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# Biomimicry of Surfactant Protein C

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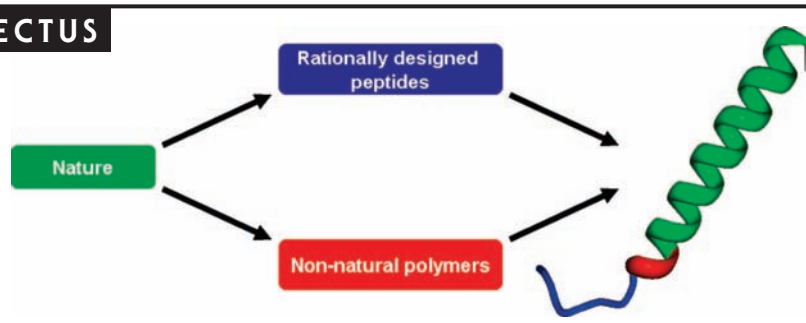
## CONSPECTUS




Since the widespread use of exogenous lung surfactant to treat neonatal respiratory distress syndrome, premature infant survival and respiratory morbidity have dramatically improved. Despite the effectiveness of the animal-derived surfactant preparations, there still remain some concerns and difficulties associated with their use. This has prompted investigation into the creation of synthetic surfactant preparations. However, to date, no clinically used synthetic formulation is as effective as the natural material. This is largely because the previous synthetic formulations lacked analogues of the hydrophobic proteins of the lung surfactant system, SP-B and SP-C, which are critical functional constituents. As a result, recent investigation has turned toward the development of a new generation of synthetic, biomimetic surfactants that contain synthetic phospholipids along with a mimic of the hydrophobic protein portion of lung surfactant.

In this Account, we detail our efforts in creating accurate mimics of SP-C for use in a synthetic surfactant replacement therapy. Despite SP-C's seemingly simple structure, the predominantly helical protein is extraordinarily challenging to work with given its extreme hydrophobicity and structural instability, which greatly complicates the creation of an effective SP-C analogue. Drawing inspiration from Nature, two promising biomimetic approaches have led to the creation of rationally designed biopolymers that recapitulate many of SP-C's molecular features. The first approach utilizes detailed SP-C structure–activity relationships and amino acid folding propensities to create a peptide-based analogue, SP-C33. In SP-C33, the problematic and metastable polyvaline helix is replaced with a structurally stable polyleucine helix and includes a well-placed positive charge to prevent aggregation. SP-C33 is structurally stable and eliminates the association propensity of the native protein. The second approach follows the same design considerations but makes use of a non-natural, poly-*N*-substituted glycine or "peptoid" scaffold to circumvent the difficulties associated with SP-C. By incorporating unique biomimetic side chains in a non-natural backbone, the peptoid mimic captures both SP-C's hydrophobic patterning and its helical secondary structure.

Despite the differences in structure, both SP-C33 and the SP-C peptoid mimic capture many requisite features of SP-C. In a surfactant environment, these analogues also replicate many of the key surface activities necessary for a functional biomimetic surfactant therapy while overcoming the difficulties associated with the natural protein. With improved stability, greater production potential, and elimination of possible pathogenic contamination, these biomimetic surfactant formulations offer not only the potential to improve the treatment of respiratory distress syndrome but also the opportunity to treat other respiratory-related disorders.

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## Introduction

Despite the seemingly simple building blocks from which they are constructed, proteins constitute enormously complex and powerful molecular tools. Utilizing a bottom-up design approach, Nature has engineered proteins to perform a wide range of life-essential tasks that participate in almost every biological process. Proteins are able to carry out such diverse functions because they adopt specific, compact conformations with high precision and fidelity, allowing exact three-dimensional positioning of the functional groups and facilitating the necessary interactions of the active site.

