 7–9), or ALLN (lanes 10–12). The concentrations of the inhibitors were 10, 10, and 100 μM, respectively. *Top,* SDS-PAGE analysis of apoB immunoprecipitated from cell lysates; *bottom,* SDS-PAGE analysis of apoB immunoprecipitated from conditioned medium. Panel B, the apoB signals on fluorograms, such as those shown in panel A, were quantified by PhosphorImager analysis (Molecular Dynamics). After normalization to total (lysate + medium) trichloroacetic acid-insoluble counts/min, the results were expressed as the total labeled apoB recovered relative to that recovered from control culture wells, and are plotted as the means ± S.D. (*p* < 0.001 versus control for each inhibitor, *n* ≥ 5 for each condition). Panel C, cells were labeled with [35S]methionine/cysteine for 2 h in media containing BSA (control, lanes 1–6) or 0.8 mM OA/BSA (lanes 7–12) in the absence of proteasomal inhibitors (lanes 1–2 and 7–8) or in the presence of lactacystin (lanes 3–4 and 9–10) or MG132 (lanes 5–6 and 11–12). ApoB was immunoprecipitated from samples of cell lysate (top) and conditioned medium (bottom) followed by SDS-PAGE analysis. The resulting fluorogram is shown. The treatments for each lane were: control (lanes 1 and 2); lactacystin (lac, 3 and 4); MG132 (5 and 6); OA/BSA (OA, 7 and 8); lactacystin + OA (lac+OA, 9 and 10); MG132 + OA (11 and 12). Panel D, cells were pretreated with either lactacystin or buffer for 45 min and pulse-labeled with [35S]methionine/cysteine during the last 15 min. Monolayers were washed and cells were incubated with isotope-free chase medium. 20 min after the initiation of the chase period, OA/BSA or BSA alone was added, and apoB was immunoprecipitated from conditioned medium 0, 30, 90, and 150 min following this addition. The amounts of apoB after SDS-PAGE separation were quantified by PhosphorImager analysis. Labeled apoB recovery from the medium at each time point is expressed relative to the labeled cellular apoB content at the end of the 15-min labeling period (data not shown). Symbols represent cells treated with BSA alone ( ), lactacystin and BSA ( ), or lactacystin and OA ( ).
not other proteolytic activities.

We next investigated whether the apoB that accumulated when proteasome function was blocked could be secreted when lipid synthesis was stimulated. HepG2 cells were incubated as in Fig. 3A, but in some wells OA was added along with the proteasome inhibitors. A large fraction (typically over 50%) of the labeled apoB was recovered from the conditioned medium at the end of the experiment (Fig. 3C, bottom). In pulse-chase experiments, a similar result was obtained: cells were incubated for 15 min with [35S]methionine/cysteine in the presence or absence of lactacystin. This medium was then replaced by one that was isotope-free and the incubation continued for 20 min to allow for all of the incomplete radiolabeled apoB molecules to be translated into full-length proteins (8, 42). OA was then added to some wells, and aliquots of conditioned medium from the three treatment groups (no OA or lactacystin, lactacystin, OA + lactacystin) were collected for immunoprecipitation analysis at different times following OA addition. As shown in Fig. 3B, over the duration of the 150-min incubation with OA, the secretion of radioiodinated apoB synthesized in the presence of lactacystin was stimulated. These two experiments (Fig. 3, C and D) demonstrated that the apoB spared from degradation by the proteasome could be incorporated into lipoproteins and secreted if lipid synthesis was stimulated either during or after the translation of apoB. Similar results (i.e. the stimulation of apoB secretion) were obtained even when OA was added to lactacystin-treated cells as long as 2 h after the 15-min labeling period (data not shown). None of the inhibitors significantly affected the total protein content of the cells or the synthesis and secretion of total radiolabeled protein (n = 6 for each parameter). In addition, the cellular content of another apolipoprotein, apoA-I, which is the major protein of high density lipoproteins, was not significantly affected by proteasomal inhibition (n = 5).

We next investigated the cellular location of apoB that accumulated during proteasomal inhibition. Cells were incubated with [35S]methionine/cysteine for 2 h and some cells were treated with either MG132 or lactacystin. Cell lysates were centrifuged at 4 °C and 100,000 × g for 1 h to obtain microsomal (pellet) and supernatant (S100) fractions. Whatever the treatment, over 80% of the labeled apoB was found typically in the microsomal fraction (data not shown), suggesting that apoB protected from degradation remained membrane-associated.

Another approach was to study the cells by immunofluorescence, which showed (Fig. 4) an accumulation of apoB in the cells after proteasomal inhibition, in agreement with the subcellular fractionation data and Fig. 3A. Without the inhibitor (Fig. 4A), most of the apoB signal appeared to be localized to the Golgi, based on its appearance and its co-localization with the 58-kDa Golgi-specific protein, but not with the ER-specific protein, calnexin (data not shown). This was expected, since we have previously shown that once apoB was transported to the Golgi, it was relatively stable (43), whereas rapid degradation of apoB in the absence of inhibitor would have resulted in a small steady-state pool of ER-associated apoB. After inhibition of proteasomes (Fig. 4B), the apoB signal was not only distributed more generally than in the untreated control cells (Fig. 4A), but it also substantially overlapped with the signal for calnexin (data not shown). These findings suggested that pre-Golgi apoB was now stable and did not accumulate in the cytosol. Moreover, these immunofluorescence results were specific for apoB, since lactacystin treatment did not alter the distribution of apoA-I in the cells (data not shown).

