DERLEME

REVIEW

Alkylphenols in the Environment and Their Adverse Effects on Living Organisms

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Anahtar Kelimeler
Alkilfenol
Çevre kirliliği
Canlı organizma
Karsinojenik
Endokrin sistem bozucusu

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SUMMARY

The compounds of alkylphenolpolyethoxylates (APEs), one of the ubiquitous estrogenic environmental endocrine disrupters, are widely used as non-ionic surfactants or antioxidants in detergents, pesticides, herbicides, emulsifiers, paints, cosmetics, plasticwares and even in jet fuel. It has been reported that APEs and their derivatives called alkylphenols (APs) exert adverse effects on both aquatic and terrestrial organism. Moreover, they have been shown to have toxic, estrogenic and carcinogenic effects. Therefore, the occurrence of APEs and their derivatives in the environment as well as their structures, biodegradation, bioaccumulation, metabolic fate and their adverse effects on living organisms including human were reviewed from a complex literature to understand their potential danger to human being. From this review of literature, it is concluded that the pollution level of APEs or their derivatives in the environment are not lethal to the most organisms. However, the sub-lethal levels of these substances exert estrogenic and carcinogenic effects in almost all organisms including human being.

Çevredeki Alkifenoller ve Canlı Organizmalara Olan Zararlı Etkileri

ÖZET

Alkilfenol bileşikleri (AFEO) iyonic olmayan yüzey aktif maddesi olarak deterjanlarda, ot ve böcek ilaçlarında, kozmeiklerde, plastik eşyalarda emülsifikatörlerde, boyalarda ve hatta uçak yakıtlarında çok yaygın kullanılan östrojenik endokrin sistem bozucularıdır. Alkilfenol adı verilen AFEO bilişiklerinin türevlerinin hem suda hem de karada yaşayan canlılara zararlı etkileri olduğu rapor edilmiştir. Ayrıca, bunların hem östrojenik, hem toksik hem de karinojenik etkileri olduğu ortaya konmuştur. Bu nedenle, AFEO bileşikleriri ve türevlerinin doğadaki varoluşları, yapıları, biyolojik bozunumları, biyolojik birikimleri, metbolize edilme yolları ve insan dahil bütün canlı organizmalara olan zararlı etkileri karmaşık literatür taranarak potnsiyel zararlı etkileri anlaşılmaya çalışılmıştır. Bu literatür taraması ile doğada bulunan AFEO bileşikleri ve türevlerinin miktarlarının öldürücü dozlarda olmadığı anlaşılmıştır. Fakat, bu bileşiklerin ve türevlerinin öldürücü olmayan dozlarının da insan dahil bütün canlı organizmalarda hem östrojenik hem de karsinojenik etkilerinin olduğu ortaya konmuştur.

INTRODUCTION

A number of man-made and naturally occurring compounds in natural environment have been shown to have hormone like activity and these types of compounds are called as environmental endocrine disrupters.[1] Continuous exposure to high levels of these compounds led to the occurrence of acute and chronic health problems in humans and in wildlife.[2, 3] Adverse effects of environmental endocrine disrupters are originating from their ability to mimic or antagonize the effects of endogenous hormones and thus, they disrupt the metabolism or the synthesis of endogenous hormone receptors in an organism. Majority of environmental endocrine disrupters such as diethylstilboestrol (DES), DDT, polychlorinated biphenyls (PCB), and alkylphenolpolyethoxylates (APEs) are shown to have estrogenic effects in both aquatic and terrestrial organisms.[4]

Considerable evidence has been accumulating that exposure to the ambient levels of estrogenic compounds in the environment may have adverse effects on the development of gonads and the fertility of animals and human being.[2,5,6] It has been widely recognized that "human milk is the most important form of nourishment for newborn children". Ademollo et al[7]. reported that the presence of nonylphenol and octylphenol in human breast milk is a potential risk for the growth and development of children. José G. Dórea[8] also reported that nonylphenol and octylphenol is contaminating not only human breast milk but also cow milk as well. This is a very important for risk assessment of these endocrine-disruptor substances (EDS).

