Despite rising interest in the health problems of the elderly, information on senescence-related alterations in essential metabolic pathways and their responses to various chemicals is scarce. Although peroxisomal pathways are involved in a multitude of cellular functions, little attention has been given to the potential relationship between senescence of these organelles and the process of aging and disease. Although the prevailing experimental evidence points to a decline in liver peroxisomal enzyme activities and a muted response to peroxisome-proliferating chemicals in aged animals, it is also evident that aged animals are more susceptible, in comparison to their young counterparts, to the hepatocarcinogenic effects of these chemicals. Furthermore, little is known about extraperoxisomal effects of peroxisome proliferators in aged animals. This review evaluates published studies on the impact of aging on basal hepatic peroxisomal metabolism, response to peroxisome proliferators, and changes in signal transduction pathways involved in these processes, with the aim of stimulating research efforts in this important area. The potential intricate relationship among senescent peroxisomes, aged hepatocytes, and health are also discussed.

**Key words:** aging, peroxisomal disorders, peroxisome proliferator-activated receptors, peroxisomes. Environ Health Perspect 107:791–797 (1999). [Online 31 August 1999]
http://ehpnet1.niehs.nih.gov/docs/1999/1107p791-797/yowssefaract.html

**Aging:**
Despite the myriad of peroxisomal enzymes involved in essential cellular metabolic pathways, little attention has focused on the relationship among changes in the basal activities of these enzymes, their response to peroxisome-proliferating chemicals, and the process of cellular aging. This paucity of information is particularly alarming in light of the fact that the elderly population, particularly in the industrialized world, is exposed over its life span to an ever-increasing number of peroxisome-proliferating chemicals in the form of therapeutic agents and environmental pollutants.

**Aging of the Liver**
The liver, like most organs, is altered morphologically and functionally in old organisms; many age-related changes in hepatocytes are similar to alterations observed in other cell types (Table 1; [4,5]). Therefore, common mechanisms of aging may operate at the cellular level and cause widespread decline in physiologic processes, eventually culminating in death ([5]). The aged liver has a reduced ability to regenerate (6,7). Increases in mitosis in response to hepatocarcinoma or to chemicals are diminished in old animals, as compared to their young counterparts ([6–9]). Furthermore, the pool of proliferating hepatocytes is smaller in old animals, and is more sharply localized to perportal hepatocytes ([7]). The importance of these changes in aging is unclear.

Studies have produced conflicting data on the effect of aging on the size of hepatocytes. Although most studies described conspicuous enlarged as well as small hepatocytes in old rats ([7]), others found far fewer variations of cell size, especially in female rats ([7]). Interestingly, hepatocellular organelles age faster than the cell ([7]). Mitochondria appear to generally decrease in number and size as a function of aging; the smallest mitochondria with shortest cristae were observed in the oldest rats ([5,7]). Similar observations have been documented in both mice and humans ([7,10,11]). Overall mitochondrial phosphorylation and turnover of mitochondrial proteins are reduced in aged liver ([7,12]). Similarly, the endoplasmic reticulum seems diminished in aged animals ([7,13]). In rats, studies have documented the existence of an age-related decline in several monoxygenase activities without any evidence of significant changes in cytochrome 450 or reduced nicotinamide adenine dinucleotide phosphate cytochrome redactase content ([14]). In contrast to the situation in aging rats, mice demonstrate a decline in several monoxygenase activities with age ([14]). However, benzo(a)pyrene hydroxylase and 7-ethoxycoumarin deethylase showed significant increases in senescent mice ([14]). Conversely, activity of liver cytosolic alcohol dehydrogenase increased with age in male Fischer 344 rats ([15]). However, activity of this enzyme was not altered with age in female rats, but was higher than in males in both young and old rats ([15]). In contrast, hepatic aldehyde dehydrogenase activity was similar in both male and female rats and was unchanged with age ([15]). Lysosomes are conspicuously increased in aged animals, with their volume density reaching approximately five times at birth ([7]).

**Peroxisomes: Function and Proliferation**
The peroxisome is a ubiquitous single membrane-limited cytoplasmic organelle present in animal, plant, and fungal cells ([16]). Peroxisomes are most abundant in the liver and kidney ([7]) and morphometric studies show that there are between 370 and 620 peroxisomes per rat hepatocyte, occupying 1.5–2% of the cell volume ([18]). The average peroxisome is approximately one-fifth the volume of a mitochondrion ([19]). Peroxisomal proteins are synthesized on free polyribosomes, with most of them made at the time of their synthesis. These proteins are then post-translationally imported from the cytosol into peroxisomes, with half-lives ranging from 1 to 15 min or longer ([16]).

**Peroxisome function.** Peroxisomes are respiratory organelles that play a role in cellular oxygen consumption as well as in production and removal of reactive oxygen species ([20]). Peroxisomes are involved in a number of essential cellular functions, including, but not limited to, cellular respiration, gluconeogenesis, lipid metabolism, thermogenesis, and synthesis of plasmalogens ([21]). Among the oxidases identified in peroxisomes are urate oxidase, acyl-CoA oxidase, amino acid oxidase, polyamine oxidase, trihydroxycisolestanoyl-CoA oxidase, and pristanoyl-CoA oxidase ([22]). Peroxisomal β-oxidation

Address correspondence to M. Badr, University of Missouri-Kansas City, 2411 Holmes Street, M3-115, Kansas City, MO 64108-2792 USA. Telephone: (816) 235-1842. Fax: (816) 235-1776. E-mail: badrm@umkc.edu

We thank B.S. Warren, J.A. Handler, and R.N. Miranda for their valuable suggestions in preparing this review. Aging studies in our laboratories are supported by a grant from the National Institutes of Health, CA/OD 74384.