The diverse and precise bioactivities of proteins have made them coveted therapeutic agents, and they are currently being used in a variety of applications including in the treatment of diabetes and cancer and to combat infection. Because of their size, peptides have several advantages over small molecules and antibodies for therapeutic applications. Therapeutic peptides are large enough (10−50 amino acids) that their interaction with desired targets are more specific with lower toxicity profiles than other small molecules. Likewise, peptides are small enough to have improved storage/stability, tissue penetration, and immunogenicity over antibody-based therapeutics. Despite this, peptide-based therapeutics are only recently seeing more widespread use, largely because peptides are generally more difficult than small molecules to produce on a large scale, are rapidly cleared *in vivo*, and lack *in vivo* stability due to protease degradation.[1]

To date, the vast majority of engineered synthetic and recombinant peptide therapeutics are based upon sequences originating from natural sources, because these peptides have undergone natural selection for both efficacy and stability.[1] Recent advances in genomics and structural proteomics have led to a deeper understanding of how a protein's amino acid sequence lends itself to the formation of stable secondary structure elements such as α-helices, $\beta$-sheets, and turns and how these structural elements give rise to tertiary and quaternary structures of folded, biologically active proteins. This improvement in translating amino acid sequences into protein structures and related functions, while not complete and still developing, has led to the ability to prepare some peptide sequences *de novo* with predictable structures.[2] In addition, researchers are also expanding Nature's code with the inclusion of non-natural amino acids and, in some cases, use of totally synthetic polymeric scaffolds or "foldamers".[3] These novel structures are beginning to become increasingly efficacious and have the possibility to possess improved properties over native peptides, increasing the tools available to the protein scientist.

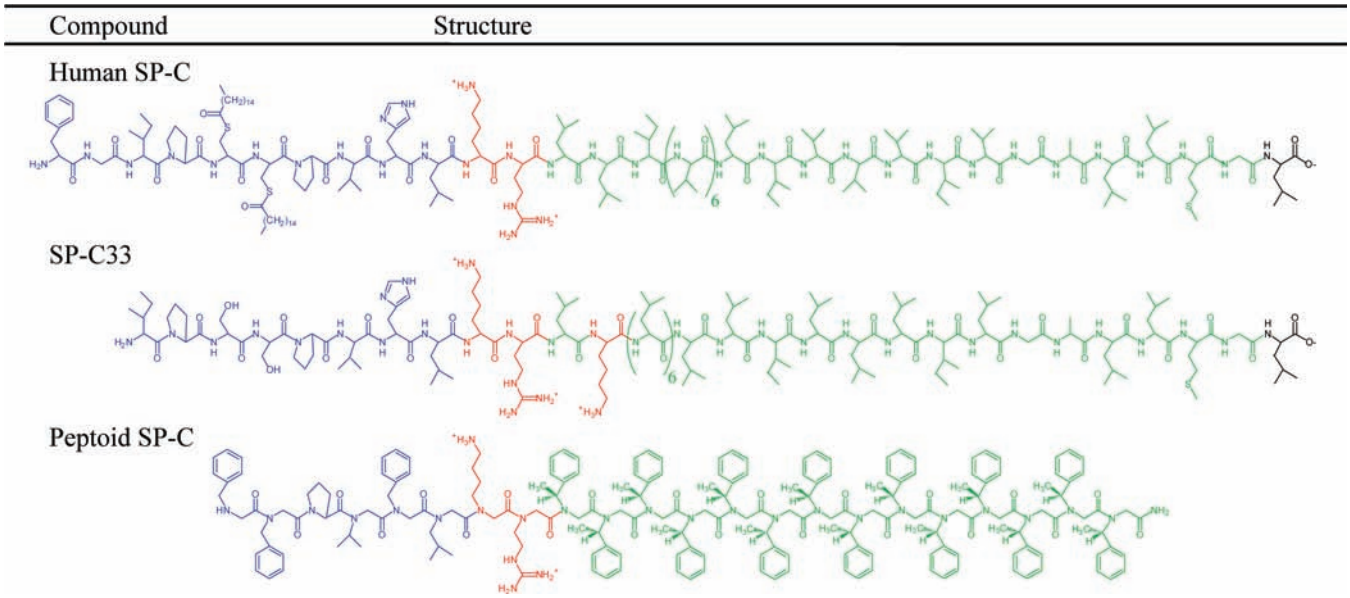
Researchers have developed novel sequences of amino acids, non-natural polymers, and hybrids of the two that can be efficiently produced with improved stability, bioavailability, and, in some instances, functionality in comparison to the mimicked peptide. One interesting application with clinical relevance concerns the mimicry of the predominantly helical, hydrophobic protein of the lung surfactant (LS) system, surfactant protein C (SP-C). SP-C, in addition to SP-B, is an important constituent of exogenous LS material for the treatment of respiratory distress syndrome (RDS). However, working with the natural protein and sequence-identical analogues is extremely challenging due SP-C's extreme hydrophobicity and metastable secondary structure.[4] These attributes make the isolation and storage of SP-C very difficult and may also contribute to the limited shelf life of therapeutic preparations containing the protein.[5] The utilization of recombinant and synthetic peptides as well as non-natural peptidomimetic approaches offer promise for overcoming these difficulties. Below, the requisite surface activities of a functional LS formulation and the key SP-C structural and molecular features are detailed, followed by ongoing approaches to mimic this difficult protein.