DES is one of the widely known environmental estrogens that their adverse effects in both animals and humans are well documented.[9,10] Although the general use of DES is prohibited, it is still used for the treatment of prostate cancer, birth control and also used as cattle feeds for growth promoter. Organochlorine pesticides such as PCBs and DDT are another commonly known groups of environmental estrogens shown to accumulate in breast milk and adipose tissues of human.[7,11,12] Recent findings have indicated that APEs especially NP is ubiquitously found in terrestrial food including fresh fruits and vegetables[13,14], human milk[7,15], in aquatic as well as livestock products and in rice.[16] The pathway for nonylphenol contamination to food occurs due to the use of cleaning agents in the food processing industries as well as the use of pesticides and herbicides.[18]

There has also been converging evidence that exposure to environmental estrogens may disrupt reproductive and endocrine function in fish, birds and mammals, especially during development.[18,19]

As mentioned above, one of the most ubiquitous estrogenic environmental endocrine disrupters known as the compounds of alkylphenolpolyethoxylates (APEs) that have been widely used as non-ionic surfactants or antioxidants in detergents, pesticides, herbicides, emulsifiers, paints, cosmetics and plasticware[20]. Besides their estrogenic effects[21,22], APEs have also been shown to have carcinogenic[23,24,25] and toxic effects[26,27,28,29] in both aquatic and terrestrial organisms.

Nonylphenol (NP), one of the most abundant derivatives of APEs, has been demonstrated to stay biologically active state for a longer period of time in the body than that of natural estrogen.[30] It seems that the major source of APEs in Human occurs through diet. In a diet survey in Germany, NPs was found in all the investigated food samples with concentrations ranging from 0.1 up to 19.4 μg/kg[13] and 5.8 to 235.8 μg/kg were determined in Taiwan foodstuff samples.[16]

Therefore, the occurrence of APEs and their derivatives in the environment as well as their structures, biodegradation, bioaccumulation and metabolic fate in living organisms and their adverse effects on living organism including human were reviewed in this review paper from a complex literature to understand their potential danger at cellular and molecular level.

2. Structure and biodegradation of APEs and APs

The surfactant activity of APEs derived from an alkylphenol hydrophobe and a para-substituted long chain of repeating ethylene oxide units as the hydrophilic moiety (Figure 1).

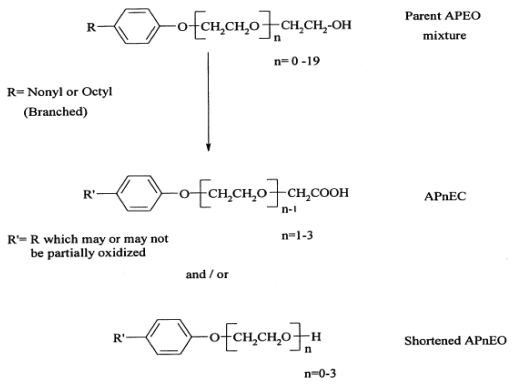

Figure 1. Structure of alkylphenol polyethoxylates[30]
Şekil 1. Alkilfenol polietoksilatların yapısı

The longer the chain, the more soluble the compound is, therefore, repeating units of ethylene oxide may extend from one to 100 repeating units in the alkylphenolic compounds and their alkyl groups are typically a branched nonyl, octyl, and butyl chain.[1]

The main biodegradation of APEs is the hydrolytic removal of ethoxylate groups to give more lipophilic degradation products with estrogenic potential such as nonylphenol (NP), octylphenol (OP) and butylphenol (BP) (Figure 2).