Received 10 March 1999; accepted 25 May 1999.
of fatty acids is among the best characterized peroxisomal metabolic pathways. This system is not a functional duplicate of the mitochondrial system, as it is now clear that peroxisomes are involved in the degradation of a distinct set of compounds such as the very long-chain fatty acids and branched-chain fatty acids (22). Oxidation of erucic acid, arachidonic acid, and tetracosatetraenoic acid is initiated in peroxisomes (22). Mammalian peroxisomes are also a site for cholesterol and ether lipid synthesis (22).

**Peroxisome proliferation.** Various chemicals, conditions, and factors cause peroxisome proliferation. This phenomenon was first described in the 1960s. It was observed that numerous electron dense single membrane-limited structures appeared in rat liver following clofibrate feeding (23). Since that time, a myriad of therapeutic agents, industrial chemicals, and environmental pollutants (Figure 1 and Table 2) of diverse chemical structure have been shown to cause peroxisome proliferation, particularly in rodent livers (17,24). This group of chemicals has thus been referred to collectively as peroxisome proliferators. In addition to increasing the number of peroxisomes, these chemicals induce the activities of peroxisomal enzymes and cause hepatomegaly (17). These effects are not necessarily coupled, although they are dose and time dependent (17). Furthermore, these effects are tissue and species specific (25–30), with the liver being the most responsive tissue (17).

In addition to peroxisome-proliferating chemicals, various other conditions and factors cause peroxisome proliferation, albeit not to the same degree as with xenobiotics. Among these conditions and factors are diabetes (31,32), changes in thyroid hormone levels (33,34), cold adaptation (17), endotoxin exposure (35), high-fat diets (36), and partial hepatectomy (37). These conditions and factors are all believed to produce an effect similar to that initiated by the peroxisome-proliferating xenobiotics.

Mechanisms involved in the response to such a diverse group of compounds, factors, and conditions are unclear. However, a receptor-based mechanism for the pleiotropic response to peroxisome proliferators in rodents has been suggested, where a nuclear peroxisome proliferator receptor belonging to the steroid hormone receptor superfamily has been identified (38,39). This receptor is activated by several peroxisome proliferators; thus, it is termed the peroxisome proliferator-activated receptor (PPAR). Activation of PPAR stimulates the expression of genes encoding peroxisomal proteins (40). Recently, three isoforms of the PPAR have been found and cloned (41).

Prostaglandin 15-deoxy-Δ12,14, PGJ2 and the antidiabetic thiazolidinediones are ligands for the PPARy subtype of this receptor (Figure 1) (42). These chemicals promote differentiation of preadipocytes and act as an insulin-sensitizing agent (43). Activation of PPARδ subtype, however, was not enough to potentiate preadipocyte differentiation, nor did it result in modulation of glucose or triglyceride levels in vivo (43). On the other hand, leukotriene B4 and several known peroxisome-proliferating agents including the hypolipidemic and hypocholesteremic WY-14,643 and fibrates, as well as fatty acids and eicosanoids, bind specifically to and activate PPARα (Figure 1) (44–47). Activation of this receptor subtype significantly reduces serum triglycerides, but with minimal effects on hyperglycemia (43).

Studies have documented the existence of a human form of PPARα (hPPARα) (48,49) and PPARy (hPPARy) (50). The tissue distribution pattern of hPPARα mRNA is similar to that of the rat PPARα. Both are highly expressed in the liver and kidneys, whereas both are expressed at low levels in the brain and lung, with lower levels in most human tissues as compared to rats (49,51). Interestingly, the relative expression of PPARα mRNA in human skeletal muscle is higher than in rodent skeletal muscle (52). This finding may be significant because studies focus almost exclusively on the liver as a site to compare the role of PPARα in gene transcription in humans and rodents.

**Effect of Aging on Peroxisomal Metabolism**

Information on the relationship between aging and peroxisomal metabolism is scarce. Studies point to a general decrease in peroxisomal function with aging (52,53). However, several studies failed to document measurable differences in peroxisomal enzyme activities...
between young and old animals (54,55). Peroxisomes become smaller but more numerous in older rats (7). In aged rats, peroxisomes showed decreased catalase and acyl-CoA oxidase content, but increased content of thiolase and urate oxidase (56). The decrease in catalase activity coupled with the net increase in activities of oxidases are thought to contribute to oxidative stress (57). It has been shown that peroxisome proliferation was not accompanied by enhanced levels of oxidative damage in young mature animals (24,58). However, whether aging lowers the threshold for peroxisome proliferator-induced oxidative stress is not known, but may be implied from the enhanced susceptibility of aged animals to the hepatocarcinogenic effects of peroxisome proliferators (55,59).

