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Review Article

NSAID injury to the gastrointestinal tract: evidence that NSAIDs interact with phospholipids to weaken the hydrophobic surface barrier and induce the formation of unstable pores in membranes

Lenard M. Lichtenberger, Yong Zhou, Elizabeth J. Dial and Robert M. Raphael

Abstract

In this review, we have discussed our current understanding of the barrier properties that are in place to protect the upper gastrointestinal mucosa from luminal acid, and the pathogenic mechanism by which nonsteroidal anti-inflammatory drugs (NSAIDs) induce injury to the gastrointestinal tract. The changes in our view of the importance of NSAID-induced cyclo-oxygenase (COX) inhibition on the pathogenesis and prevention of NSAID-induced gastrointestinal injury is presented. The focus of this paper has been placed on the effects of NSAIDs on the mucosal surface, and specifically the effect of these powerful drugs in inducing changes in the hydrophobicity, fluidity, biomechanical and permeability properties of extracellular and membrane phospholipids. Lastly, recent evidence is presented that salicylic acid and related NSAIDs may alter the stability of membranes, inducing the formation of unstable pores that may lead to back-diffusion of luminal acid and membrane rupture. This understanding of the interaction of NSAIDs with membrane phospholipids may prove valuable in the design of novel NSAID formulations with reduced gastrointestinal side-effects.

The Department of Integrative Biology & Pharmacology, The University of Texas Medical School, Houston, TX, USA

Lenard M. Lichtenberger, Elizabeth J. Dial

The Department of Biochemistry and Cell Biology, Rice University, Houston, TX, USA

Yong Zhou

The Department of Bioengineering, Rice University, Houston, TX, USA

Robert M. Raphael

Correspondence: L. M. Lichtenberger, Department of Integrative Biology & Pharmacology, The University of Texas Medical School at Houston, 6431 Fannin Street, Houston, TX 77030, USA. E-mail: lenard.m.lichtenberger@uth.tmc.edu

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Introduction

The mechanism by which nonsteroidal anti-inflammatory drugs (NSAIDs) induce injury to the gastrointestinal tract remains controversial, and the dogma that all gastrointestinal side-effects and other actions (anti-neoplastic actions) are attributable to an inhibition of cyclooxygenase (COX) and a depletion of mucosal prostaglandin levels is under increased scrutiny. In this paper we will review our current understanding of the molecular and biophysical basis of the barrier properties of the gastrointestinal tract and specifically the gastric mucosal barrier and the effect of NSAIDs on these barrier properties. We will also introduce recent work from our laboratory and others demonstrating that NSAIDs have the capability of partitioning into synthetic and biological membranes, undergo chemical interactions with the component phospholipid molecules and cause marked changes in the membrane's fluidity, biomechanical, structural and permeability characteristics.

Characteristics of the barrier properties of the gastrointestinal tract

One of the properties of the gastrointestinal tract that has yet to be fully elucidated is what protects the epithelium from the noxious, corrosive and proteolytic chemicals that are contained within its lumen. This barrier property is particularly striking in the stomach, where the pH of gastric juice can fall to a pH of ~1.0 between meals, yet the tissue is protected against the million-fold proton gradient established between the lumen and the intracellular environment. The concept that the stomach is protected by a gastric mucosal barrier was eloquently proposed and supported by a series of papers by Horace Davenport in the mid 1960s (Davenport 1964, 1965, 1972). Over the past 40 years, evidence generated by many laboratories has implicated a number of gastric properties as being important in contributing