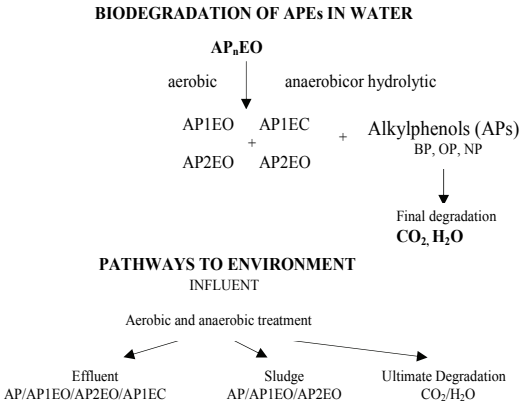

Figure 2. Primary and final biodegradation of APEs and environmental fate[1]
Şekil 2. APE'nin primer ve son biyodegrasyonu ve degradasyonu ve çevresel akıbeti[1]

This step is relatively fast and resulted to produce the degradation intermediates. For example, if the alkyl group in the APEs is nonylphenol, the intermediates are nonylphenol ethoxylates (NP_nEO).[31] Biodegradation of NP occurs more slowly since there is a benzene ring in the compound.[32] The UK Environmental Agency estimated that the biodegradation half-life of NP is about 150 days in water.[33] Furthermore, recent studies have identified that nonylphenol is the most critical metabolite among APs because of its enhanced resistance towards biodegradation, severe toxicity, high lipophilicity, less water solubility and its ability to bioaccumulate in aquatic organisms more than that of other APs.[34, 35, 36, 37, 38]

Nonylphenol polyethoxylates (NPE_n where n=3 to more than 20 ethoxylates units) posses the properties of good wetting detergency, low foaming, applicability at low temperatures and low cost.[39] Because of environmental concern, their use in the household detergent formulations has been voluntarily stopped in North America. However, NPE_n are now primarily used in the textile, the pulp and paper industries. Also, up to 1 kilo tons of NPE_n is annually used in the production of pesticides as the emulsification agents.[40]

Octylphenol is the second largest group of alkylphenolic compounds present in the natural environment.[1] The compound 4-*tert*-octylphenol (4-*t*-OP) is a degradation product of octylphenol polyethoxylates surfactants and may also be present as a minor component in nonylphenol polyethoxylates preparations.[39]

As shown in Figure 3, alkylphenolic compounds may also be microbially carboxylated to give APECs with carboxylic acid functional groups at the terminal end of the ethoxylate chain.[41]

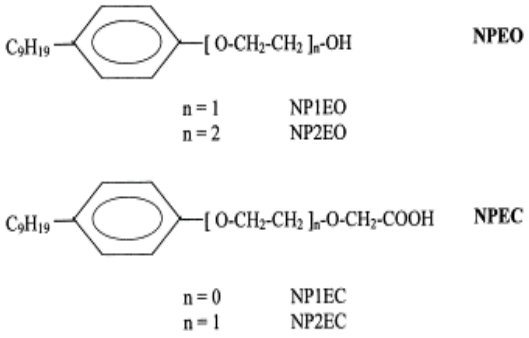

Figure 3. Carboxylated structure of nonylphenol ethoxylates.
Şekil 3. Karoksillenmiş nonilfenol etokislat yapısı

There are a few reports about the presence of alkylphenol polyethoxylates in the natural environment. However, there are many reports about the concentrations of the biodegradation products of alkylphenols (APs) such as NP or OP in sewage effluents, river waters and sediment and these derivatives are more stable for degradation than that of APEs.

Hawerlak et al.[42] showed that APEs concentrations in the field of farms declined by 84 % over a period of 14 weeks. However, these changes in the chromatographic patterns of acetylated 4-nonylphenol indicates that there is a group of recalcitrant nonylphenol isomers, which are degraded more slowly than the other isomers.