## Lung Surfactant and Respiratory Distress Syndrome

LS is a complex mixture of over 50 lipid species and four surfactant proteins that lines the alveolar air−liquid interface and is required for proper respiration, as its absence or dysfunction leads to severe respiratory disease.[6,7] No single component is solely responsible for LS's unique biophysical activity.[7,8] Phospholipids are the main constituent of LS, representing >80% of its mass, while neutral lipids and surfactant proteins each constitute approximately 10%.[8,9] In the airways, the lipid−protein mixture has three main properties that are essential for respiration: (1) rapid adsorption to the air−liquid interface of the alveoli, (2) dramatic reduction of the alveolar surface tension to near-zero values upon compression or exhalation, preventing collapse of the alveoli due to contractile forces of the alveolar liquid lining, and (3) rapidly respread to the air−liquid interface upon expansion or inhalation, reducing maximum surface tension and diminishing the work of breathing.[10]

An absence of functional LS in premature infants leads to the occurrence of neonatal RDS (nRDS). Without properly functioning LS, lung compliance is reduced and respiration is greatly impaired, ultimately resulting in alveolar collapse and

**CHART 1.** Structure of Native SP-C and SP-C Analogues[a]




[a] The chart shows how two classes of SP-C analogues utilize different techniques to mimic the various conserved regions. Colored residues signify the different conserved regions of SP-C: residues in the flexible *N*-terminal region of SP-C are blue, the basic residues are red, and the hydrophobic helical region is green.

suffocation without treatment. While nRDS was once a leading cause of infant mortality, it is now routinely treated by administration of animal-derived exogenous surfactant preparations into the lungs of afflicted individuals.[7] These surfactant replacement therapies (SRTs) consist of modified mammalian surfactant either extracted directly from animal lungs by lavage or isolated from homogenized lung tissue. Despite the efficacy of natural SRTs, there are some drawbacks associated with their use.[11] Natural preparations present a possibility of cross-species transfer of infectious agents as well as high production costs and batch-to-batch variability. In addition, SRTs may also be beneficial in the treatment of other respiratory-related disorders such as acute RDS; however, this requires significantly more material than current isolation techniques can supply.[12] To address these concerns, synthetic surfactants composed of synthetic phospholipids and surface-active chemical additives were developed. However, these synthetic preparations had reduced efficacies compared with natural surfactants and are no longer used, likely a result of the absence of the hydrophobic LS proteins, SP-B and SP-C, which are both critical constituents of functional LS.[13,14] Therefore, there has been increasing interest in the development of a third category of SRTs: biomimetic SRTs that utilize an entirely synthetic surfactant containing a mimic of the hydrophobic protein portion of LS that functions as well as the natural material and eliminates the concerns associated with animal-derived surfactants.[12,15] This endeavor requires a detailed understanding of the molecular interactions between the hydrophobic surfactant proteins and the LS phospholipids and how these interactions relate to surface activity.[16]

## Surfactant Protein C

Both SP-B and SP-C are key functional constituents of LS and are included in exogenous SRTs for the treatment of nRDS.[13,16] SP-B catalyzes the transport of surfactant to the air−liquid interface and assists in the folding and respreading of surfactant material during the respiration cycle. While many of SP-C's biophysical properties overlap with those of SP-B, SP-C is ubiquitously expressed in all mammals and other nonmammalian species that breathe air via alveolar-like lung structures and lacks sequence homology to any other known protein.[16] These observations suggest that SP-C's role in LS is both unique and necessary in addition to SP-B.