to the barrier properties of the stomach, including: gastric mucus, and specifically mucus glycoproteins, which can form an adherent gel 100–200 μm in thickness over the tissue (Atuma et al 2001; Allen & Flemstrom 2005); intercellular junctional complexes, whose strand density has been implicated in the regulation of the paracellular resistance between cells (Anderson & Van Italie 1995; Ma et al 1999; Turner et al 1997; Fasano 2000; Amieva et al 2003); the ability of gastroduodenal tissue to secrete bicarbonate that can form a solid-state mucus/bicarbonate pH gradient between the tissue and the bulk luminal fluid (Isenberg et al 1987; Konturek et al 2004; Allen & Flemstrom 2005); the resistive properties of the plasmalemmal membrane, which increases under acidic pH (Sanders et al 1985; Barreto & Lichtenberger 1992); the ability of the epithelium to rapidly reconstitute itself after injury due to cell migration and proliferation (Ito & Lacy 1985; Lacy 1988; Takahashi et al 2003); and the unique biophysical properties of the surface of the upper gastrointestinal tract, which may specifically relate to the non-wettable hydrophobic characteristics of mucus or the membrane itself (Lichtenberger 1995, 2001; Darling et al 2004). It is this latter property that will be the focus of this paper.

Effect of damaging and protective agents on the barrier properties

One of the major ways of investigating and characterizing the barrier properties of the upper gastrointestinal tract is to expose the tissue, under either in-vivo or in-vitro conditions, to test agents and to study their ability to either induce tissue injury or to protect the mucosa from damaging agents. Using this approach, Davenport and other investigators (Davenport 1964, 1965, 1972; McCormick & Brune 1987) have demonstrated that aspirin, salicylate and other weak acids are potent damaging agents, along with natural (e.g. bile acids) and synthetic detergents, alcohols, hypertonic salt solutions, strong HCl (0.3–0.75 M) and proteolytic (pepsin) and lipolytic (phospholipase) enzymes. Furthermore, using these model systems, a number of protective agents were described, such as prostaglandins, growth factors (EGF, FGF, trefoil peptides), defensins (e.g. lactoferrin) and phospholipids (Davenport 1964, 1965, 1972; McCormack & Brune 1987; Lichtenberger 1999, 2001; Wallace 1997; Podolsky 2000; Dial et al 2004). These diverse groups of protective factors were demonstrated to act by stimulating mucus secretion or its gel-forming, pH gradient or hydrophobic properties, increasing epithelial cell proliferation or migration, or the cell's resistance to bacteria or their toxins. These studies, in addition to identifying both damaging and protective factors, also provided valuable evidence of the limits of one or more of these gastrointestinal properties in accounting for the tissues' barrier characteristics. For example, it has been demonstrated that mucus glycoproteins present a weak diffusion barrier to strong acid, and that the mucus gel layer is disrupted when the luminal pH < 2 (Allen & Flemstrom 2005). These findings therefore suggest that the gastrointestinal barrier properties when the gastric juice is highly acidic (pH < 2) must reside in intracellular or extracellular elements other than those residing with mucus glycoproteins. It also has been demonstrated that intercellular junctions are frequently found intact in tissue exposed to a damaging agent, although mucosal barrier disruption has occurred, as determined by changes in the permeability properties of the tissue, and the extrusion of plasma proteins and haemoglobin into the lumen (Davenport 1972). Alternatively, it has been demonstrated that the CagA protein of *H. pylori* may cause gastric injury by binding to and altering the apical/junctional complex (Amieva et al 2003). Also, although epithelial proliferation and migration are a dynamic property and play a role in tissue reconstitution and healing after injury, most investigators agree that the kinetics of the responses are too slow to account for the tissue's intrinsic barrier characteristics. All these properties have been reviewed in detail by this and other investigators in previous publications (Wallace 1997; Lichtenberger 1999, 2001).

Role of extracellular and membrane phospholipids in gastrointestinal barrier properties

