Alkyl Ethoxylated and Alkylphenol Ethoxylated Nonionic Surfactants: Interaction with Bioactive Compounds and Biological Effects

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This review deals with recent advances in the study of interactions of nonionic surfactants with proteins, peptides, amino acids, membrane phospholipids, and organisms. The effect of surfactants on the structure and biological activity of the interacting biomolecules and organisms is discussed, with emphasis on the impact of hydrophobic and hydrophilic molecular substructures on biological efficiency. *Key words*: alkyl ethoxylated nonionic surfactants, alkylphenol ethoxylated nonionic surfactants, phospholipids, proteins, surfactants. *Environ Health Perspect* 103:358–364 (1995)

Nonionic surfactants are amphipathic molecules consisting of a hydrophobic (alkylated phenol derivatives, fatty acids, long-chain linear alcohols, etc.) and a hydrophilic part (generally ethylene oxide chains of various length). Although not as important commercially, tertiary amine and various sugar surfactants are also nonionic surfactants. Due to their favorable physicochemical properties, nonionic surfactants are extensively used in many fields of technology and research. The application of nonionic surfactants in various biotechnological processes has been recently reviewed (*1*). Surfactants have been successfully used to decrease the foaming of fermentation broths during solvent extraction (*2*), increase the conversion of linoleic acid to its hydroperoxide (*3*), and enhance the rate of cellulose hydrolysis (*4*). Nonionic surfactants are an integral part of the majority of pesticide formulations (*5*). They increase the leaf retention of spray solutions (*6*), enhance adhesional forces of aqueous droplets on crop leaf surfaces (*7*), and generally improve the effectiveness of active ingredients (*8,9*). However, not only do surfactants influence the performance of pesticides, but the pesticides exert some effects on the fate of surfactants; for example, pesticides promote or inhibit the photolytic degradation of nonionic surfactants (*10*).

Nonionic surfactants are also used in pharmaceuticals to increase their stability (*11*) and to enhance the dissolution rate of active ingredients from suppositories (*12*) and solid dispersions (*13*), for example. The pharmaceutical industry also uses nonionic surfactants to facilitate solubilization (*14*) and to increase the stability of drug-carrier emulsions (*15*). Surfactants markedly modify the particle size of precipitated drugs, too (*16,17*). Due to strict regulations, nonionic surfactants have only limited application in the food industry, where they are employed to change the stability of various emulsions (*18*) and to decrease the retrogradation of amylopectin (*19*). Nonionic surfactants also have been used in analytical chemistry to increase the fluorescence of dansylated amino acids (*20*), improve protein separation in capillary zone electrophoresis (*21*), and mask side effects in spectrophotometry (*22*).

This review presents a critical evaluation of recent results of studies on the interaction of alkyl ethoxylated and alkylphenol ethoxylated nonionic surfactants with various bioactive macromolecules and with organisms. The fate of surfactants in various ecological systems has been extensively studied. Nonionic surfactants are generally easily degradable; however, in some cases the persistence of intermediates has been observed. Due to the limited scope of this review, investigations of intermediates will not be discussed in detail.

Interaction with Bioactive Macromolecules

The mode of action of nonionic surfactants and the hydrophilic (electrostatic) or hydrophobic character of their interaction with bioactive molecules, organs, and organisms have been extensively discussed. The results are sometimes contradictory, and the character of interaction depends considerably on the interactive molecular species.

Proteins, peptides, and amino acids. Many studies have indicated that nonionic surfactants readily bind to various proteins. This phenomenon has been frequently exploited to extract and solubilize sparingly soluble proteins such as membrane proteins (*23*). Nonionic surfactants derived from tris(hydroxymethyl)-aminomethane perform well in the solubilization of subcellular proteins of rat hepatocytes and membrane antigens from tumor cells (*24*). Nonionic surfactants are generally less effective than ionic surfactants; for example, Tween 80 and polyoxyethylene-9-laurylether have a negligible effect on the dissociation, γ-chymotryptic degradation, and enteral absorption of insulin hexamers (*25*). Surfactants also modify the adsorption capacity of proteins and peptides: Tween 80, Triton X-100, and PEG 6000 decrease the adsorption of urokinase on glass surfaces; however, they were less effective than gelatin (*26*). The adsorption of fibrinogen was markedly lower on polyoxyethylene-polyoxypropylene-coated polystyrene latex (*27*), and the adsorption on self-assembled monolayers of fibrinogen, lysozyme, pyruvate kinase, and RNAse A was inhibited by oligoethyleneoxides (*28*).