In addition to hydrolytic and microbial degradation, Ahel et al.[43] reported that NP is susceptible to photochemical and temperature dependent breakdown. For example, it has been reported that the rate of APEs biodegradation is fluctuating seasonally due to temperature changes. Marcomini et al.[44] reported that temperature over 20°C is causing rapid degradation of APEs. It has also been discovered that NP has a half-life of approximately 10-15 h under continuous summer sun in the surface water layer and the rate of breakdown decreases 1.5 times at the depths of 20-25 cm.[33]

3. Production and environmental distribution of APEs and APs

The APEs are nonionic surfactants that constitute 6% of the total surfactant production and 25% of the total non-ionic surfactant production in the

USA. Talmage[45] reported that 55% of the total APEs production is used for industrial application such as plastics and elastomers, textiles, agricultural chemicals and paper product, while 30 % of them are used for institutional cleaning products. Remaining 15 % of the total APEs are used for house-hold cleaning and personal cleaning products in the world.[45]

As mentioned above, APEs are commonly used chemicals with a wide variety of commercial and domestic applications consisting industrial detergents, paint ingredients, wetting agents, emulsifiers, and a number of miscellaneous applications including use in cosmetics or cleaning agents or as a spermicide such as nonoxynol-9[20,46,47].

The annual production of APEs in the world was estimated to exceed the amount of 500,000 (180,000 tons alone in the USA) metric tons.[48,49] It has been estimated that 60 % of the total production has been released into the rivers, lakes and estuaries around the world[82]. Due to their high lipophilicity, APs have been reported not to accumulate in the water. Instead, they accumulate in sewage sludge and sediments and bioaccumulate in aquatic biota.[36,37] Although at "hot spots" like in sewage effluents, APEs concentrations have been reported to reach 1000µg/L, APs concentrations in environmental waters are rarely found to exceed the amount of 10µg/L.[31,38,50,51]

The main ports of APEs entry into the natural environment are sewage treatment plants and industrial sources such as pulp mills and wool scouring.[45] NP has also been shown to leach from PVC tubing during milk processing[52] and from plastics used in food packaging.[53] Hale et al.[54] reported that the concentration of APEs in sediment of rivers in North America is reached up to 70 mg/kg-sediment.

Concentrations of alkylphenolic compounds in the environment are reasonably well documented but they are geographically variable.[55] Domestic sewage effluents can contain alkylphenolic compounds up to 100 µg/l, whereas alkylphenolic compounds originating from pulp mills and textile industries can contain mg/L quantities.[36,41] In the UK, for example, industrial effluents may contain concentrations of NP that may exceed 1000 µg/l.[38]

APEs and APs pollution levels have been reported in many countries in Europe and in North America. For instance, reported that the concentration of NP, the most abundant APs in natural environment, has been ranging from 0.3 to 0.45 µg/l in Swiss Rivers[34] reaching up to 336 in British Rivers and µg/l.[50] The maximum APEs concentration has so far been reported by Hale et al.[54] that NP reached 14 mg /l in sewage effluent in the rivers of the USA. Also, Blackburn et al.[51] reported that nonylphenol level reached up to 15µg/g-sediment in British Rivers. However, maximum APs pollution in natural environment was reported in North America. O'Halloran et al.[56] reported that NPs concentrations were ranging 3-300µg/L in Canadian Rivers and the highest NP detected so far was 6,300 µg/L in sewage effluent and 70,000 µg/kg-sediment in the USA Rivers.[54]

As mentioned above, nonylphenol ethoxylates (NPEs) are the most common of the APEs, constituting 82% of the total production, and are also referred to as nonoxynol, ethanol nonylphenol, polyoxyethylene nonylphenol ether or nonylphenoxy poly (ethyleneoxy) ethanol, while octylphenol ethoxylates constitute for 15 to 18% of the total APEs.[1]

4. Bioaccumulation APs and APEs

Bioaccumulation of many estrogenic agents has aroused considerable concern, especially those that are lipophilic and have long half-lives. NP is a lipophilic compound with octanol:water partition coefficient (K_{ow} 4.48), while NP1EO, NP2EO and NP3EO are slightly less lipophilic with log of K_{ow}s 4.2[35]. These kinds of agents might be accumulated over time until reaching a final critical level that is capable of eliciting an effect.[57]

