RESEARCH ARTICLE

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Tailoring Butyl Methacrylate/Methacrylic Acid Copolymers for the Solubilization of Membrane Proteins: The Influence of Composition and Molecular Weight

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Low-molecular weight (MW) amphiphilic copolymers have been recently introduced as a powerful tool for the detergent-free isolation of cell membrane proteins. Herein, a screening approach is used to identify a new copolymer type for this application. Via a two-step ATRP/acidolysis procedure, a 3 × 3 matrix of well-defined poly[(butyl methacrylate)-*co*-(methacrylic acid)] copolymers (denoted BMAA) differing in their MW and ratio of hydrophobic (BMA) and hydrophilic (MAA) units is prepared. Subsequently, using the biologically relevant model (T-cell line Jurkat), two compositions of BMAA copolymers are identified that solubilize cell membranes to an extent comparable to the industry standard, styrene-maleic acid copolymer (SMA), while avoiding the potentially problematic phenyl groups. Surprisingly, while only the lowest-MW variant of the BMA/MAA 2:1 composition is effective, all the copolymers of the BMA/MAA 1:1 composition are found to solubilize the model membranes, including the high-MW variant (MW of 14 000). Importantly, the density gradient ultracentrifugation/sodium dodecyl sulfate-polyacrylamide gel electrophoresis/Western blotting experiments reveal that the BMA/MAA 1:1 copolymers disintegrate the Jurkat membranes differently than SMA, as demonstrated by the different distribution patterns of two tested membrane protein markers. This makes the BMAA copolymers a useful tool for studies on membrane microdomains differing in their composition and resistance to membrane-disintegrating polymers.

1. Introduction

Membrane proteins play a prominent role in biological systems and represent a major group of pharmacological targets; however, studies on their structural and functional aspects are challenging, particularly due to their destabilization outside of their native lipid environment.[1–3] Until recently, detergents were almost exclusively used to isolate membrane proteins. However, during the isolation, the protein is largely stripped of the surrounding lipids, which might impact on the protein structure and function.[4] In 2009, the use of low-molecular-weight (MW) styrene-maleic acid (SMA) copolymers for detergent-free membrane protein solubilization was reported.[5] SMA copolymers cut the membrane into nanodisc-shaped particles (sometimes called SMA lipid particles, SMALPs) containing membrane proteins embedded in native-like lipid environment.[2,5,6] A number of proteins have been successfully isolated and characterized using SMA.[7–10]

Unfortunately, SMA copolymers also suffer from certain limitations. First, the variants exhibiting the best solubilization performance, i.e., these with the 2:1 and 3:1 styrene/maleic acid ratio, are derived from styrene/maleic anhydride copolymers produced industrially via free-radical polymerization in a continuous stirred tank reactor (CSTR),[11] which enables achieving the desired composition. However, the products are still highly disperse with respect to MW,[12,13] with a potentially negative impact on the nanodisc homogeneity and extraction efficiency.[12,14] Further, the strong perturbation of the phospholipid bilayer inflicted by SMA's phenyl groups may, in some cases, affect the structure and function of encapsulated proteins.[15] In addition, the UV absorbance of styrene units limits the study of the reconstituted proteins by certain spectroscopic techniques,[16] and the copolymers precipitate at acidic pH and at biologically relevant concentrations of divalent cations (Ca^{2+}, Mg^{2+}) due to the interactions of the maleic acid carboxyls.[13] To address some of these shortcomings, better defined SMA copolymers were prepared by various approaches,[12,17–20] and a number of derivatives of the original

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SMA structure were also introduced.[21–28] Apart from SMA and SMA-like copolymers, several other types of amphiphilic copolymers for membrane protein isolation were developed.[29] These included diisobutylene/maleic acid copolymers (DIBMA) and their derivatives,[16,30–33] alkylamine-modified poly(acrylic acid) (APAA),[34] methacroylcholine chloride/butyl methacrylate copolymers (PMA),[35] acrylic acid/styrene copolymers (AASTY),[14,36] modified inulin,[37,38] methylstilbene/maleic acid copolymers (STMA),[39] and cycloalkylamine-modified poly(acrylic acid) (CyclAPol).[40] In addition, in a very recent comprehensive study, Kopf et al. presented a number of SMA derivatives with diverse substitution mainly at the aromatic ring, as well as acrylic acid copolymers with substituted styrenes.[41] Finally, new amphiphilic small molecules (nonpolymeric but of higher MW) for membrane protein solubilization have been introduced recently, further blurring the boundary between the originally used detergents and the newly developed (mostly low-MW) polymers.[42,43]