**Basal peroxisomal enzyme activities in aged animals.** Apparent inconsistencies in age-related effects on peroxisomal β-oxidation (Table 3) may have to do with species differences and the age of animals at the time of sacrifice, as opposed to their age at the beginning of the experiment. For example, in some studies animals were kept for extended periods, which resulted in younger and older animals being senescent at the time of sacrifice (Table 3). Another potential reason for these apparent disparate results may reside in the ages of animal groups used, as peroxisomal enzyme activities appear to change dramatically and abruptly at certain age points during development and aging (52). The decline of catalase activity with aging in male Fischer 344 rats has been observed in various studies (Table 4). In our laboratories, hepatic catalase activity declined by 20% between the ages of 4 and 10 weeks, with most of the decline (30%) observed between the ages of 50 and 100 weeks (Table 4). Similar findings were also observed by others (52) for both peroxisomal β-oxidation and catalase in both CSWV and OFi mice strains (Tables 3 and 4), and similar findings were reported for the enzymatic activity and mRNA coding for catalase in rat liver (63,64). Transcription of the catalase gene decreased by approximately 60% between 6 and 29 months of age (64). Thus, the age-related decline in catalase mRNA levels appears to arise primarily from a decrease in the transcription of the gene (64). Quantitative analysis of kinetic parameters (rates of protein synthesis and degradation) revealed that the aged rat liver exhibited a decrease in rates of catalase synthesis without significant changes in degradation rates (65).

**Peroxisome proliferation in aged animals.** Aging also interferes with the ability of the liver to respond to peroxisome-promoting chemicals (60). Peroxinsol area relative to hepatocellular cytoplasmic area increased by approximately 7-fold in both 8- and 52-week-old rats following 14 days of daily 200 mg/kg clofibrate doses in diet; it increased only by 4-fold in 117-week-old rats (60). In the same rats, increases in hepatic peroxisomal β-oxidation showed a similar pattern, with increases approaching 15- and 11-fold in 8- and 52-week-old rats, but only 3-fold in the 117-week-old rats (60). In our laboratory, perfluorooctanoic acid (PFOA) induced peroxisomal β-oxidation significantly in male Fischer 344 rats of varying age groups to different levels. Forty-eight hours after 150

---

**Table 2. Peroxisome proliferators in the environment.**

| Chemical            | Major uses                      | Production in the United States | Source and concentration in the environment | Human body burden | Observed human toxicity |
|---------------------|---------------------------------|---------------------------------|---------------------------------------------|-------------------|-------------------------|
| Diethylhexylphthalate | Plasticizer, Lubricating oils, Insect repellent, Cosmetic formulations | 292,500,000 lb (1986) | Drinking water (~30 ppb) Groundwater (2.4 ppb), Surface water (1-83 ppb), Surface sediment (170 ppm), Atmosphere (~14 ng/m³), Fish (0-16,000 µg/kg), Milk (0.8 mg/L) | 0.3-1.15 ppm in adipose tissue | Hepatic peroxisome proliferation in dialysis patients, Necrotizing enterocolitis in neonates after insertion of umbilical catheters, CNS depression, Opacification of the lung |
| Dibutylphthalate    | Insect repellent, Plasticizer, Solvent, Solid rocket propellant | 6,662,000 lb (1993) | Drinking water (0.1-470 ppb) Groundwater (0.73-2.38 ppb), Surface water (0.1-0.8 ppb), Canned tuna (0.7-38 ppb), Fish (0.02-35 mg/kg) | 0.1-0.8 ppm in adipose tissue | Severe keratitis, Toxic nephritis, Higher incidence of miscarriages among exposed female workers |
| Trichloroethylene   | Degreasing agent, Chemical and pharmaceutical industry, Refrigerant | 320,000,000 lb (1991) | Groundwater (30 ppb), Surface water (1-24 ppb), Atmosphere (15-16 ppb), Chees (3 mg/kg), Vegetable oil (7 mg/kg), Fruit juices (5 mg/kg), Fish (1-1 ppm), Milk (0.3 mg/kg) | 1-32 ppb in mother's milk, 0.1-9 mg% in blood | Coma with eventual death from renal or hepatic failure, Headache, dizziness, and sleepiness, Skin irritation, CNS toxicities, Carcinogenicity |
| 2,4-Dichlorophenoxyacetic acid | Herbicide and other agriculture uses | Over 5,000 lb (1991) | Atmosphere and water near application or spill sites (concentration varies), Food (few mg/kg, except in the vicinity of spills), Fish (0.1 mg/kg) | 0.4-6.3 mg/kg body weight in urine of larynx care specialists using the herbicide | Coma, myotonia, fever, pulmonary emphysema, liver necrosis, degeneration of kidney tubule, death |

**Table 3. Effect of age on hepatic peroxisomal β-oxidation.**

| Species           | Young a,b | Old a,b | Reference |
|-------------------|-----------|---------|-----------|
| Male Fischer      | 100 (17)  | 117 (28) | (59)      |
| 344 rats          | 100 (8)   | 115 (20) | (54)      |
| Female CSWV mice  | 100 (11)  | 28 (20)  | (52)      |
| Female OFi mice   | 100 (11)  | 61 (24)  | (53)      |

*Rates are expressed as percent of specific activity in young animals within the same study. Numbers in parentheses indicate age of animals, in months, at time of sacrifice.*

**Table 4. Liver catalase in animals of various ages.**

| Species           | Young a,b | Old a,b | Reference |
|-------------------|-----------|---------|-----------|
| Male Fischer      | 100 (3)   | 83 (24) | (61)      |
| 344 rats          | 100 (5)   | 48 (26) | (62)      |
| Female CSWV mice  | 100 (6)   | 61 (24) | (64)      |
| Female OFi mice   | 100 (6)   | 48 (29) | (64)      |

*Rates are expressed as percent of specific activity in young animals within the same study. Numbers in parentheses are age of animals in months.*

---

CNS, central nervous system.