It has been reported that algae, fish, and ducks, as well as marine organisms, have all been bioaccumulating NP, NP1EO, and NP2EO from fresh water environment. Lewis and Lech[58] have demonstrated that [^{14}C]4-nonylphenol was uptaken from water by rainbow trout and stored in muscle and liver tissues with a 40-100 bioaccumulation factors. Blackburn et al.[51] reported that nonylphenol concentrations measured in fish were 50 fold higher than those measured in water. It has also been shown that NP is bioaccumulating more in algae than in other higher organisms.[59]

5. Metabolic fate of APEs and APs

In order to understand the mechanism of the effects of alkylphenol in an organism, it is crucial to understand the metabolic fate of alkylphenols in the organisms.

Previous studies on metabolism and the disposition of radio labeled NP in an organism revealed that the metabolites of NP are present in both bile and urine after water exposure[58] and dietary[60,61,62]. NP-derived radioactivity in fish exposed through water was reported to be more evenly distributed in the organs than that of the intragastric exposure and were observed in the intestinal contents, liver, kidney, gills, skin, abdominal fat and brain.[63] The excretion of alkylphenols in salmon mainly occurs through urine but not in trout. These differences may be attributed to the differences in the living conditions of these fishes since trout is a fresh water fish while

salmon is an anadromous fish that mainly live in the sea.[64]

Alkylphenols were hydroxylated by cytochrome P450 monooxygenase enzymes (CYP) at C-8 and C-9 positions of the alkyl chain to give catechols or conjugated catechol structure namely (8- or 9-hydroxy alkylphenol).[64,65] Hydoxylation of the alkylphenols by CYP enzymes is the phase I of the metabolic pathway in alkylphenol metabolism (Figure 4). Then, the glucouronidation of the alkylpehnols occurs by ß-glucouronidase enzyme. Thibaut et al.[60] reported that 4-nonylphenol is modified by ß-glucuronidase hydrolysis.

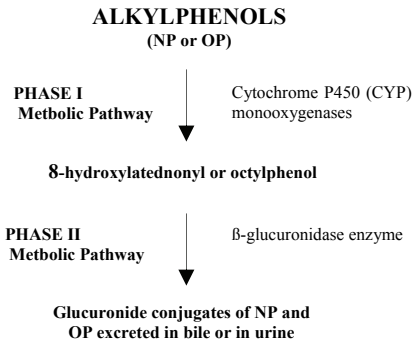

Figure 4. Metabolism and excretion of alkylphenols in the liver of rainbow trout, Atlantic salmon (adapted from Arukwe[64]) and rat (Pedersen and Hill[65]).

Şekil 4. Gök kuşağı alabalığı, Atlantik somon balıkalrının (Arukwe[64]'ten uyarlandı) ve ratların karaciğerinde nonilfenolün metabolizması ve dışarı atılımı (Pedersen and Hill[65]).

Therefore, most of the 4-nonylphenol metabolites are found as glucuronic conjugates in the organisms. Nonylphenol is hydroxylated at both omega-1 and omega-2 positions of the alkyl chain, giving 9-hydroxynonylphenol and 8-hydroxynonylphenol. 9-hydroxynonylphenol is oxidized and then subsequently beta-oxidized to yield 7-(4-hydroxyphenyl) heptanoic acid, 5-(4-hydoxyphenyl) pentonoic acid, 3(4-hydoxypehnyl) propionic acid and 3-(4-hydroxyphenyl)-2-propenoic acid. Predominant metabolites of 4-nonylphenol and 4-*tert*-octyphenol were found in bile as a glucouronid conjugate.[66,67] Other metabolites include glucouronid conjugates of ring or side chain of hydroxylated 4-nonylphenol. Despite the relatively rapid metabolism and excretion, a substantial amount of the parent compound remains in the muscle indicating the maintenance of 4-nonylphenol residues and associated biological activity in the organisms.[66]

6. Adverse effects of APEs and alkylphenols on living organisms

As mentioned above, alkylphenolic compounds have been shown to have estrogenic,[68] carcinogenic[69] and toxic effects[70] in both aquatic and terrestrial organisms. Therefore, it is necessary to focus on the mechanism of APEs effects on different organisms.