Surfactants exert a protective effect on proteins. At a 2% concentration, Tween 20 completely prevented the denaturation of rabbit skeletal myosin by freezing and thawing, and glycerol enhanced synergistically the protective effect (*29*).

The majority of research on protein–surfactant interaction has focused on the binding of surfactants to enzymes and the effect of surfactant binding on enzyme activity. These results will be discussed later. Because the molecular basis of the binding of surfactants to proteins has not been elucidated in detail, some investigators have tried to pinpoint individual amino acids accounting for the binding. Charge-transfer chromatographic methods indicate that nonylphenyl hexaethoxylate only interacts with some amino acids, with the order of relative strength of interaction Tyr> Glu>Phe>Hyp>Gln>Cys>Gly. A significant linear relationship has been found between the interactive strength and the hydrophobicity of amino acids. The authors concluded that the interaction of individual amino acids with the surfactant is fairly low and does not explain the strong interaction of surfactant with proteins observed in many studies (*30*). It was assumed that the long surfactant molecule lies parallel with the protein surface, contacting more than one amino acid residue. The strength of interaction varies according to the amino acid sequences, and hydrophobic forces are probably involved in the interaction (*30*). A similar study dealing with the interaction of amino acids with ethoxylated stearic acid surfactants found that surfactants interact with free amino acids in the following order: Cys>Phe>Tyr>Asn>Met>Nle>Leu> Gln>Lys>Ser>Trp. In this case the electronic parameters of surfactants had a significant impact on the strength of interaction (*31*).

The forces involved in the binding of nonionic surfactants to proteins are being characterized. The results indicate that the hydrophobic moiety of surfactants can bind to the apolar amino acids, whereas the hydrophilic ethyleneoxide chain can inter-

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act with the peptide bond and with one or more polar amino acid residues, probably by electrostatic forces and hydrogen bonding.

Membrane phospholipids. Results of many studies indicate that nonionic surfactants interact not only with proteins but also with membrane phospholipids by modifying their structure and permeability. As phospholipids are chemically simple compounds, the principles of various surfactant–phospholipid interactions and the character of forces involved are fairly well known.

Surfactants generally increase the permeability of phospholipid membranes and vesicles, causing leakage of compounds with low molecular mass. The loss of ions, amino acids, etc., may result in cell damage or cell death. It is generally accepted that the increased permeability is the result of membrane disruption. Supramolecular surfactants (polyethylene glycol + dicarboxylic acid esters) as well as Triton X-100 readily disrupt egg yolk phosphatidylcholine membranes (*32*). An increase in permeability has been observed in many model systems: Triton X-100 and some new synthetic surfactants caused leakage from palmitoyloleoyl phosphatidylcholine/cholesterol large unilamellar vesicles (*33*). The concentration and aggregation state of surfactants also exert a considerable effect on their membrane-damaging capacity: monomeric Triton X-100 causes leakage of dipalmitoyl-phosphatidylcholine vesicles, whereas micellar solutions result in the catastrophic rupture of membrane (*34*). Some new surfactants, (HO $(C_2H_4O)_6CO(CH_2)_{14}CO_2C_2H_4O)_6H$ and $HO(C_2H_4O)_6CO(CH_2)_6CH{=}CH(CH_2)_6CO_2C_2H_4O)_6H$ and their polymeric counterpart, were synthesized and their capacity to disrupt egg yolk phosphatidylcholine and palmitoyloleoyl phosphatidylcholine bilayers determined at various cholesterol concentrations in the bilayer. It was established that the effect of new synthetic surfactants depends on the cholesterol concentration in the bilayer, whereas the effect of Triton X-100 is not affected by the cholesterol concentration (*35*). Unfortunately, the cause of the damaging behavior of the new surfactants was not explained in detail. The same surfactants caused leakage or rupture of palmitoyloleoyl phosphatidylcholine vesicles depending on the membrane packing (*36*). The condensation product of hexaethyleneglycol and various dicarboxylic acids considerably increased the release of 5(6)-carboxyfluorescein from the large, unilamellar vesicles of palmitoyloleoyl phosphatidylcholine (*37,38*).