*Data extracted from a comprehensive search of the Hazardous Substances Data Bank (National Library of Medicine, Bethesda, MD).
mg/kg PFOA administration, hepatic peroxi-
somal β-oxidation increased in 100-week-old
rats to levels equaling approximately 60% of
those achieved in 10-week-old rats (Table 5).

In contrast to the PFOA findings, and in agree-
ment with results reported in the liter-
ature (54), young and old male Fischer 344
rats given various concentrations of the
peroxisome proliferator WY-14,643 in the diet
exhibited comparable hepatic peroxisomal β-
oxidation activity (Table 5). Similarly, a
study in male Wistar rats showed that age did
not seem to influence either basal or
nafenopin-induced induction of peroxisomal
β-oxidation (55). In these long-term feeding
studies (54, 55), animals were given diets
containing peroxisome proliferators for
22–59 weeks, which placed the age of
young and old rats between 68 and 116
weeks at the conclusion of the experiment.
Potentially, these are ages where responses
to peroxisome proliferators are similar. In
a more detailed study in our laboratories (66),
striking results showing compound-specific
effects were observed (Table 6). Although
aging-related differences appeared to exist
in the response of the liver to certain perso-
xisomal proliferators, aging did not seem to
influence the response to other peroxisome
proliferators studied (Table 6). The lack of
uniformity in the response to various perox-
osome proliferators may be due to potential
aging-related alterations in specific pathways

involved in the metabolism of specific pro-
liferators. In other words, peroxisomal
signal transduction pathways are most likely
altered in old animals, as evidenced by the
significant differences observed between
young and old animals in response to the
nonmetabolizable PFOA.

Mechanisms Involved in the
Aging-Related Decrease in
Peroxisomal Enzyme
Activities
Factors responsible for the reported decline
in peroxisomal enzyme activities and the
diminution of their response to peroxisome-
proliferating chemicals in aged animals are
unclear. A schematic depiction of a proposed
mechanism involved in this effect is present-
ed in Figure 2. The aged liver suffers from a
deffect in gene transcription and translation
of certain messengers (67). This defect may
be the result of deficiency in important
receptors and/or impaired signal transduc-
tion pathways. Recent findings show that
expression of nuclear thyroid hormone
receptor mRNA is reduced by 50% in the
livers of male Wistar rats 24 months old as
compared to their 6-month-old counterparts
(68). This decrease was not accompanied by
changes in the binding characteristics of the
receptors, although $B_{max}$ and $K_I$ decreased
by approximately 2-fold in the older rats.
Similar changes were also observed for the
mRNA of retinoic acid receptors (68). An
overlap between the gene networks regulated
by peroxisome proliferators and those for
retinoids and thyroid hormones has been
documented (57). PPARs, thyroid receptors,
and retinoid receptors (RXR) recognize the
same DNA response sequence and there is
cross-talk among these receptors (57, 69).
PPAR binds to peroxisome proliferator-
response elements as heterodimers with RXR
and activate gene transcription in response
to activators (70). Furthermore, RXR lig-
ands produce effects similar to those pro-
duced by PPAR ligands (70).

Peroxisome proliferators induce malic
enzyme gene transcription, which is a
known response to treatment with thyroid
hormones (68), through the action of
heterodimers of PPARγ and RXR (71).
In addition to changes in receptor abundance
and affinity, aging also causes modifications
in cellular transduction pathways, leading to
altered gene expression, which may be
responsible for some important impairment
in physiologic functions (67). The decrease
in receptor affinity in aged animals is
hypothesized to be due to changes in the
phosphorylation–dephosphorylation cycle in
these animals (68). The potential impact of
aging on the expression of the various perox-
osome proliferator-activated receptor isoforms
and/or binding of peroxisome-proliferating
chemicals to these receptors is shown by a
decline in the abundance of PPARγ mRNA
expression in the liver of aged rats (72).

Aging, Peroxisomes, and
Health
The importance of the peroxisome to animal
as well as to human life is evidenced by the
fact that their absence, or the dysfunc-
tionalit-
y of one or more peroxisomal enzymes, is
invariably fatal (73, 74). In addition, inhibit-
ing peroxisomal enzyme activities resulted in
the accelerated death of animals (75), which
suggests the existence of a linkage between
peroxisomal metabolism, aging, and death.
However, the exact nature of a potential
relationship is not known.

Inherited peroxisomal disorders.