Our laboratory originally proposed that the barrier properties of the stomach, and perhaps other regions of the gastrointestinal tract, exposed to noxious luminal agents is attributable, in part, to it's unique hydrophobic characteristics that makes it non-wettable to luminal acid and other aqueous damaging agents (Hills et al 1983; Lichtenberger 1995). Furthermore, we proposed and provided evidence for the hypothesis that this biophysical barrier property was dependent on the ability of mucus cells to secrete surfactant-like phospholipids that form extracellular layers, perhaps a monolayer, that adsorbs directly to the luminal interface of the mucus gel layer or the plasmalemmal membrane itself (Figure 1) (Goddard & Lichtenberger 1987; 1990; Lichtenberger 1995, 2001). This hydrophobic characteristic, as measured by contact-angle analysis, has been demonstrated to significantly correlate with the barrier property of the tissue, being reduced by damaging agents and maintained or increased by protective agents. It is also conceivable, if not likely, that phospholipids contribute to the barrier property of the gastrointestinal tract by forming tightly packed bilayers that provide a formidable transcellular resistive pathway to the movement of protons, as demonstrated by our and other laboratories (Sanders et al 1985; Barreto & Lichtenberger 1992).

The effect of NSAIDs on the barrier properties of the gastrointestinal tract

Aspirin and salicylic acid were one of the first series of damaging agents described by Davenport, which rapidly partitioned into the mucosa and caused barrier disruption, bleeding and ulcer disease (Davenport 1964, 1965, 1972). Since these early studies it has been demonstrated by Vane and others (Vane 1971; Whittle et al 1980; Mitchell et al 1993) that these weak acids are members of a much larger family of molecules, called NSAIDs, that share potent anti-inflammatory, analgesic and anti-pyretic activity and appear to act primarily by inhibiting the biosynthesis of prostaglandins, by inhibiting the enzyme cyclooxygenase (COX). This important finding was followed shortly afterwards by the demonstration by Robert and associates that prostaglandins have the capacity to protect the gastrointestinal mucosa from a number of damaging agents and

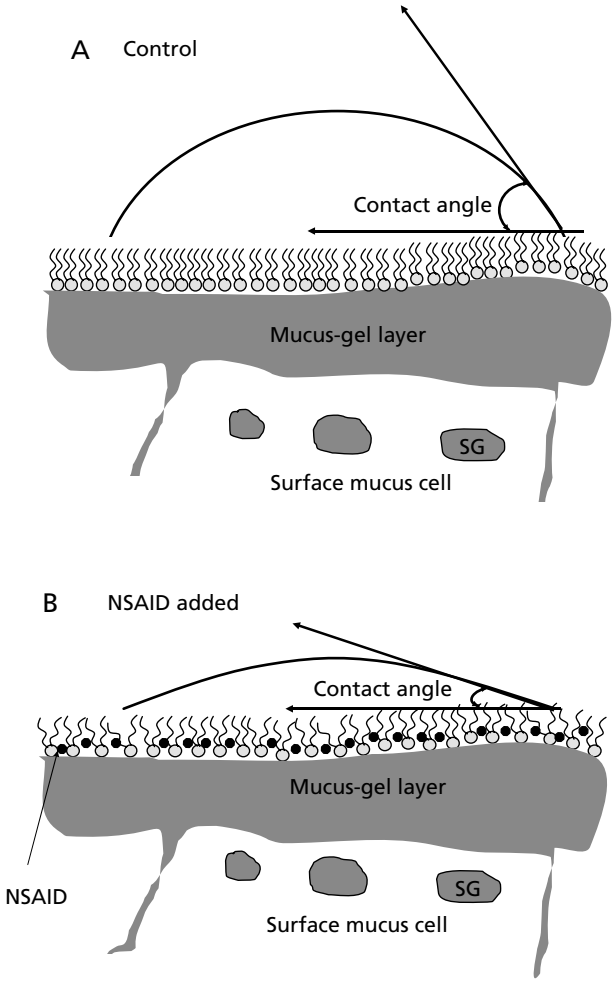

Figure 1 Schematic molecular model of putative phosphatidylcholine-enriched monolayer at the interface between the gastric mucus gel and the gastric juice to provide an acid repellent surface hydrophobic barrier. This property can be determined by measuring the contact angle at the air–liquid–solid interface. Schematic depiction of the hydrophobic properties of the gastric mucosa under control conditions (A) (contact angle readings between 60–80°) and after the stomach has been exposed to a luminal NSAID and surface hydrophobicity is reduced (B) (contact angle readings <40°). SG, secretory granule.