6.1. Estrogenic Effects

Endocrine disrupters especially estrogenic environmental disrupters have received great media attention due to their possible contribution to the reported declines in human sperm count over the past 50 years. Exposure of environmental estrogen may also be linked to breast cancer in women and in men and the observations of hermaphroditic fish in the aquatic environment.[71,72,73]. NP has been reported to cause change in the expression level of mRNA for gonadotropin-releasing hormone (sGnRH), and estrogen receptor α(ERα) isoforms in the brain.[74]

Broadest definition of environmental estrogen is that exogenous estrogenic compounds mimicking the endogenous action of 17ß-estradiol, a female specific hormone, in organisms because they resemble the natural structure of estradiol (Figure 5).

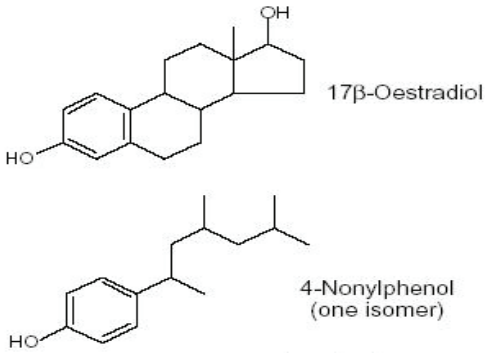

Figure 5. A Comparison of the structures of 17ß-estradiol and 4-nonylphenol (Warhurst, 1995).

Şekil 5. 17ß-östradiol ile 4-nonilfenolün yapılarının bir karşılaştırılması (Warhurst, 1995).

Their molecular mechanism of action may be the direct interaction of parent compound or their metabolites with estrogen receptors.[68] Giesy et al.[75] demonstrated that 3.4 µg/L water borne NP exposure to female fathead minnows resulted 900 % elevation of endogenous 17ß-estradiol in the blood and this elevation of estradiol induced vitellogenin production in the liver.

APs also exert estrogenic activity in fish, birds, reptiles and mammals.[76,77] It has been shown that NP induced the synthesis of vitellogenin in the liver

of rainbow trout under *in vitro* and *in vivo* conditions.[78,79,80,81] Sumpter and Jobling[82] indicated that vitellogenesis is a biomarker for estrogenic contamination in aquatic environment. Harries et al.[83] demonstrated that the caged male rainbow trout exposed to effluents of sewage treatment works in different rivers in UK caused an increase in the level of vitellogenin (Vtg), a large lipoglycophosphoprotein synthesized, lipidated, glycosylated, and phosphorylated in the liver of female individuals in the egg laying organisms, secreted into the blood stream and is finally taken up by oocytes through the receptor mediated endocytosis.

Synthesis of vitellogenin is regulated by endogenous 17ß-estradiol and is not detectable in males and immature females.[84] However, it has been shown that the exogenous administration of 17ß-estradiol and NP stimulated Vtg synthesis in male individuals of various fish species including rainbow trout[55], eelpot [80], and carp.[85] It has also been shown that NP could induce the production of the eggshell zona radiata proteins (Zrp), precursors of inner layer of egg-shells and are only found in the female fish in which hepatic synthesis of them are estrogen dependent.[87] However, because of environmental estrogen exposure, these precursors have also been found in male fishes including rainbow trout[87,88] and medaka.[89] Nice[90] demonstrated that the concentrations of 750 ng/l NP impairs the motility of oyster sperm and it has detrimental effects on sperm viability and embryo development in sea urchin.[91,92,93]
It has been reported that the estrogenic effects of nonylphenol may also induce morphological changes in the organism. For example, the inhibition of spermatogenesis in adult male rainbow trout[55] and the cytological changes of germ and Sertoli cells in male eelpot,[80] and increase in ovosomatic index in Juvenile rainbow trout were observed.[94] On the contrary to these reports, Adeoya-Osiguva et al.[95] demonstrated that the estrogenic activity of NP stimulated the capacitation and the acrosome reaction (AR) of mammalian sperm. Fraser et al.[96] indicated that even a very low concentration of xenobiotic or a combination of xenobiotics present in female tract could have adverse effects on the ability of sperm to fertilize egg in-vivo by accelerating capacitation and AR in mouse and human sperm.