Peroxisomal disorders are a group of inher-
ited metabolic diseases that are classified into
10 complementation groups (76). These
diseases involve multiple genes (76) and are
characterized by impairment of one or more
functions of peroxisomes (73, 74, 76). Recent
studies have suggested that at least 16 genes
may be involved in the observed abnormali-
ties in the assembly of peroxisomes (74, 77),
and the list of peroxisomal disorders con-
tinues to grow (76). The combined incidence
for the occurrence of these diseases is esti-
mated at 1 in 20,000 or greater (74). The
cerebrohepatorenal syndrome known as
Zellweger syndrome was the first peroxi-
osomal disorder identified (74, 78). This
syndrome is characterized by the absence of
morphologically distinguishable peroxisomes
(74). In contrast to Zellweger syndrome,
patients suffering from X-adrenoleukodys-
trophy have peroxisomes of normal number
and morphology, and the disease is caused
by a single enzymatic abnormality; ligo-
oceroyl-CoA ligase is suggested as the missing
enzyme (74, 79). Other diseases such as
rhizomelic chondrodysplasia are recognized
as multiple peroxisomal enzymatic deficien-
cies, although they have intact peroxisome mor-
phology, normal size, and number (74, 80).

Aging, apoptosis, and response to peroxi-
some proliferators. Aged animals are more
susceptible to the hepatocarcinogenic effect
of peroxisome proliferators (55, 59). Long-
term treatment with either of the peroxi-
some-proliferating chemicals nafenopin or
WY-14,643 produced numerous hepatocel-
lar adenomas and carcinomas in old male
Wistar and Fischer 344 rats while producing
only a few changes in young animals
(55, 59). Because the enhanced susceptibility
of old rats did not appear to result from
enhanced hepatic oxidative damage, it was
hypothesized that peroxisome proliferators
may promote spontaneously occurring pre-
neoplastic foci. These foci are more abundant

---

Table 5. Induction of liver peroxisomal β-oxidation by various peroxisome proliferators in aged animals.

| Species            | Young   | Old       | Reference |
|--------------------|---------|-----------|-----------|
| Male Fischer       | 100 (3' | 44 (25)   | (67)      |
| 344 rats           | 100 (2')| 68 (13')  | (60)      |
| Female Ofr mice    | 100 (8')| 85 (20)   | (54)      |

*Results are expressed as percent of peroxisome proliferator-induced specific activity in young animals within the same study. Numbers in parentheses are age of animals in weeks. Animals were given perfluorooctanoic acid (150 mg/kg, orally in corn oil 48 hr prior to sacrifice). Animals were given clofibrate in the diet (various concentrations to correspond to 200 mg/kg/day for 14 days). Animals received WY-14,643 in the diet (0.1% for 22 weeks). Animals received clofibrate-supplemented diet (0.5% for 5 days).

Table 6. Age-related responses of hepatic peroxisomal β-oxidation to various peroxisome proliferators.

| Compound          | Young (10 weeks old) | Old (100 weeks old) |
|-------------------|----------------------|---------------------|
| PFOA              | 100%                 | 62%                 |
| WY-14,643         | 100%                 | 93%                 |
| DEHP              | 100%                 | 137%                |

Abbreviations: DEHP, diethylhexylphthalate; PFOA, perfluorooctanoic acid.

*Results are expressed as percent of hepatic enzyme specific activity in young rats. **150 mg/kg, *250 mg/kg.
†2.2 g/kg. Drugs in corn oil were given orally 48 hr prior to sacrifice.
‡Significantly different from corresponding young animal group.
in the livers of aged animals as a natural consequence of aging (59). Hypolipidemic drugs are preferentially prescribed to older individuals who may have high levels of preneoplastic hepatocytes. Promotion of these cells by peroxisome proliferators has the potential to result in liver cancer.

As an alternative mechanism responsible for the hepatocarcinogenic effect of peroxisome proliferators, several studies have shown that peroxisome proliferators suppress apoptosis in the liver in a process mediated by PPARα (81,82). Apoptosis appears to be a safeguard to prevent cells with DNA damage from progressing to a tumor (83). In primary cultures of rat hepatocytes and in an FaO rat hepatoma cell line, apoptosis was induced by transforming growth factor β1, an effect that was significantly reduced by the coadministration of the peroxisome proliferator nafenopin (84). Furthermore, nafenopin suppressed hepatic apoptosis in vivo and its withdrawal resulted in a 100- to 200-fold elevation in apoptosis (84). Hepatocytes generated during nafenopin-induced hyperplasia were not the same as those that underwent apoptosis upon nafenopin withdrawal (84). Hepatocytes resistant to apoptotic death may represent preferential targets for promotion by peroxisome proliferators; thus, suppression of apoptosis may play a role in the hepatocarcinogenicity of this class of nongenotoxic compounds. Current evidence indicates that the signal transduction pathways for apoptosis and the cell cycle overlap (85). Senescent cells are resistant to both proliferative and apoptotic stimuli, possibly because of their failure to induce the expression of G1 genes, which are required for both outcomes, when subjected to stimulation (85). An alternative hypothesis to explain the resistance of senescent cells to apoptosis stipulates that these cells have the protein terminin predominantly in its 60-Kda form, in contrast to the 90 Kda in apoptotic-prone cells (85). Resistance to apoptosis may contribute to the enhanced vulnerability of the aged liver to the hepatocarcinogenic effect of peroxisome proliferators.