conditions (e.g. stress) by fortifying the barrier properties of the tissue (Robert et al 1979). The discovery that there are at least two isoforms of COX, with the constituitively active COX-1 being primarily present in the gastrointestinal tract and kidney and the inducible COX-2 being expressed at sites of inflammation, fortified the concept of Vane that NSAIDs primarily induce gastrointestinal injury by inhibiting the COX-1 isoform of the gastrointestinal tract and the biosynthesis of cytoprotective prostaglandins in the mucosa (Mitchell et al 1993; Masferrer et al 1994, Vane & Botting 1998). This led to the development of NSAIDs that selectively inhibit the COX-2 isoform (coxibs) and the blockbuster drugs rofecoxib (Vioxx) and celecoxib (Celebrex) with preclinical and clinical evidence of increased gastrointestinal safety (Masferrer et al 1994; Emery et al 1999; Laine et al 1999; Bombardier et al 2000; Silverstein et al 2000). However, the entire class of coxibs are presently under scrutiny due to questions concerning their improved gastrointestinal safety (Juni et al 2002; Hipisley-Cox et al 2005) and also to reports that they cause adverse effects to the cardiovascular system, which resulted in the withdrawal of both Vioxx and valdecoxib (Bextra) from the US market as recommended by the FDA (Mukherjee et al 2001; Bresalier et al 2005; Nussmeier et al 2005; Soloman et al 2005).

Although there is pre-clinical and clinical evidence that coxibs are less injurious to the gastrointestinal tract than less selective NSAIDs, there is also compelling evidence that the mechanism by which NSAIDs induce injury to the gastrointestinal tract is not entirely dependent on their ability to inhibit mucosal COX-1 activity. This controversial subject, which was the basis of a previous editorial (Lichtenberger 2001), will be briefly reviewed below. First, a number of in-vivo and in-vitro studies have demonstrated that the gastrointestinal injurious potential of a number of NSAIDs can be dissociated in time, dose and route of administration from their ability to inhibit COX-1 (Whittle 1981; Ligumsky et al 1982, 1983, 1990). Second, in studies by our laboratory and others it has been demonstrated that COX-1 knockout mice do not have ulcer disease (as would be predicted by Vane's concept), whereas the mice readily undergo gastrointestinal ulceration and bleeding when challenged with aspirin and other non-selective NSAIDs (Langenbach et al 1995; Darling et al 2004). Also, in a recent paper, we demonstrated that in COX knockout mice there is a statistical association between aspirin's ability to reduce surface hydrophobicity and induce gastric injury, whereas there was no such association between aspirin's ability to affect gastric mucosal prostaglandin concentration and the development of stomach haemorrhagic lesions (Darling et al 2004). Lastly, the preclinical and clinical evidence that COX-1 selective inhibitors are not injurious to the gastrointestinal tract and that both COX isoforms need to be blocked for mucosal damage to occur has made investigators reassess the entire concept originally proposed by Vane (Wallace et al 2000). As emphasized in an earlier publication, the ability of NSAIDs to topically injure the mucosa, as originally described by Davenport 40 years ago, may have greater importance than appreciated in more recent times.

Evidence supporting the concept that the interaction of NSAIDs with phospholipids is important in the drugs' mechanism of pathogenesis

A concept being evaluated by our laboratory and others is that one of the central mechanisms by which NSAIDs induce gastrointestinal injury is by chemically associating with membrane or extracellular phospholipids that provide important barrier properties to the tissue. In previous publications, biochemical and biophysical evidence has been presented to support this hypothesis with regards to NSAID-induced changes in phospholipid solubility, fluidity and membrane integrity (Lichtenberger et al 1995; Giraud et al 1999). In a recent paper, Ferreira et al (2005a, b) demonstrated that a number of NSAIDs share the ability to increase the fluidity of both synthetic and biological membranes, using steady-state anisotropic measurements. It also should be noted that in this and other studies, different NSAIDs had differing and in some cases contrasting