SP-C is synthesized in the airways by the alveolar type II cells and is secreted as a final 35 amino acid surface-active lipopeptide. The mature protein contains a high number of valine, leucine, and isoleucine residues as well as a dipalmitoylation modification near the *N*-terminus (Chart 1). The abundant hydrophobic residues, along with the palmitoyl motif, make SP-C one of the most hydrophobic proteins known to exist in any biological system.[7] SP-C's unique sequence is highly conserved among all species expressing the protein.[17]

The three-dimensional structure of SP-C in an apolar solvent has been determined by 2D-NMR, and the protein was found to lack any tertiary structure, while its secondary struc-

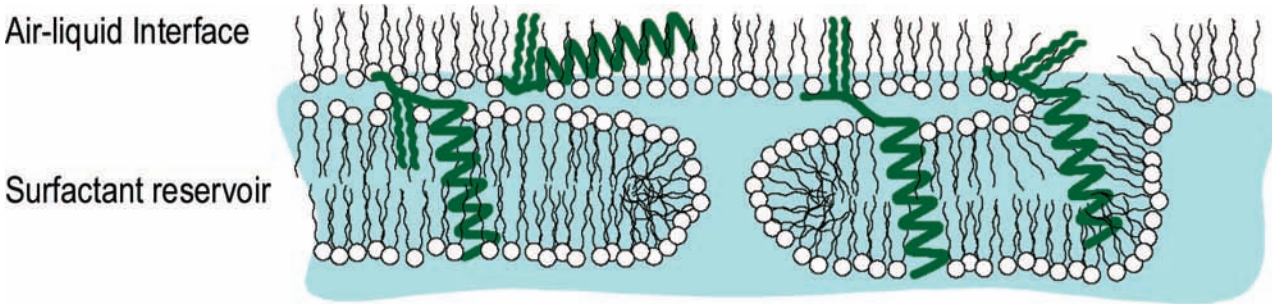



**FIGURE 1.** Hypothesized SP-C disposition in a LS film at the alveolar air−liquid interface. In a monolayer film, the SP-C helix is oriented nearly parallel to the air−liquid interface, while in a phospholipid bilayer, the SP-C helix aligns nearly parallel with the lipid acyl chains. The figure also shows how the palmitoyl chains of SP-C may act to anchor either the flexible *N*-terminal region in the same phospholipid film or to an adjacent layer, forming a surface-associated surfactant reservoir.

ture is dominated by a large α-helical region encompassing residues 9−34.[18] The length of this helical region, 37 Å, matches that of a fluid DPPC bilayer and within this region resides a 23 Å long, valyl-rich stretch of hydrophobic amino acids that also closely matches the alkyl chain length of a DPPC bilayer. The unstructured *N*-terminal region of the mature peptide was found to assume a flexible disordered orientation in solution, but more recent studies have suggested that this segment adopts an amphipathic conformation in the presence of phospholipid monolayers and membranes.[19,20]

SP-C's unique molecular characteristics make it ideally suited for interacting with LS phospholipids.[7] Strong hydrophobic forces and the lack of charged or bulky side chains cause SP-C's helical region to associate and interact with the interior of phospholipid acyl chains. In a DPPC monolayer, SP-C orientates at a 70° tilt relative to the interfacial plane and orients nearly parallel with the lipid acyl chains within a bilayer, maximizing interactions between the polyvalyl helix and the acyl chains.[21,22] The two positively charged residues, lysine and arginine, also promote binding to a phospholipid monolayer or bilayer through ionic interactions between the basic residues and the head groups of the anionic phospholipids.[23] The exact function of the palmitoylated cysteines is not completely known, but they appear also to contribute to SP-C's association with phospholipids, as well as in the possible maintenance of its helical structure.[22,24−26] A depiction of SP-C's believed disposition in a surfactant film is shown in Figure 1.