**Conclusion**

The importance of peroxisomes to normal cell functions and cellular survival makes the investigation of whether these organelles play a role in aging and aging-related diseases an urgent task. Research in the field of peroxisomes has until recently focused on these organelles and their proliferation, particularly in the rodent liver. Because peroxisomes do not seem to proliferate to significant levels in the livers of humans exposed to peroxisome-proliferating chemicals, pre-mature conclusions were drawn that dismissed the potential risk these chemicals pose to humans. However, recent advances show that signal transduction pathways linked to peroxisomes are involved in a myriad of extraperoxisomal effects. Examples of these effects include adipocyte differentiation, regulation of glucose homeostasis, inhibition of macrophage and monocyte activation, and inhibition of angiogenesis (42,43,86,87). Although several of these effects appear beneficial, others may potentially be harmful. Therefore, it is imperative that a concerted and comprehensive effort is channeled toward investigating a wide array of potential effects due to peroxisome-proliferating chemicals. A significant portion of this effort should be directed toward the elderly population, which might be more vulnerable to these effects.

Membrane structure, fluidity, and permeability to ions undergo some age-dependent alterations (53,88,89). These alterations have serious inhibitory effects on the enzymatic catalysis rates involved in protein

---

**Figure 2. Schematic depiction of the effect of aging on proposed signaling pathways involved in maintaining and proliferating hepatic peroxisomes.** Abbreviations: GR, glucocorticoid receptor; PPAR, peroxisome proliferator-activated receptor; RXR, retinoic acid receptor; TR, thyroid hormone receptor. Aging is associated with a decline in GR, TR, and RXR. The compromised integrity of these systems, along with a potential decline in PPAR expression and/or activation with aging may be responsible for the diminished basal levels of peroxisomal enzyme activities and their response to peroxisome proliferators in aged animals.
synthesis that may be responsible for the decline in protein synthetic capacity in aged cells and may also contribute to the onset of age-related diseases (53,88,89). For example, membrane fatty acid composition affects membrane structure and function (90). Membrane fatty acid composition is maintained through a balanced process of synthesis and degradation. Although degradation of long-chain fatty acids occurs in both mitochondria and peroxisomes, very long-chain fatty acids are metabolized exclusively in peroxisomes (91). Thus, peroxisomes appear to play an important role in the maintenance of membranes and membrane function by maintaining fatty acid balance. Consequently, the decline in peroxisomal catabolism of fatty acids with aging may represent a potential mechanism by which these organelles are involved in the process of aging.

An aging-related decline in peroxisomal protein synthesis, among other proteins, is expected to diminish peroxisomal capacity to metabolize very long-chain fatty acids. This effect may be manifested in an altered composition of the structure of cellular and organelle membranes. For example, it has been reported that aging causes an increase in the cholesterol:phospholipid ratio in rat liver microsomal, mitochondrial, and cellular membranes (88,90). Phospholipids are essential for the activation of various signal transduction pathways. This includes the modulation of protein kinase C activity by phophatidylserine and various other membrane phospholipids (92). Accordingly, changes in membrane phospholipids as a result of age-related diminished peroxisomal metabolism may trigger a cascade of harmful events and/or interrupt normal important cellular functions.

Aging-associated changes in membrane phospholipid composition are also reflected by an increase in Na, K-ATPase activity, and intracellular potassium content (88). Increased intracellular ion concentrations are postulated to reduce the activity of the whole translational machinery and increase aggregation of macromolecules, which retard all enzymatic catalysis (88,93) and interfere with mitotic activity (94).

The impact of aging on extracellular matrix, growth factors, cytokines, and non-parenchymal cells, among other factors, will be of utmost importance to our understanding of the relationship between aging and peroxisomal metabolism. Because these factors play important roles in liver cell division (95,96), their alterations with aging may explain the potential interrelationship between peroxisomal metabolism and aging-related disease and death. Peroxisomes grow by the posttranslational incorporation of new content and membrane proteins into preexisting peroxisomes, which then divide to form daughter peroxisomes in a dynamic fission process (16). Consequently, a vicious cycle involving hepatocellular senescence and a decline in peroxisomal metabolism may ensue, leading to the demise of the liver and ultimately to death.

Peroxisome-proliferating chemicals produce a myriad of extraperoxisomal effects in the liver and other tissues of experimental animals (24). Such effects include metabolic as well as hormonal alterations, in addition to effects on ion homeostasis and interference with metabolism and effects of other chemicals (24). These extraperoxisomal effects may be directly or indirectly related to the effects on peroxisomes or may be totally independent of these events. Whether any or all of these effects occur in humans exposed to peroxisome proliferators is not known. Focusing only on the peroxisomal effects of these chemicals may grossly underestimate the harmful potential of peroxisome proliferators to humans, especially the elderly, who are more susceptible to toxic insults. Furthermore, because of the essential roles played by peroxisomal metabolism, the possibility that modulation of peroxisomal functions may be a contributing factor to the process of aging should not be ignored.