effects on membrane fluidity and order, which may relate to differences in the biophysical characteristics of the NSAID (pK_a, hydrophobicity), membrane or synthetic phospholipid (differences in head group, and length and level of saturation of fatty acid side-chains) and the probe used to assess fluidity and how its fluorescent properties are altered by the specific NSAID being studied (Giraud et al 1999; Ferreira et al 2005b). It has also been documented by our laboratory and others that NSAIDs can have a direct effect on cellular and synthetic membranes to induce alterations in permeability, which may be the basis of NSAID-induced cytotoxicity. This has been most clearly demonstrated in the studies of Tomisato et al (2004), who demonstrated that a number of NSAIDs induced the dose-dependent release of the fluorescent probe calcein from synthetic membranes composed of egg phosphatidylcholine, and in some cases (celecoxib), changes in membrane permeability were seen at micromolar concentrations.

In very recent studies reviewed below, we have acquired additional evidence that salicylate and perhaps other NSAIDs rapidly enter phospholipid membranes and induce pronounced changes in the membrane's thickness and micromechanical properties, which result in an increased susceptibility for the membranes to rupture and form pores or aqueous channels for protons and other noxious water-soluble agents to move from the lumen into the tissue (Zhou & Raphael 2005). It is our proposition that these NSAID-induced biophysical changes in the properties of phospholipid bilayers and monolayers may be critical in the pathogenic mechanism by which these drugs disrupt the mucosal barrier properties and induce gastrointestinal injury and ulceration. The rationale and evidence supporting this hypothesis is presented below.

Mechanical properties are important to the stability of the gastrointestinal mucosa

The mechanical properties of any material will determine a material's strength and ability to resist deformation. Lipid membranes are primarily characterized by two mechanical parameters: the elastic compressibility modulus and bending stiffness. These continuum parameters are a function of the membrane's chemical composition and are important in describing membrane stability. The compressibility modulus (K) characterizes the ability of the membrane to resist in-plane area changes resulting from forces that stretch or compress the membrane (Evans et al 1976, 2003; Israelachvili et al 1980). At the molecular level, this parameter arises from cohesive interactions between lipid molecules and correlates with the membrane's permeability. Because the gastrointestinal tract is constantly stretched and deformed, the lipid layer covering the gastrointestinal mucosa must be able to withstand these forces if it is to function as a permeability barrier. When a membrane is stretched in the plane of the bilayer, as the lipid bilayer covering the gastrointestinal tract is stretched during digestion, its elasticity determines how well the membrane can resist deformation and remain intact. A larger K for a material like gastrointestinal mucosa means this material is more resistant to stretching and more likely to function as a hydrophobic barrier. The bending modulus (k_c) describes the membrane's ability to resist out-of-plane bending and changes in curvature (Evans & Rawicz 1990; Zhou & Raphael 2005). The ability to bend is particularly important in the gastrointestinal mucosa because the bending pattern and the curvature of the lipid layer of the gastrointestinal mucosa are constantly changing in the course of forming complex rugae in the gastrointestinal tract and due to changes in gastrointestinal contractile activity. Moreover, the bending modulus is even more intimately correlated with membrane permeability, because (as explained below) it determines the likelihood that both transient and stable pores will form in the membrane.

To measure membrane mechanical properties, a special technique has been developed for deforming the membrane in a controlled manner and measuring the extent of deformation (Zhou & Raphael 2005). This micro-mechanical technique, micropipette aspiration, utilizes a pipette with a diameter of 8–10 μm to apply pressure onto the membrane. The resulting deformation of the membrane into the pipette can be visualized on a microscopic scale and recorded for later geometric analysis (Figure 2). When expanded, lipid molecules in the membrane are pulled apart. Because biomembrane stability depends upon the complex intermolecular interactions among lipids, the response to any perturbation can reflect the fundamental packing properties of the membrane.