While only a minor component of LS, ∼1% (w/w), SP-C has a substantial effect on the surface activity of the LS phospholipids. The addition of SP-C to surfactant phospholipids dispersed in an aqueous subphase accelerates the adsorption and transfer of surfactant phospholipids from the subphase, rapidly forming a surfactant layer at the interface.[27] SP-C also promotes further binding of dispersed lipid vesicles to the newly formed surfactant layer, creating a surface-associated surfactant reservoir.[26]

Once at the air−liquid interface, SP-C acts as a molecular lever and hydrophobic lipid anchor during respiration.[21] Upon exhalation, the LS film is compressed causing a reduction of surface tension. This compression leads to the exclusion of some material from the interface, resulting in the formation of bilayer and multilayered structures of surfactant that remain associated with the interface.[28] This multilayer formation is significantly enhanced by the presence of the hydrophobic SP-C protein.[28] Figure 1 depicts how SP-C may facilitate the formation of three-dimensional surfactant structures. As the film is compressed, SP-C's helical region reorients, aligning its helix with the lipid acyl chains.[29] At the same time, a number of lipid molecules are also drawn out of the monolayer surface and inverted to form a retained local bilayer structure.[30] SP-C's helix as well as the palmitoylated cysteines are thought to assist in surfactant retainment, whereby the palmitoyl groups remain associated with the compressed surface film while the helix is anchored with the excluded phospholipids.[30,31] This surfactant reservoir could then readily be inserted upon expansion or inhalation. The SP-C-specific catalyzed multilayered structures are also believed to be responsible for SP-C's regulation of surfactant viscosity, which prevents surfactant outflow from the alveoli to the upper airways at very low surface tensions.[32]

Despite SP-C's many important biophysical properties, working with and studying the native protein is quite difficult because SP-C's polyvalyl helix is discordant, that is, it is composed of amino acids with a high propensity to form β-strands.[4] The SP-C helix is consequently metastable in solution and can spontaneously convert into β-sheet aggregates and amyloid fibrils that have inferior surface activity compared with the correctly folded helical protein.[33−35] The instabilities of SP-C pose serious challenges either in isolating the

native protein or in the creation of synthetic analogues that retain the SP-C's requisite α-helical structure and corresponding surface activity. As a result, researchers have employed a variety of unique strategies to overcome these problems by producing and mimicking SP-C in heterologous systems, synthetic peptide synthesis, and non-natural peptidomimetic materials.

## Synthetic SP-C Analogues

The high degree of SP-C sequence conservation and uniqueness suggests the main molecular and structural features of SP-C must be preserved to retain the full repertoire of SP-C-related functionality.[17] These features include SP-C's extreme hydrophobicity, its helical secondary structure, the flexible *N*-terminal region, and the two basic charged residues. The palmitoylation motif has also been shown to have significant *in vitro* functionality; however, the *in vivo* importance of this motif has yet to be definitively proven.[19,20,31] In addition, the resulting analogues must resist misfolding and degradation over time and be amenable to efficient, large-scale production.

**Recombinant SP-C Analogues.** The expression of SP-C in heterologous systems allows for more efficient, large-scale production of the troublesome protein than isolation of the native protein with the ability to introduce designed sequence variations, improving handling and efficacy.[36−38] A form of recombinant SP-C (rSP-C) is currently used in Venticute.[39] This rSP-C analogue from Nycomed shares a nearly identical sequence with human SP-C except that the palmitoylated cysteines are absent and have been replaced with phenylalanines to eliminate protein oligomerization.[38] The phenylalanine substitution makes this protein analogue similar to canine SP-C, which contains one phenylalanine at position 6 and preserves, to some extent, the hydrophobicity of the palmitoyl groups. The addition of palmitoyl chains in a similar analogue did not enhance *in vivo* efficacy of the rSP-C-based formulation.[36]

rSP-C has shown good efficacy, improving lung function in premature animal models of RDS.[38] In clinical trials for the treatment of ARDS resulting from both indirect and direct lung injury, Venticute has shown a positive effect on gas exchange but, ultimately, did not have a beneficial effect on long-term survival.[39] However, a posthoc analysis suggested that Venticute may have survival benefits in patients with direct lung injury. As a result, Venticute is currently being used in a phase III clinical trial for the reduction of mortality in patients with RDS resulting only from severe acute pneumonia or aspiration of gastric contents (NCT00074906). The forthcoming results of this study are highly anticipated by clinicians and researchers of the LS community.

**Synthetic SP-C Peptide Analogues.** SP-C's lack of a tertiary structure and relatively simple secondary structure make it feasible to mimic the native protein by synthetic peptides. This is especially enticing given the difficulties associated with isolation, purification, and storage of the native protein from natural or recombinant sources. Utilizing structural and biophysical information gained from the native protein, synthetic SP-C analogues have been created to mimic the key lipid-interaction domains.[5,10,40,41] These synthetically engineered peptides not only address some of the inherent difficulties associated with SP-C but also facilitate the production of sufficient quantities for use in a synthetic SRT. In fact, many of these polypeptide biomimetics have been found to have comparable *in vitro* activity as the native protein as well as favorable *in vivo* performance in animal models. However, the metastability of the helical region still remains a challenging problem in sequences that retain the polyvalyl helix.