References and Notes

1. Dice JF. Cellular and molecular mechanisms of aging. Phys Rev 72:143-159 (1953).
2. Cutter RG. Longevity is determined by specific genes. In: Testing the Theories of Aging (Adelman RC, Roth GS, eds). Boca Raton, FL: CRC Press, 1982:25-114.
3. Rubin H. Cell aging in vivo and in vitro. Mech Ageing Dev 98:1-35 (1997).
4. Makrides S. Protein synthesis and degradation during aging and senescence. FEBS Lett 343:422 (1993).
5. Dice JF. Cellular theories of aging as related to the liver. Hepatology 5:508-513 (1985).
6. Tauchi H, Sat0 T. Hepatic cells of the aged. In: Liver and Aging (Kita R, ed). Amsterdam: Elsevier, 1976:19-19.
7. Popper H. Aging and the liver. Prog Liver Dis 8:659-683 (1986).
8. Bucher NLR. Regeneration of mammalian liver. Int Rev Cytol 15:245-300 (1963).
9. Schaprio H, Hotta SS, Dutton WE, Klein AW. The effect of aging on rat liver regeneration. Experientia 36:1075-1076 (1980).
10. Wilson P, Fratse. The effect of age on mitochondrial ultrastructure. Gerontology 21:84-19 (1975).
11. Herberg GH. A morphometric study of age dependent changes in mitochondrial populations of mouse liver and heart. J Gerontol 31:12-12 (1976).
12. Weindruch RH, Cheung MK, Verity MA, Walford RL. Modification of mitochondrial respiration by aging and dietary restriction. Mech Ageing Dev 12:375-392 (1980).
13. Schmucker DL, Wang MM. Age-related changes in liver drug metabolism: structure versus function. Proc Soc Exp Biol Med 165:178-187 (1980).
14. Birnbaum LS. Hepatic drug metabolism in senescence. Exp Gerontol 18:263-267 (1980).
15. Rikans LE, Moore DR. Effect of age and sex on allyl alcohol hepatotoxicity in rats: role of liver alcohol and aldehyde dehydrogenase activities. J Pharmacol Exp Ther 220:23-20 (1957).
16. Lazarow P, Fujiki Y. Biogenesis of peroxisomes. Annu Rev Cell Biol 1:489-530 (1985).
17. Bentley P, Elcombe C, Grasso P, Stringer D, Wiegand H. Hepatic peroxisome proliferation in rodents and its significance for humans. Food Chem Toxicol 31:857-907 (1993).
18. Staubli W, Feng L, Weibel E. Correlated morphometric and biochemical studies in rat hepatocytes. J Cell Biol 42:52-112 (1969).
19. Weibel E, Staubli W, Gangi H, Hess F. Correlated morphometric and biochemical studies on the liver cell. I. Morphometric model, stereometric methods, and normal morphometric data for rat liver. J Cell Biol 42:86-119 (1969).
20. del Rio LA, Sandalio LM, Palma JM. A new cellular function for peroxisomes related to oxygen free radicals. Experientia 46:909-922 (1990).
21. Tolbert N. Metabolic pathways in peroxisomes and glyoxysomes. Annu Rev Biochem 50:133-157 (1981).
22. Van den Bosch H, Schuengens RBH, Wanders RJAJ, Tager JM. Biochemistry of peroxisomes. Annu Rev Biochem 61:157-192 (1992).
23. Paget G. Experimental studies of the toxicity of atorganic with particular reference to line structural changes in the rat. Proc Soc Exp Biol Med 31:729-736 (1963).
24. Youssef J, Badr M. Extraperoxisomal targets of peroxisome proliferators: mitochondrial, microsomal, and cytoxic effects: implication for health and disease. CRC Rev Toxicol 21:1-33 (1990).
25. De La Iglesia F, Lewis J, Buchanan R, Marcus E, McMahon G. Light and electron microscopy of liver in hyperlipoproteinemic patients under long-term gemfibrozil treatment. Atherosclerosis 23:9-21 (1982).
26. Easocho P, Fowenby P, Johnson W, Hoyer D, White S. Hepatic peroxisomal changes induced by a tetratole-substituted alkyloxycyclohexane in rats and comparison with other species. Toxicol Appl Pharmacol 63:430-437 (1983).
27. Dech P, Schmitt L, Steinberg P, Thomas H. Concomitant induction of cytosolic epoxide hydrolase and peroxisomal B-oxidation by hypolipemic compounds in rat and guinea pig liver. Arch Toxicol 12:suppl:248-255 (1988).
28. Lake B, Evans J, Gray T, Koren S, Netof C. Comparative studies on naifen-inhibited hepatic peroxisome proliferation in the rat, Syrian hamster, guinea pig and mouse. Toxicol Appl Pharmacol 89:146-160 (1988).
29. Watanabe T, Horie S, Yamada J, Imai S, Nishigaki T, Naito J, Sugita T. Species differences in the effect of bezafibrate, a hypolipemic agent, on hepatic peroxisome-associated enzymes. Biochem Pharmacol 30:367-371 (1989).
30. Sakuma M, Yamada J, Sugita T. Comparison of the inducing effect of dehydroxyandrosterone on hepatic peroxisome proliferation-associated enzymes in several rodent species. Biochem Pharmacol 43:1209-1213 (1992).
31. Horie S, Ishii H, Sugita T. Changes in peroxisomal fatty acid oxidation in the diabetic rat liver. J Biochem 90:1991-1996 (1981).
32. Thomas H, Schultz L, Knebe M, Dech P. Effect of diabetes and starvation on the activity of rat liver epoxide hydrolase, glutathione S-transferase and peroxisomal B-oxidation. Biochem Pharmacol 38:4231-4237 (1989).
33. Fringes B, Reith A. Titania biogenesis during adaptation to mild hyperthyroidism in rat liver: a morphometric/stereologic study by electron microscopy. Lab Invest 47:19-26 (1982).
34. Reith A, Horie S, Yamada J. Peroxisome biogenesis in rat liver during adaptation to mild hyperthyroidism: a morphometric/stereologic study by electron microscopy. Am J Anat 200:514-516 (1982).
35. Levy E, Weiss PM. Hepatic B-oxidation produced by a single dose of endotoxin in the mouse. Am J Pathol 52:477-502 (1968).
36. Neat C, Thomasen M, Osmundsen H. Induction of peroxisomal B-oxidation in rat liver by high fat diet. Biochemistry 180:369-371 (1980).
37. Rigas JS, Legg PG, Wood RL. Microbody formation in regenerating rat liver. J Histochem Cytochem 18:893-900 (1970).
38. Laszlo A, Tripathi B., Thiesbock J, Stasium G, Di Lallo AM, Di Lallo M, De Lisio L. The mechanisms of action of steroid/thyroid receptor family members. Annu Rev Biochem 63:431-466 (1994).
39. Gottlicher M, Widmark E, L J, Kulthong J, J A. Fatty acids activate a cholesterol c GOTFIC acid-activated receptor and the glucocorticoid receptor. Proc Natl Acad Sci USA 89:4653-4657 (1992).
Environmental Health Perspectives • Volume 107, Number 10, October 1999