Membrane mechanical properties reflect changes in the lipid–lipid interactions

It is well known that "oil and water do not mix". The physical reason for this is that hydrophobic forces arise from the unfavourable interactions between lipid acyl chains and water. These forces drive the self-assembly and contribute to the stability of biomembranes (Cevc & Marsh 1987). This hydrophobic force is cohesive and gives rise to a surface tension at the aqueous–lipid interface, which tends to pull lipid molecules together. The surface tension is opposed by a surface pressure caused mainly by the electrostatic interactions among lipid headgroups.

Both surface tension and surface pressure are determined by the geometry of the lipid molecule, such as the lipid packing area and the length and level of saturation of the fatty acid side-chains (Israelachvili et al 1980). When the membrane structure is perturbed by incorporation of amphiphilic molecules (Figure 3) such as salicylate, ibuprofen or other related NSAIDs, lipid–lipid interactions will be changed, thereby changing surface tension or the surface pressure. Traditionally, Langmuir troughs have been utilized to measure surface pressure of phospholipid monolayers, but this technique has significant limitations in modelling the properties of biological membranes under physiological conditions and after exposure to pharmaceuticals.

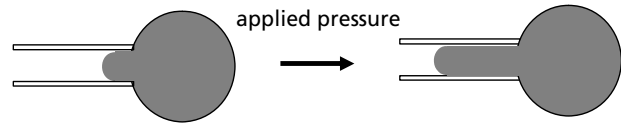

Figure 2 Schematic images of an aspirated vesicle at low pressure level and at high pressure. The dimensions are measured to calculate membrane tension.

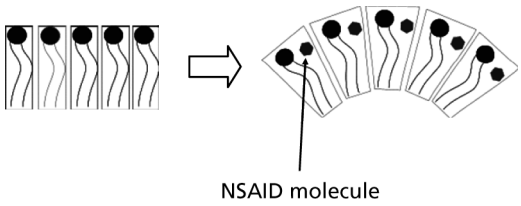

Figure 3 When amphiphilic molecules, such as NSAIDs, partition into the membrane, the packing shape of the lipids will be changed, ultimately altering the mechanical properties and stability of the bilayer.

The studies of the condensed phase membranes have been conducted using simple and highly versatile systems of unilamellar vesicles, such as giant unilamellar vesicles (GUVs). The vesicles can be synthesized by various methods, depending on the desired size of the vesicles. To synthesize GUVs, a variation on a method of electroformation developed by Angelova et al (1992) has been utilized, combined with the technique of micropipette aspiration (Figure 2). GUVs can be used to study membrane mechanics, determining parameters such as elasticity, bending stiffness and permeability. It is also possible to study the strength of the membrane by raising the aspiration pressure to a level that causes the membrane to lyse: the corresponding tension is referred to as the lysis tension. As described below, the membrane lysis tension is a function of the rate of pressure loading, and studying this relationship enables a detailed characterization of the nature of membrane pore formation.

This microaspiration technique has been applied to investigate effects of short-chain alcohols, bile acids and lysolipids on the membrane elasticity, bending stiffness and lysis tension (Zhelev 1998; Ly & Longo 2004). For large surfactants, such as bile acids and lysolipids, the effects on the elastic compressibility were attributed to tension-dependent partitioning (i.e., the incorporation of more molecules into the membrane as the membrane is stretched by increases in applied pressure). In addition, small surfactants are predicted to change the profile of the bilayer lateral pressure, which arises from the repulsive interactions between lipid acyl chains, leading to changes in membrane mechanical moduli and thickness (Cantor 1999).

Effects of salicylate on membrane mechanics

Our results indicate that salicylate has only a modest effect on the elasticity of membranes composed of 1-stearoyl-2-oleoyl-phosphatidyl-choline (SOPC) lipids whose structure is similar to the most abundant lipids found in the gastrointestinal mucosa (Zhou & Raphael 2005). SOPC is composed of two 18-carbon acyl chains, with one fully saturated and the other one monounsaturated. The microaspiration results showed that the compressibility modulus of SOPC GUVs was not affected within the range of physiological concentrations of salicylate that may be found in the gastrointestinal lumen (1–10 mM).