As noted earlier, the increased apoB degradation in cells with elevated levels of Hsp70 was inhibited by ALLN. To test more conclusively whether this degradation was also mediated by the proteasome, HepG2 cells were either mock-transfected or transfected with rat hsp72 cDNA, and the effects of lactacystin examined. Cells were radiolabeled with [3H]leucine for 15 min and incubated for 20 min in isotope-free medium. Transient transfection resulted in elevated Hsp70 levels (Fig. 5A, lanes 3 and 4). Hsp70 overexpression, however, did not affect albumin synthesis or secretion (data not shown). In contrast, radiolabeled cell apoB levels in the hsp72 cDNA-transfected cells were approximately 50% lower than in the mock-transfected (control) cells (n = 3, p < 0.01; Fig. 5A, lane 3 versus 1). The addition of lactacystin to either the mock or hsp72 cDNA-transfected cells increased the total recovery of apoB by approximately 2- and 4-fold, respectively (Fig. 5A, lane 1 versus 2, lane 3 versus 4; 5B, bar 1 versus 2, bar 3 versus 4). Thus, both basal apoB degradation and that induced by high levels of Hsp70 were mediated by the proteasome.

Although some polypeptides may be directly degraded by 20
S proteasomes, the selective elimination of many regulatory proteins and proteins with highly abnormal conformations involves their covalent conjugation to multiple ubiquitin molecules. This modification leads to their rapid hydrolysis by the 26 S proteasome complex (30). We therefore tested in HepG2 cells whether degradation of apoB involved its ubiquitination. After a 1-h incubation with [3H]leucine, radiolabeled material with a higher molecular weight than apoB was detected after cell lysates were first immunoprecipitated with the anti-ubiquitin antibody, resuspended, and then immunoprecipitated with the anti-apoB antibody (Fig. 6A, lane 2). That this material represented apoB-ubiquitin conjugates was also supported by competition experiments in which the addition of unlabeled apoB (in the form of low density lipoprotein) to samples before the second immunoprecipitation step reduced the recovery of labeled material migrating at or above the expected position of apoB (Fig. 6A, lane 3). There was no evidence for apoB-ubiquitin conjugates in the conditioned medium (data not shown).

Furthermore, when a 4% polyacrylamide gel was used to better resolve high molecular weight proteins (Fig. 6B), treatment with lactacystin led to the accumulation of apoB-ubiquitin conjugates (Fig. 6A, lane 3). Immune complexes were collected with Protein A-Sepharose 4B beads. The beads were then treated with 2% SDS at 100 °C for 4 min. The supernatant was diluted 20-fold with 1% Triton X-100 and immunoprecipitated with rabbit anti-human apoB antibody. In some samples (lane 3), unlabeled apoB in the form of low density lipoprotein (1.5 mg) was added prior to the second immunoprecipitation. After the two immunoprecipitations, immune complexes were separated by SDS-PAGE (3–15% gradient gel) followed by fluorography. Panel B, HepG2 cells were pretreated with or without lactacystin (10 µM) for 4 h before radiolabeling. After the two immunoprecipitations, immune complexes were separated by SDS-PAGE using a 4% gel.

**DISCUSSION**

Overall, the results in this report show that in the standard model of human liver lipoprotein metabolism, the HepG2 cell line, apoB degradation is mediated by the proteasome and that Hsp70 participates in this process. Although apoB is a secretory protein, and Hsp70 and the proteasome are found in the cytosol, recent studies have shown that proteasomes can degrade other proteins that are either secretory (44–46) or trans-membrane (29, 47–49). These examples, however, primarily involve the degradation by the ubiquitin-proteasome pathway of a non-secretory or mutant protein (e.g. CFTR (29) or α1-antitrypsin Z (45)). In contrast, apoB is a wild-type mammalian secretory protein targeted to the proteasome as part of a metabolic regulatory mechanism; i.e. degradation by the ubiquitin-proteasome pathway is increased when lipid synthesis is low. In this novel mechanism, the itinerary of a secretory protein is governed in the disruption of the ribosome-translocon junction (60, 61) and the cytosolic exposure of hydrophobic apoB domains. Note that the ER protein MTP is a heterodimer of a 97-kDa subunit and protein disulfide isomerase (PDI). Path 1, when adequate core lipids (CE, cholesterol ester; TG, triglyceride) are available and transferred by MTP, forward translocation is accelerated (or re-initiated), and apoB is assembled into a lipoprotein and targeted for secretion. Under these conditions, newly synthesized apoB does not tend to associate with Hsp70 (18). Path 2, when there is inadequate synthesis of core lipids, apoB is associated with Hsp70 (18), ubiquitinated, and degraded by the proteasome, possibly in conjunction with retrograde movement of the amino-terminal portion of apoB.
ApoB Degradation Involves the Proteasome and Hsp70

ApoB Degradation Involves the Proteasome and Hsp70 pathway have the potential to regulate the hepatic production of atherogenic lipoproteins.

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