It has been well established that the steroid hormones can affect the direction of sex differentiation in many species of fishes and estrogens induce feminization and androgens induce masculinization.[97,98] For example, sex differentiation could be altered by 100% with exogenous steroid hormones in tilapia,[99] chinook salmon,[100] rainbow trout[101] and largemouth bass.[102]

Since exogenous hormone application can induce sex reversal in fish, there is a considerable possibility of sex reversal caused by the exposure of environmental endocrine disrupters including alkylphenolic compounds. Indeed, it has been shown that alkylphenols can induce sex reversal in Japanese medaka.[103] It has also been observed that the development of testis-ova in Japanese medaka[103,104] and the appearance of an oviduct in genetic male carp were observed upon the due to the exposure of nonylphenol.

Sex reversal in response to increasing amount of environmental endocrine disrupters including alkylphenolic compounds may have the deleterious effects on the conservation of both aquatic and terrestrial organisms of lower vertebrates including fishes.

6.2. Carcinogenic effects

It has been reported that endogenous estrogens can induce the development and growth of breast cancer in women.[105,106] Along with these findings, it has also been debated that environmental estrogens may contribute the development of breast cancer in women and may decrease the male and female reproductive capacity.[107]

The human breast cancer cell line (MCF-7) has been shown to respond to estrogen and estrogenic alkylphenols stimuli by increasing DNA-synthesis. Thus, this cell line is frequently used to investigate estrogen or estrogen mimicking substances to understand the mechanism of their action on the induction of cell proliferation.[108] MCF7 cells exposed to NP, bisphenol A or DES induces dose dependent-increase in telemoric association and chromosome breaks and this action of alkylphenols are reported to be independent form their estrogenic effects.[109] The incidence of testicular cancer has been reported to increase three-to four-fold over the past 50 years in several human populations, particularly in the Western world.[110]

6.3. Toxic effects

Argese et al.[111] reported that APs were toxic to animals, plants, and microorganisms, probably due to their hydrophobic alkyl residue that causes alteration in cell membranes and alkylphenols lead to uncoupling of energy production in the mitochondrial membranes. In mammals, the toxicity of alkylphenols usually increases as the length of ethoxy units increases because long ethoxy units accelerate the absorption of APEs in mammals.[112]

6.3.1. Mammalian toxicity

Actually, acute toxicity of APEs to mammals is very low. For example, oral LD_{50} dose for rats and mice range from 2 to 4 g/kg. However, dermal irritation was observed with lower doses (500 mg/ kg) and severe eye irritation was observed with 5mg/kg dose.

Humans are exposed to APEs and their degradation products via water supply, sewage sludge used for fertilizer, aquatic flora and fauna serving as food, and directly use of APE spermicides to avoid conception. Commonly, the surfactant nonoxynol-9 is used intravaginal spermicide in humans. Nonoxynol-9 consists 9 ethylene oxide units and has been shown to be absorbed rapidly following intra-vaginal administration in rats[113,114] and was rapidly metabolized to NP which subjected to glucuronidation.[115] Jick et al.[116] was initially addressed the occurrence of teratogenesis due to the usage of APE spermicide. It was also found that nonoxynol induced transformations in a mouse embryo cell lines.[117]

6.3.2. Aquatic toxicity

As Nimrod and Benson reviewed,[1] the toxicity of alkylphenols to aquatic organism increases with a decreasing number of ethylene oxide units and increasing hydrophobic chain length. Therefore, the toxicity of parent molecules of alkylphenols is less than that of the degradation products NP, NP1EO and NP2EO. For example, LC_{50} value of NP16EO at the 48-hr for Japanese medaka was 110 mg/l and this decreased to 11.2 and 1.4 mg/L for NP9EO and NP, respectively.