However, salicylate reduces the bending rigidity of an SOPC membrane in a dose-dependent manner (Zhou & Raphael 2005). In particular, 1 mM salicylate is able to decrease the bending stiffness by approximately 40%. As for any thin material, membrane elasticity and bending stiffness are related by the thickness of the membrane (Rawicz et al 2000). As the effects of salicylate on both the membrane elasticity and bending stiffness have been measured, salicylate's effect on membrane thickness can be predicted. It was found that salicylate reduces the thickness of the membrane in a dose-dependent manner (Zhou & Raphael 2005).

Salicylate induces membrane pore formation: characterization of membrane strength by dynamic tension spectroscopy

The micropipette method discussed above has also been utilized to study the strength of lipid membranes (Evans et al 2003). The failure of a thin amphiphilic material usually begins with the formation of a pore, which expands to cause the material to rupture. However, the formation of a membrane pore is preceded by the formation of a pre-pore, or a defect, because a lipid membrane is dynamic and fluid-like (Figure 4). During this pre-pore formation, lipids in the membrane can spontaneously tilt into a defect, and then tilt back to their original orientation normal to the bilayer plane (Evans et al 2003). As more energy is put into the membrane, the opening of the defect is enlarged to form a pore, which leads to the rupture of the membrane (Figure 4).

A recently developed technique, dynamic tension spectroscopy (DTS), explores the details of the process of membrane lysis (Evans et al 2003). During a typical DTS experiment, a GUV is aspirated into a micropipette. The pressure is applied at various speeds from vesicle to vesicle. The lysis tension of the membrane, which is the highest tension a vesicle can withstand, is dependent upon the speed of the pressure loading. The DTS indicates the correlation between lysis tension and (slope of the plot) the pressure loading rates and characterizes the pattern of the formation of defects and pores. All plots can be grouped into two categories, a high lysis tension region and a low lysis tension domain, which demonstrate the presence of transient defects and the formation of pores in the membrane, respectively (Evans et al 2003).

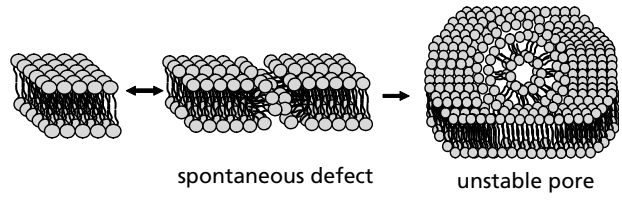

Figure 4 Lipids in a smooth membrane can momentarily tilt toward the bilayer core and spontaneously swing back to their original position, which is perpendicular to the plane of the bilayer, to form a defect. When more energy is put in, some of those lipids that momentarily tilt toward the bilayer core would keep the new orientation and form a pore. This pore will grow until a critical size is reached, which will cause the membrane to rupture. To form a pore in the membrane, lipids have to reorient to bend. The partitioning of salicylate molecules can thin the membrane and decrease the bending stiffness, thus inducing membrane pore formation.

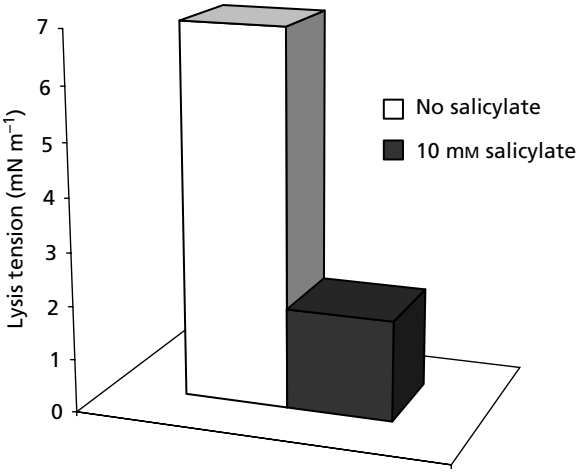

Figure 5 The dynamic tension spectroscopy (DTS) of SOPC when the pressure was applied slowly in the low tension domain and the pressure loading rates were held constant. The lysis tension for vesicles in 10 mM salicylate was shown to be much larger than that of vesicles with no salicylate. It also indicates that salicylate can potentially lower the energy barrier for forming a pore.