To address the metastability of the polyvalyl helix, a novel series of valine-to-leucine substituted SP-C analogues have been created.[40,42,43] One of the first was SP-C(Leu), which is a nonacylated SP-C variant based upon the sequence of human SP-C but replaces the valine residues in the α-helical region with leucine residues and the palmitoylated cysteines with serines.[40] The substitution of the $\beta$-sheet-promoting valine residues with the α-helix promoting leucines of SP-C(Leu) results in an analogue that spontaneously forms an α-helical structure in solution, even after acid treatment, which can be produced efficiently on a large scale. When combined with a phospholipid formulation, SP-C(Leu) adopted a transbilayer orientation and showed favorable *in vitro* surfactant spreading properties, similar to the native protein, reducing both minimum and maximum surface tensions during dynamic cycling. However, in an animal model of nRDS, the biomimetic formulation offered only a modest improvement in lung function.[40] This was due to unwanted oligomerization of the polyleucine helix at high concentrations, caused by leucine zipper-like association.

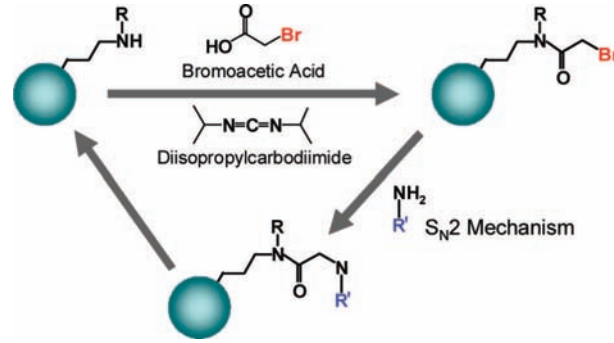
To circumvent this problem, SP-C(LKS) was created in which three evenly spaced leucine residues in the helical region were replaced with lysine residues.[43] The location of the positive charges around the helical circumference prevented self-association and when combined with phospholipids, exhibited rapid adsorption and favorable modulation of surface tension during dynamic cycling.[43] However, SP-C(LKS) was unable to produce sufficient improvement of lung function *in vivo*. The reason for the poor *in vivo* performance is not well understood, but the introduction of the positively charged lysine res-

idues in the nonpolar helix may either disrupt the region's hydrophobic interaction with the lipids or the SP-C dipole moment.[42]

The latest generation of the leucine-based synthetic peptide analogues, SP-C33, utilizes specific design features from both SP-C(Leu) and SP-C(LKS) to overcome the solubility and aggregation problems of SP-C(Leu) with improved *in vivo* performance relative to SP-C(LKS).[42] SP-C33 is similar to the other valine to leucine substituted analogues but contains an *N*-terminal region that is truncated by two residues to increase synthesis efficancy and, most notably, replaces Leu14 with lysine to prevent peptide oligomerization while not disrupting the hydrophobicity of the aliphatic helix. This analogue was found to prevent oligomerization and improve the dynamic surface activity of a lipid formulation.[42] When used to treat premature newborn animals, a SP-C33-containing formulation was as effective in increasing inspiration volumes as a modified natural surfactant preparation that contained both SP-B and SP-C.[44] Surprisingly, despite the demonstrated *in vitro* importance of the SP-C palmitoyl groups, an acylated SP-C33 analogue did not enhance the *in vivo* performance.[42] Airway openness at end expiration was lower in animals treated with SP-C33 than those treated with a natural SRT, suggesting that other components, such as SP-B, are necessary to maintain alveolar stability.[13] Despite this difference, the synthetic peptide is as effective as native SP-C in improving surface properties of lipids *in vitro* and *in vivo* and represents a successful engineering approach to overcome the numerous difficulties associated with the native protein.