The interaction of surfactants with artificial membranes modifies many physicochemical parameters of the phospholipids: A fluorescence depolarization study indicated that alkanoyl-*N*-methylglucamide surfactants decrease the fluidity of dipalmitoyl phosphatidylcholine membranes (*39*). Nonionic surfactants decreased the phase transition temperature of negatively charged dilauroylphosphatidic acid membrane. The interaction between surfactant molecules incorporated in the lipid membrane was also observed (*40*).

The effect of surfactants on natural membranes has also been observed. Surfactant can disrupt not only artificial membranes but also modify the physicochemical characteristics of natural membranes. Nonionic surfactants were able to increase the permeability of sarcoplasmic reticulum vesicles (*41*), and Pluronic L81, a hydrophobic surfactant, markedly influenced the cholesterol homeostasis of intestinal mucosa; however, it was not specified whether this effect was due to the direct surfactant–cholesterol interaction or due to the result of other, not well known biochemical or biophysical processes (*42*).

The number of studies dealing with the elucidation of the relationship between surfactant structure and membrane-damaging activity is surprisingly low. Adiabatic differential-scanning calorimetric measurements indicated that [2-(alkoxy)-phenyl]-2-(1-piperidinyl)ethyl esters of carbamic acid interact with dipalmitoyl phosphatidylglycerol model membranes, and the effect depends on the length of ethyleneoxide chain (*43*). The effect of polyoxyethylene cetyl ethers on the vesicle to micelle transitions of egg yolk phosphatidylcholine liposomes also markedly depends on the length of polar ethyleneoxide chain (*44*). It has been found that polyoxyethylene-polyoxypropylene block copolymer molecules are intercalated with phosphatidylcholine monolayers (*45*).

Although the binding of surfactants to proteins and phospholipids seem to be two independent procedures, a comparative study suggested that there is a strong relationship between the skin irritation potential of surfactants and their capacity to increase dye leakage from egg yolk phosphatidylcholine unilamellar liposomes (*46*).

It can be concluded that the interaction of nonionic surfactants with membrane phospholipids involves the insertion of the hydrophobic moiety of surfactants into the apolar fatty acid domain of phospholipids. However, this insertion is not enough to disturb the membrane organization. Linear substructures (fatty acids, long-chain alcohols) are well accomodated and do not disturb the membrane organization. Bulky hydrophobic moieties (alkylated phenols) cause severe disturbances between the apolar fatty acid chains, resulting in increased permeability and leakage. The hydrophilic ethyleneoxide chain probably has two functions: it regulates the insertion depth of hydrophobic moiety (longer ethyleneoxide chain draws the hydrophobic moiety toward the aqueous outer phase), indirectly influencing its membrane damaging effect, or it binds to the polar head group of phospholipids. As the long ethyleneoxide chain can contact more than one head group, it can stabilize the membrane organization. The effects observed are the result of the interplay of the interactions outlined above.

Proteins and membrane phospholipids simultaneously occur in many living cells. In these instances surfactants can bind both to the proteins and phospholipids. The preference of surfactants either for proteins or for phospholipids in a complicated living system has never been studied in detail.

Biological Effects

Stimulation and inhibition of enzymes. Nonionic surfactants readily bind to various proteins, and the binding modifies protein solubility and structure. These changes may also result in the stimulation or inhibition of the biological activity of enzymes. Unfortunately, most studies dealing with the effect of surfactants on enzyme activity are limited to determining the degree of stimulation or inhibition and do not elucidate the underlying molecular mechanism.