Figure 5 shows the effect of salicylate on the low tension region of a phospholipid bilayer member. In the presence of salicylate, the lysis tension lowers dramatically when the pressure loading rate is held constant. This indicates that salicylate, and perhaps other NSAIDs, can potentially lower the energy barrier for forming a pore. In contrast, for the high lysis tension domain, the tension versus time plot for SOPC is not significantly affected by the presence of salicylate, indicating that the NSAID has marginal effects on the formation of these membrane defects (data not shown) (Zhou & Raphael 2005). These results demonstrate that salicylate mainly influences the membrane rupture process by lowering the energy barrier of pore formation. Thus, salicylate and perhaps other NSAIDs stabilize membrane pores (Figure 4) to facilitate the back-diffusion of toxic chemicals in the lumen (e.g. H^+), which can lead to membrane rupture.

Consequences of NSAID–membrane phospholipid interactions

The mechanical measurements and DTS spectra together characterize a unified picture that salicylate induces the formation of a highly curved structure, such as a pore, by decreasing the thickness and the bending stiffness of the membrane. When the lipid area is spanned by insertion of salicylate molecules, the packing shape of the lipid molecule within the membrane is changed from an approximate cylinder to an inverted cone with the headgroup region being enlarged. The increase in the lipid packing area, and resulting freedom for lipid acyl chains to move around, causes the membrane to increase in fluidity and be reduced in thickness, if the volume of the membrane remains constant (Raphael et al 2000). A decreased bending rigidity means that it takes less energy to deform the membrane. For instance, formation of a pore whose discrete structure has a highly curved edge is made easier when it requires less energy to bend the lipids located around the pore. Thus, we hypothesize that there are two inter-related effects by which salicylate or other NSAIDs may contribute to an increase in membrane permeability and a consequent reduction in mucosal barrier characteristics. Firstly, NSAID intercalation in the bilayer increases the propensity for defects to form in the membrane, which could lead to the development of erosions and small increases in permeability. Secondly, salicylate and other NSAIDs may stabilize pores in the membrane, which would lead to an even higher permeability. Under acidic conditions, normally present in fasting gastric juice, this could ultimately lead to a total disruption of both extracellular and membraneous lipid layers protecting the gastrointestinal tract from acid back-diffusion and the development of a gastroduodenal ulcer and its associated complications (haemorrhage and obstruction).

This concept of salicylate's effect on membrane stability can help the design of new NSAID drugs. This view raises the possibility that the origin of a drugs' toxicity could be related to the molecules' amphiphilic structure and the ability to interact with lipids, not solely their ability to interact with enzymes (e.g. COX). This could point to a new direction for designing new NSAIDs that would have fewer side-effects on the gastrointestinal tract while retaining their therapeutic efficacy.

Conclusion

NSAIDs have been shown to induce remarkable changes in the properties of phospholipid membranes with regards to changes in hydrophobicity, fluidity, thickness, bending stiffness, permeability and pore formation. The new data suggest that NSAIDs like salicylate affect membrane stability, in addition to the inhibitory activity on COX. Therefore, NSAIDs that are mainly amphiphiles can be hypothesized to affect the stability of gastrointestinal mucosa by partitioning into both the extracellular and membrane phospholipid layers and inducing an alteration in their physical properties, which may ultimately lead to the formation of membrane pores in the mucosa to act as low resistance pathways for the back-diffusion of luminal acid. This concept may not only provide insight into how NSAIDs may topically injure the gastrointestinal mucosa by a mechanism independent from COX inhibition but also may provide a new direction to designing a novel class of NSAIDs that will have fewer side-effects while retaining their therapeutic activity.

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