**Peptoid Analogues.** In an alternative approach to creating recombinant or synthetic peptide analogues, recent work has been aimed at the development of a new category of synthetic surfactant protein mimics utilizing poly-*N*-substituted glycines or "peptoids".[3] Peptoids have close structural similarity to peptides, but have their side chains appended to the amide nitrogens rather than to the α-carbons. This feature renders peptoids essentially invulnerable to protease degradation, making them more biostable than peptides and reducing specific recognition by the immune system.[45] So far, studies have shown peptoids to induce only very low-level antibody response, and certain sequences have been found to be bioactive, nontoxic, and nonimmunogenic.[46,47] Despite the achirality of the *N*-substituted glycine backbone and its lack of hydrogen bond donors, peptoids are able to adopt extraordinarily stable chiral helices. When substituted with α-chiral, sterically bulky side chains, steric and, in some instances, electronic repulsion between adjacent residues cause peptoids to adopt a secondary structure similar to a polyproline type I

**SCHEME 1.** Submonomer Synthesis Scheme for Peptoidoligomers.




helix with approximately three residues per turn and a pitch of ~6 Å.[48,49] These helical structures are extremely stable and do not appreciably denature over time because their formation does not depend on hydrogen bonding along the backbone.[50] Peptoids also have the added advantage of being relatively easy and cost-effective to synthesize compared with peptides.[3] Utilizing a solid-phase, submonomer synthesis approach (Scheme 1), peptoids up to 50 residues in length can be synthesized in high yield, with coupling efficiencies comparable to those of Fmoc peptide synthesis (>98.5%). Efficient synthesis and the ability to form stable helical structures make peptoids an excellent platform for mimicry of bioactive molecules that rely on helical structure for proper function, such as the hydrophobic protein SP-C.

Utilizing design strategies similar to those that were used in the design of recombinant and synthetic peptide analogues of SP-C, a variety of nonacylated, sequence-specific peptoid analogues of SP-C(5−32) have been developed.[51,52] The sequences of the biomimetic analogues were designed to emulate the key structural features of the native protein, with a hydrophobic, α-chiral helical region, mimicking SP-C's helix, and an *N*-terminal amphipathic achiral region containing side chains that are largely analogous to those present in human SP-C. To gain a greater understanding of the structural requirements in peptoid-based analogues of SP-C, systematic alterations were introduced in the sequences.[51] Two main classes of mimics were created, containing either α-chiral aromatic- or α-chiral aliphatic-based helices that contained 8, 11, and 14 residues to determine both the side chain preference and the optimal helix length for a peptoid-based mimic of SP-C. Circular dichroism spectroscopy showed that the analogues were predominantly helical in solution with the longer mimics having the highest helical propensity. In addition to being highly helical, the analogues were stable and not prone to misfolding in solution over time.

When combined with a synthetic phospholipid formulation, the peptoid-based SP-C mimics all showed improvement

in surface activity over the lipid system alone; however, the mimics containing the more rigid aromatic helix yielded superior surface activity in comparison to the more biomimetic aliphatic-based mimics. The optimal surface activity of the SP-C mimics was also dependent on the presence of a helical region of 14 peptoid monomers in length (~28 Å), which is similar in length to the polyvalyl region of native SP-C (26 Å). In a lipid film, the aromatic-based mimic altered the surface pressure−area isotherms (obtained on a Langmuir−Wilhelmy surface balance) in a manner similar to a synthetic SP-C peptide. Film morphology studies revealed that the presence of the aromatic-based mimic but not the aliphatic-based mimic resulted in the nucleation of bright vesicle domains, likely three-dimensional structural changes in the film, at higher surface pressures, similar to SP-C. In a pulsating bubble surfactometer, the aromatic-based mimic also accelerated surfactant adsorption to the air−liquid interface as well as greatly reduced the minimum and maximum surface tensions, all of which properties were similar to the SP-C peptide.

These results were somewhat surprising, because the native protein contains only aliphatic side chains in the helical region; however, it is believed that the aromatic monomers more effectively constrain the peptoid backbone into the polyproline type I like helical conformation because it exhibits both steric repulsions between bulky side chains and electronic repulsions between aromatic $\pi$ and carbonyl lone-pair electrons.[48] This indicates that it is more important to capture the extreme hydrophobicity and highly helical structure of SP-C rather than its exact side chain chemistry, which is consistent with previous studies of peptide analogues.[53]

Given these properties, peptoid-based mimics of SP-C seem very promising, since they exhibit *in vitro* biophysical activities highly similar to that of the SP-C peptide. It was also found that the same design considerations occurring in SP-C peptide variants are applicable in the development of peptoid-based mimics of SP-C. Therefore, the results observed seem to provide a basis for the development of a peptoid-based biomimetic SRT for the treatment of RDS or other medical applications, and this work is ongoing.