An *N*-acetyl-D-glucosaminyltransferase detected in human carcinoma Colo 205 cells showed optimum activity in the presence of the nonionic detergent Triton CF-54 (*47*). Glycolipid glucuronyltransferase isolated from embryonic chicken brain shows optimum activity in the presence of neutral detergents such as Triton CF-54, Triton DF-12, and Nonidet P-40 (*48*). Triton X-100 activated lecithin:cholesterol acyltransferase (*49*), stimulated the activity of rat liver mitochondrial phosphatidylserine decarboxylase (*50*), and, together with other nonionic surfactants (Myrj 52, Myrj 59, Tween 20, Tween 80, etc.) at 0.1% (w/v), increased the activity of human leukocyte proteinase elastase and cathepsin G (*51*). Nonionic surfactants having a polyoxyethylene chain have been shown to effectively increase the activity of *Chromobacterium viscosum* lipase in aerosol bis(2-ethylhexyl)sodium sulfosuccinate reverse micelles (*52*). Octaethylene glycol dodecyl ether induced the dissociation of membrane-bound Na^+/K^+-ATPase purified from dog kidney (*53*). An interesting study indicated that the effect of surfactant strongly depends on its concentration: Triton X-100 stimulated the activity of the ATPase-active P-glycoprotein at low concentrations and inhibited it at higher concentrations (*54*).

The hydrophobic or hydrophilic character of surfactant–enzyme interactions has been established only in a few instances. Nonhomologous series of nonionic surfac-

tants increased the activity of papain and modified its structure as determined by differential scanning calorimetry. Both the hydrophobic and hydrophilic molecular characteristics of surfactants influenced their effect on the activity and structure of papain (*55*). In contrast, similar surfactants markedly inhibited the activity of horseradish peroxidase. Also in this instance both the hydrophobic and hydrophilic molecular characteristics of surfactants influenced their effect on the activity of the enzyme (*56*). Triton X-100 activated the plasma membrane ATPase. This effect was tentatively explained by the alteration of the hydrophobic environment around the enzyme (*57*). Reduced lysozyme at pH 2.5 bound polyoxyethylene alkylethers (C10E6, C12E6, and C12E8 surfactants); the maximum bond reached 0.5–0.7 mol/mol amino acid residue (*58*). It was further established that the interaction most likely takes place between the hydrocarbon tail of the surfactant and the hydrophobic domain of reduced lysozyme (*59*).

Many results prove that nonionic surfactants can considerably modify the activity of various enzymes. This effect can be both beneficial (biotechnological processes) or harmful (toxicity toward humans, animals, plants, etc.). It is currently impossible to predict the behavior of surfactant–enzyme systems. We need much more data on the molecular basis of mode of surfactant binding to proteins for the rational design of surfactants with optimal biological efficiency and with minimal toxic side effects.

Microorganisms and insects. Due to their capacity to interact with proteins and phospholipids, nonionic surfactants exert many biological effects on microorganisms and insects. These effects have been successfully exploited in some biotechnological and immunological processes. Tween 80 enhanced the ligninase production and growth of the fungi *Phanerochaete chrysosporium* (*60*). Polyethylene glycol 600 increased the γ-amylase production of *Bacillus subtilis*, whereas polyethylene glycol 3350, Triton X-100, and Tween 80 were ineffective, proving again that the character of surfactant has a marked influence on its biological efficiency (*61*). Tween 80 modified invertase secretion by *Neurospora crassa* and the cell-wall-less slime secreted by an *N. crassa* mutant(*62*). Polyethylene oxide-polypropylene oxide block-polymers up to 7.90 Da molecular mass stimulated the secretion of antibodies against *Streptococcus pneumoniae*-derived hexasaccharide–protein conjugates (*63*). The same block-polymers enhanced the avidity of antibodies in polyclonal antisera against *Streptococcus pneumoniae* type 3 in normal and Xid mice (*64*). Nonionic surfactants such as hexa-, octa-, and decaethylene glycol monohexadecyl ether in combination with alkyl phosphates inhibit the adherence of *Streptococcus mutans* on a hydroxyapatite surface (*65*).

Nonionic surfactants have toxic effects too. They increased cell fusion caused by polyethylene glycol (*66*). Triton X-100 and Triton XR suppressed spore germination and germ tube growth of *Mucor mucedo* on tomatoes in storage (*67*). Triton X-100 caused the cell death of *Bacillus subtilis* by inducing cell autolysis (*68*). It has been suggested that the surfactant interacts with the regulatory system of autolysis and thus affects the activation of autolysis in *B. subtilis* (*69*). Three to four orders of magnitude differences were found between the sensitivity of various algae species and surfactant toxicity (*70*). Two types of nonylphenol ethylene oxide-acetate did not influence the growth of *Acinetobacter calcoaceticus*, *Photobacterium phosphoreum*, or *Serratia marinorubra*, but inhibited the growth of marine heterotrophic flagellates (*71*). Nonionic surfactants (Activator N.F. and Ortho X-77) were moderately toxic to larvae of the midge *Chironomus riparius* (*72*).