6.4. Alkylphenol-induced genomic instability

Increased mitotic activity and alteration of cell cycle kinetics are considered important factors for the development of genetic instability.[118] Nonylphenol and bisphenol A stimulate massive burst in the synthesis phase of cell cycle while inhibiting DNA repair and thus introducing DNA damage.[119,120,121] These events might produce genetic instability, which could be factor in the development of adverse effects in mammary glands. As mentioned above, exposure of MCF7 cells to common estrogenic environmental endocrine disrupters, such as DES, Bisphenol A, or nonylphenol stimulated dose-dependent increase in telomeric association and chromatid breaks [121]. However, Ruggeri et al.[122] reported that 4-NP is able to act through an alternative pattern to that of estradiol-17β, modulating the expression of the same genes in a different manner. Indeed, it has been reported that nonylphenol (NP) stimulated the expression of mRNAs in oocytes[123] and mRNA expression of hemocyte genes in freshwater prawn (*Macrobrachium rosenbergii*).[124]

6.5. Alkylphenol-induced histopathological changes in organisms

Mostly, alkylphenol induced histopathological lesions are attributed to estrogenic effects of these compounds. For example, inhibition of spermatogenesis and inhibition of testicular growth in rainbow trout,[55] the appearance of an oviduct in genetic male carp (*Cyprinus carpio*)[85] and the development of testis-ova and sex reversal in Japanese medaka (*Oryzias latipes*)[103] are all attributed to the estrogenic effects of alkylphenols.

Burkhardt-Holm et al.[125] reported that nonylphenol exposed to rainbow trout demonstrated the severe histopathological changes in epidermal mucous cells. The structure of epidermal cells is altered, often cell border was not visible, pavement cells were detached, vacuolation of cytoplasm and deformed cell nuclei were observed in rainbow trout skin[125]. Also, chronic toxicity test on rainbow trout induced haematological changes including decrease in leucocytes and erythrocytes count.

Although there were no effects on the secondary sex characters, histopathological lesions were also observed in fathead minnows (*Pimephales promelas*) gonads in response to nonylphenol exposure.[126] Time and dose dependent histopathological lesions were also observed in the liver of rainbow trout due to the NP exposure.[127]

6.6. Alkylphenol induces biochemical and biophysical changes in animal cells

Alkylphenols are metabolized in the liver and excreted through bile and urine. However, it was reported that NP exerts biochemical effects by down regulating the activity of microsomal CYP1A while it increases the level and activity of CYP3A protein in the liver of mammals.[128] Uğuz et al.[127] reported that NP decreased the glutathione S-transferases (GSTs) activity, which have been known as the important part of the cellular detoxification systems and evolved to protect cell against the reactive metabolites[129] in the liver of rainbow trout. Cakmak et al.[130] demonstrated by using Fourier Transformed Infrared Spectroscopy that nucleic acids exhibited increased number of hydrogen bonded phosphodiester groups in the liver cells of NP treated rainbow trout.

7. Conclusion

This review of literature clearly shows that the environmentally relevant concentrations of APEs and their derivatives such as NP, BP and OP are not lethally toxic to majority of organisms on earth. However, these substances bioaccumulates in organisms that reaches lethally toxic levels. Furthermore, even the sub-lethal concentrations of these substances exert estrogenic effects that disrupt the endocrine system. Thus, they pose a serious danger to the conservation of wildlife especially to the conservation of aquatic organisms by disrupting their reproduction. These man-made chemicals also pose serious threat to the health of almost all vertebrates including mammalians by causing various cancers such as testicular, ovarian and breast cancer ■

KAYNAKLAR

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