Reviews • Aging and hepatic metabolism

Mangelsdorf D, Umesono K, Evans R. Differential expression and activation of a family of murine peroxisome proliferator-activated receptors. Proc Natl Acad Sci USA 91:7344–7348 (1994).

42. Tontonoz P, Singer S, Forman B, Sarral P, Fletcher J, Fletcher C, Brun R, Mueller E, Atok S, Oppenheim H, et al. Terminal differentiation of human adipocytes cells induced by ligands for peroxisome proliferator-activated receptors α and the retinoid X receptor. Proc Natl Acad Sci USA 94:237–241 (1997).

43. Berger J, Laliberte M, Deobald T, Elbrecht A, Zhang B, Zhou B, Gwias B, Cullinan C, Hayes N, Li Y, et al. Novel peroxisome proliferator-activated receptor (PPARs) and PPAR ligands produce distinct biological effects. J Biol Chem 274:6181–6185 (1999).

44. Devchand P, Keller H, Peters J, Vazquez M, Gonzalez F, Wahl W. The PPARs- leukotriene B4 pathway to inflammation control. Nature 343:49–53 (1996).

45. Forman B, Chen J, Evans R. Hypolipidemic drugs, polyunsaturated fatty acids and eicosanoids are ligands for peroxisome proliferator-activated receptors α and δ. Proc Natl Acad Sci USA 94:4312–4317 (1997).

46. Wolf G. Fatty acids bind to and activate peroxisome proliferator-activated receptors and γ. Nutr Rev 56:61–63 (1998).

47. Gustafsson J-A. Fatty acids in control of gene expression. Nutr Rev 56:S20–S21 (1998).

48. Sher T, Yi H, McBride D, Gonzalez F. DNA cloning, chromosomal mapping, and functional characterization of the human peroxisome proliferator activated receptor. Biochemistry 32:5598–5604 (1993).

49. Mukherjee R, Jow L, Noonan D, McDonnell H. Human peroxisome proliferator activated receptor δ and γ and an activated receptor (PPARs) demonstrate similar tissue distribution but different responsiveness to PPARs. J Sterol Mol Biol 51:151–166 (1994).

50. Perichon R, Bourre JM. Peroxialosomal β-oxidation activity and catalase activity during development and aging in mouse liver. Biochimie 77:288–293 (1995).

51. Perichon R, Bourre JM. Age-related decrease in liver peroxisomal fatty acid oxidation in control and clorbrate-treated mice. A biochemical study and mechanistic approach. Mech Ageing Dev 97:115–126 (1998).

52. Carasso RC, Mangold DR, Kopp JA. Age-related susceptibility to the carcinogenic effects of the peroxisome proliferator WY-14,643 in rat liver. Carcinogenesis 12:469–473 (1991).

53. Huber W, Kraupp-Grasl B, Estebauer H, Schultze-Hermann R. Role of oxidative stress in age-dependent hepatocarcinogenesis by the peroxisome proliferator nafenof in the rat. Cancer Res 51:1789–1792 (1991).

54. Beier K, Voit K, Fahimi H. The impact of aging on enzyme proteins of rat liver peroxisomes-quantitative analysis by immunoblotting and immunoelectron microscopy. Virchows Arch B 63:139–146 (1993).

55. Masters C, Crane D. On the role of the peroxisome in ontogeny, aging and degenerative disease. Mech Ageing Dev 80:89–93 (1995).

56. Soliman M, Cunningham M, Morrow J, Roberts J, Badr M. Evidence against peroxisome proliferated-induced hepatic oxidative damage: levels of esterified isoprostanes in liver-fed mice a diet containing 4-chloro-6(2,4)xyldino)-2-