The fate of nonionic surfactants in soils and surface waters has been vigorously studied. It was established that they decompose relatively easily; however, the results depend slightly on the characterisitcs of the ecological system under investigation. Polyethoxylated linear alcohol derivatives were mineralized without lag periods by rhizosphere microbial communities in surface soils (*73*). The microflora of aquatic plants decompose about 30–40% on nonionic surfactants in 30 days (*74*). According to another study, the half-life of linear ethoxylated surfactants was 8.4 days as decomposed by the microbiota of submerged plant detritus (*75*). The effect of surfactants on the biodegradation of other xenobiotics has not been determined unambiguously. One study found that nonionic surfactants inhibit the mineralization of phenanthrene in soil, probably by interacting with the membrane of soil microflora (*76*), whereas another study reported that the nonionic surfactant $(CH_2)_{12\text{-}14}(OCH_2CH_2)_{5.6}OH$ added to the soil surface promoted the biodegradation of phenanthrene and biphenyl in Lima silt loam (*77*). This discrepancy may be due to the different microbial populations of soils and the different stability of surfactants against microbial decomposition.

The relationship between the microbiological effect of surfactants and their chemical structure has been studied only in a few instances. Tween compounds induced hydrogen production in aqueous suspensions of *Anabaena variabilis* in the order Tween 85>Tween 80>Tween 60. Tween 20 was ineffective (*78*). This finding indirectly proves that the effect of surfactants depends both on the character of the hydrophobic moiety and the length of the polar ethylene oxide chain. Polyalkylene glycols improved cell growth, viability, and alcohol production of *Saccharomyces cerevisiae*. The effect depended on the number of ethylene oxide groups in the surfactant molecule (*79*). Surfactants with more ethylene oxide groups showed lower toxicity toward *Mysidopsis bahia* (*80*).

Nonionic surfactants can stimulate or inhibit the growth of a wide variety of microorganisms. These effects have a marked impact on human health care, biotechnology, environmental protection, and agrochemistry. A better understanding of the underlying biochemical and biophysical processes would be of considerable interest for the safer application of nonionic surfactants.

Plants. The direct effect of nonionic surfactants on plant species has rarely been studied because surfactants generally contact plants in combination with various pesticides. It has been found that nonionic surfactants cause phytotoxic symptoms in tobacco (*Nicotiana tabacum*), sugar beets (*Beta vulgaris*), and spiderwort (*Tradescantia albiflora*). Surfactants with low and high numbers of ethylene oxide groups were less effective (*81*). It has been shown that the more hydrophilic surfactants (fewer ethylene oxide groups) had the smallest effect both on ethylene evolution and leaf growth in *Phaseolus vulgaris* (*82*). Nonionic surfactants considerably decreased the net potassium influx in roots of wheat seedlings; their effect depended on the number of ethylene oxide groups and on the overall hydrophobicity of the surfactant (*83*). The pH of the solution did not significantly influence the sorption of octylphenoxy surfactants on isolated tomato fruit cuticles, indicating that ionic interactions have a negligible effect on sorption (*84*). The toxicity of these surfactants to cowpea leaves was found to be inversely related to the length of the ethylene oxide chain (*85*).

These data suggest that the physicochemical parameters of surfactants play a considerable role in the extent of phytotoxic activity. Similar results have been found when surfactants were used in combination with pesticides. Octylphenoxy surfactants increased the foliar uptake of DDT and atrazine. The effect was inversely related to the hydrophile:lipophile balance of surfactants (*86*). Complex stability between 2-(1-naphthyl)acetic acid and surfactant micelles decreased with the logarithm of the length of ethyleneoxide chain for

Triton X surfactants. Nondissociated forms of the plant growth hormone formed more stable complexes (*87*).

Animals and animal models. The widespread use of nonionic surfactants makes it probable that organisms may absorb a great quantity of surfactants. To elucidate their toxic effects, a variety of animal models have been used.