## Concluding Remarks

While current SRT formulations are highly efficacious in the treatment of nRDS, there are still drawbacks associated with their use, which has prompted researchers to devise synthetic sources of the surface-active material, specifically creating mimics of the hydrophobic surfactant proteins, SP-B and SP-C. The importance of SP-C as a functional constituent of a biomimetic SRT is evidenced not only by its inclusion in natural exogenous surfactant formulations, but also by its ubiquitous expression in mammalian airways and its uniqueness, lacking any known sequence homologues. However, isolation and storage of either the native SP-C protein or sequence-identical analogues is extraordinarily difficult due to its extreme hydrophobicity and metastable secondary structure. As a result, researchers have drawn inspiration from Nature in redesigning the naturally selected protein to overcome these difficulties. Utilizing detailed structural information and biophysical insight about native SP-C, coupled with protein folding preferences, investigators have created a number of novel peptide and peptoid-based analogues that mimic SP-C's key molecular and structural motifs but, because of their side chain chemistry, their unique backbone structure, or both, reduce or eliminate many of the difficulties associated with the native protein and sequence identical analogues. Utilizing synthetic approaches, it is also conceivable that future SP-C analogues may contain other non-natural sequence modifications that further improve stability and activity in comparison to the native and recombinant species. Further improvements in biomimetic SRT efficacy will also likely involve the inclusion of a SP-B mimetic because both constituents play a critical role in surfactant homeostasis. With improved stability, greater production potential, and elimination of possible pathogenic contamination, not only do biomimetic SRTs offer the potential to improve the treatment of nRDS, but they may also see wider application in the treatment of other more prevalent respiratory-related disorders where greater quantities of the surface-active material are required, such as in the treatment of acute RDS.

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BIOGRAPHICAL INFORMATION

**Nathan Brown** received a B.S. in chemical engineering from Iowa State University in 2001. He then attended Northwestern University, obtaining his Ph.D. in 2008 under the advisement of Professor Annelise Barron. His research focused on structure−activity relationships of surfactant protein C peptoid analogues. He is currently working for Baxter Healthcare.

**Jan Johansson** received an M.D. and Ph.D. from Karolinska Institutet in Stockholm in 1991. He then conducted postdoctoral work

at the ETH Zurich during 1992−1993. Since 2002, he has been a professor of Medical Biochemistry at the Swedish University of Agricultural Sciences in Uppsala. His research interests include the structure and function of prosurfactant protein C, development of synthetic surfactants, and studies of antiamyloid strategies and biomimetic silks.

**Dr. Annelise Barron** received her Ph.D. in chemical engineering in 1995 from the University of California, Berkeley, and a B.S. cum laude in chemical engineering in 1990 from the University of Washington in Seattle. She was an NIH-NRSA postdoctoral fellow in pharmaceutical chemistry at the University of California, San Francisco. She joined the chemical engineering faculty at Northwestern University in 1997, rising to the level of tenured full professor, and in 2007 accepted a tenured associate professorship in Bioengineering at Stanford. She was awarded the Presidential Early Career Award for Scientists and Engineers (1999), the Beckman Young Investigator Award (1998), and the Camille and Henry Dreyfus Teacher-Scholar Award (2003). She has authored more than 75 peer-reviewed research publications. Dr. Barron served from 2005 to 2007 as a member of the Advisory Council to the NIH Director, Dr. Elias Zerhouni, and currently is a permanent member of the NIH's Synthetic and Biological Chemistry Study Section. Dr. Barron directs a group of 29 Ph.D. students and postdoctoral associates involved in diverse research projects at the interface of biotechnology, polymer science, and medicine. About half of her group's research is focused on developing novel materials and strategies for high-throughput DNA sequencing and genotyping by microchip electrophoresis. The other half of her group creates and studies novel stable peptide analogs that mimic bioactive protein domains and have promising therapeutic applications.

## FOOTNOTES

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