In rats, surfactant can enhance the toxic effects of xenobiotics when administered simultaneously. Surfactants increased the absorption of xenobiotics in rat colon (*88*). Tween 80 enhanced the intestinal absorption of the anthelminthic drug albendazole in rat gut (*89*), whereas polysorbate 80 increased the absorption of phenylalkylcarboxylic acids in rat colon (*90*).

Nonionic surfactants themselves show toxic effects. Hexaethoxylated linear primary alcohol ($C_{9\text{-}11}$) is moderately toxic by the oral route in rats. By the dermal route, it does not produce skin irritation or systemic or reproductive toxicity at concentrations used in formulated cleaning products (*91*). Lubrol PX 0.8% (v/v) (pH 6.98–0.02) and Triton X-100 0.5% (v/v) (pH 7.41–0.03) significantly increased the pH of mucosal surface of rat proximal jejunum (control pH 6.23–0.02) (*92*). Emulgen 913 (polyoxyethylene glycol nonylphenyl ether) decreased liver weight and the cytochrome P450, cytochrome b_5, and microsomal heme content in rats, whereas heme oxygenase activity was greatly enhanced (*93,94*). The nonionic surfactant nonoxynol-9 changes vaginal permeability in ovariectomized rats as determined by nigrosin staining and measurement of bioelectronic parameters, whereas Tween 80 was ineffective (*95,96*).

In mice, polysorbates (Tween 20, 21, 80, and 81) as well as poloxamer and poloxamine surfactants had only a slight influence on the permeability of methanol through a full thickness mouse skin; however, the permeability of lipophilic octanol decreased (*97,98*).

In rabbits, nonionic surfactants enhanced the systemic absorption of α-melanocyte-stimulating hormone via the ocular route in rabbits (*99*). The cytotoxicity order of surfactants on rabbit corneal epithelial cells was cationic>anionic=amphoteric>nonionic; however, Triton X-100 had a ranking similar to anionic surfactants (*100*). Poloxalene (30% polyethyleneoxide and 70% polypropylene oxide, MW 3000) inhibited neutral fat and cholesterol absorption in rabbits (*101*). The study of the uptake of neutral red by rabbit corneal cells revealed that nonionic surfactants have a lower toxic effect than cationic, anionic, and amphoteric ones (*102*).

Triton X-100 at concentrations over the critical micellar concentration induced lysis of isolated gill epithelial cells in *Oncorhynchus mykiss* (*103*); however, Triton X-100 showed a lower effect than ionic surfactants (*104*). Emulgen 913 (polyoxyethylene glycol nonylphenyl ether) significantly decreased the concentration of metal-binding proteins in the hepatopancreas and lessened the heme-oxygenase activity in the kidney of red carp (*105*). The adsorption of salicylic acid on hamster cheek pouch decreased in the presence of the nonionic surfactant polysorbate 80, while ionic surfactants enhanced adsorption (*106*).

The results discussed above clearly show that nonionic surfactants influence many biological processes, and the effect is general noxious to the living organisms. However, it has been found that Tween 20 was as efficient as natural surfactant in improving gas exchange and compliance in preterm lambs with respiratory failure (*107*).

The structure and physicochemical parameters of surfactants exert a marked impact on their biological activities. The effect of nonylphenol-polyethoxylates on the bioelectric properties of the vagina of rats showed a nonlinear relationship with the number of ethylene oxide groups per molecule (*108*). Surfactants having a linear alkyl chain greater than 8 carbons and an ethylene oxide chain length of less than 18 caused significant increases in the flux of methyl nicotinate across hairless mouse skin. Surfactants having branched alkyl chain or aromatic moieties in the hydrophobic portion were ineffective (*109*). The toxicity of polyoxyethylene alkyl ethers decreased by increasing length of the alkyl chain and increased by the length of the polyoxyethylene headgroup (*110*).

These data draw attention to the fact that the appropriate selection of surfactants and the synthesis of new surfactants with less toxic side effects may result in lower environmental pollution without losing the advantages of surfactant application.

Human aspects. Human skin has the highest probability of being in contact with surfactants. The cytotoxicity of 17 surfactants on cultured human skin fibroblasts were determined, and it was found that Brij 35, 58, and 99 are a highly cytotoxic. Addition of fetal calf serum decreased the toxicity, probably by binding the surfactants and lowering the concentration of free surfactants (*111*). Brij 78, Brij 99, and Triton X-100 were more toxic than Tween 40 and 80 (*112*). It has been stated that the method used is suitable for predicting irritation potential of surfactant *in vivo*.

Not only can surfactants cause skin irritation, they can also exert beneficial effects, such as promoting the transport of drugs accross the skin. Brij-36 increased the transport of methyl nicotinate and hexyl nicotinate across the skin, whereas sodium dodecyl sulfate was ineffective (*113,114*). Surfactants can effectively increase the transdermal permeation of therapeutic peptides and proteins (*115*). Polysorbate 80 and polyoxyl 40 markedly influenced the transepithelial permeability in monolayers of human intestinal epithelial cells (*116*). The capacity of surfactants to increase the transport of many drugs across the skin may be due to the interaction of surfactants either with the drug or with the skin: sorbitane mono-oleate and polyoxyethylene-*n*-lauryl ether can interact with both the drugs and the skin in degrees dependent on the polarity of the surfactant and the drug (*117*). Diethylene glycol laurylether increased the penetration of theophylline and adenosine into excised human skin by a factor of 2.2–2.7, respectively (*118*). Anionic and cationic surfactants exert a marked effect on the permeability of human skin, whereas the effect of Tween 60 was negligible (*119*).

Surfactants can modify the permeability of blood cells when they enter the organism. Triton X-100 caused a rapid release of ATP from human red blood cells, while the presence of Brij 58 retarded the mobilization of the intracellular ATP (*120*). A study comparing two cytotoxicity tests for predicting ocular irritancy established that the red blood cell lysis test was predictive. Surfactants caused membrane disruption; anionic and cationic surfactants were more toxic than nonionic ones (*121*). Polyethoxylated nonionic surfactants inhibit the transport of 2,4-dinitrophenyl glutathione out of intact human erythrocytes. Surfactants may modify the arrangement of integral membrane proteins such as P glycoprotein and presumably the glutathione transporters (*122*).

Nonionic surfactants show considerable therapeutic effects by synergistically increasing the efficiency of drugs. The nonionic block-polymer surfactants L101 and 31R1 stimulated the induction of delayed-type hypersensitivity on the murine humoral and cellular immune response to a synthetic peptide composed of amino acid residues 9–21 of herpes simplex virus type 1 glycoprotein D (*123*). The neuroleptic activity of haloperidol increased in the presence of the nonionic surfactant poly(55)oxypropylene/dipoly(8)oxyethylene (*124*). Parental P388 murine leukemia cell lines sensitive to adriamycin, a subline of P388 resistant to adriamycin; sarcolemma-180; and Ehrlich ascites tumor were used to study the influence of nonionic surfactants on the activity of adriamycin. An enhanced biosynthesis inhibition by adriamycin was observed when used in combination with Brij 30 or Brij 35 in all the murine tumor models.

The increase in adriamycin cytotoxicity was due to an increased accumulation of adriamycin in the tumor models (*125*). Polyethoxylated nonionic surfactants with no similarities in the hydrophobic moiety are able to reverse multidrug resistance in a human leukemia cell line (*126*). Triton X-100 prevented the net uptake of vinblastin in inside-out membrane vesicles prepared from multidrug-resistant human leukemia cells (*127*).

It can be established that nonionic surfactants are moderately toxic to humans, and they probably can synergistically increase the toxicity of other xenobiotics. However, the beneficial effect of surfactants (promotion of penetration of drugs across the skin, increase of the effect drugs) probably overshadows their eventual noxious effects, and these compounds can be a useful tool for the improvement of human health care in the future.

Conclusions

Nonionic surfactants are widely used in many fields and exert both beneficial and toxic effects. They bind to proteins as well as to phospholipids influencing (stimulating or inhibiting) enzyme activity and membrane permeability. Hydrophilic and hydrophobic forces are simultaneously involved in the binding, and the effects observed are the result of the interplay of the various interacting forces. As recent research indicates, the biological effects strongly depend on the structure of surfactants. We need additional data for the more profound elucidation of the relationship between molecular structure and biological efficiency. With the exact knowledge of this relationship, it will be possible to select for each purpose a surfactant with minimal toxicity and maximal benefits.

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