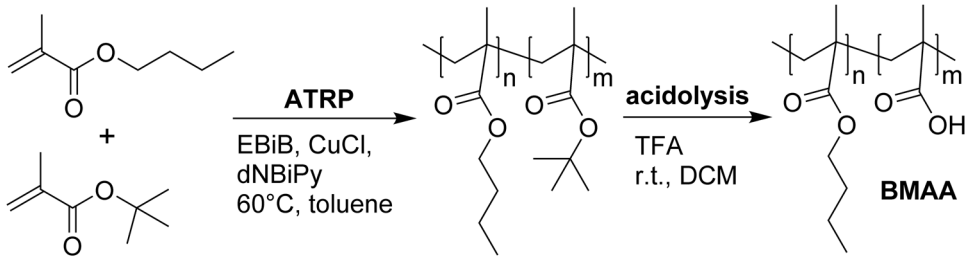
Despite the considerable progress made, the new copolymers still share some of the drawbacks of SMA mentioned above, e.g., they incorporate the potentially problematic styrenic or maleic acid moieties or show broad MW distribution. The importance of the latter parameter is being increasingly recognized because the MW (or chain length) of the copolymer is known to impact on the material's membrane solubilization efficiency and stability of the formed nanodiscs.[14,17] For this reason, reversible addition-fragmentation chain-transfer (RAFT) polymerization, that allows the preparation of low-dispersity polymers, has been applied in several recent studies to control the MW.[12,14,19,31,41] Another key property of the successful amphiphilic membrane-solubilizing copolymers is the proper balance between their hydrophilic and hydrophobic parts, where even small changes to the copolymer composition can impact significantly on the material performance in solubilization assays.[14] Moreover, the distribution of the different monomeric units within the polymeric chain also plays an important role, with compositional drift shown to be detrimental in some cases.[12,17] While the toolbox of the polymers suitable for disintegration of biological membranes and biochemical studies on native membrane proteins is expanding steadily, its extent is still rather limited. The recent finding that the type of polymer used for membrane protein solubilization can impact on the protein conformation[44] makes the search for new polymeric variants even more important. We hypothesized that a number of other copolymers could exhibit the desired membrane solubilization properties if the key copolymeric characteristics are adequately fine-tuned within a certain range. To demonstrate the validity of this approach, we used here atom transfer radical polymerization (ATRP) combined with selective acidolysis to create a library of well-defined statistical copolymers of butyl methacrylate (BMA) and methacrylic acid (MAA), termed BMAA, differing in their composition (ratio of hydrophilic and hydrophobic units) and MW. Using a biologically relevant cell model (human T cell line Jurkat), we identified the copolymers suitable for membrane solubilization, and we discuss the impact of the copolymer parameters on the solubilization process. Our results show a simple pathway for revealing new types of cell membrane solubilizing polymers.

2. Results and Discussion

2.1. Synthesis of BMAA Copolymers

ATRP is an important reversible-deactivation radical polymerization (RDRP) method providing well-defined (co)polymers of predetermined molecular weights,[45] representing thus a viable alternative to the RAFT method used so far in the synthesis of defined copolymers for cell membrane solubilization. One of the advantages of the ATRP approach is that it avoids the incorporation of an aliphatic chain, present in some of the widely used RAFT agents, into the copolymer, which could potentially influence the copolymer's performance in membrane solubilization assays, unless the RAFT end group is removed during the copolymer preparation.[17,19,20] Herein, we conveniently applied a two-step procedure to synthesize the target BMAA copolymers (**Scheme 1**). In the first step, BMA and *tert*-butyl methacrylate (tBMA) were copolymerized via ATRP, using ethyl 2-bromoisobutyrate (EBiB) as an initiator, CuCl as a catalyst, 4,4′-dinonyl-2,2′-bipyridine (dNbpy) as a ligand, and toluene as a solvent at 60 °C. To the best of our knowledge, the copolymerization parameters of this comonomer pair have not been established previously. Nevertheless, the comonomers were expected to copolymerize in a highly random fashion due to their high structural similarity, with the copolymer composition closely following the feed composition irrespective of monomer conversion. We used three different BMA/tBMA ratios in the feed (≈2:1, 1:1, and 1:2) and three different (total) monomer/initiator ratios (20, 80, and 160) to obtain nine copolymers that were, in the second step, subjected to selective acidolysis of the tBMA units using trifluoroacetic acid in dichloromethane.[46] This procedure furnished a 3 × 3 matrix of BMAA copolymers differing in their MW and the ratio of hydrophobic and hydrophilic monomeric units.

Table 1 summarizes the results of the copolymer preparation. The individual BMAA copolymers were assigned codes indicating the approximate BMA/tBMA (BMA/MAA after aci-



Scheme 1. Two-step preparation of BMAA copolymers.

Table 1. Preparation of the library of BMAA copolymers.

Code[a)]	[BMA]:[tBMA][b)]	Time [h]	Conversion[c)]		$M_{n,BMA\text{-}tBMA}$[d)]	$Đ_{BMA\text{-}tBMA}$[d)]	$M_{n,BMAA}$[e)]
			BMA	tBMA			
BMAA-2:1-L	13:7	4	0.91	0.94	4100	1.18	3500
BMAA-1:1-L	10:10	6	0.93	0.95	4300	1.22	3500
BMAA-1:2-L	7:13	4	0.88	0.92	3600	1.22	2700
BMAA-2:1-M	53:27	9	0.76	0.81	8000	1.19	7000
BMAA-1:1-M	40:40	9	0.67	0.70	9000	1.16	7000
BMAA-1:2-M	27:53	9	0.72	0.77	8000	1.18	6000
BMAA-2:1-H	107:53	33	0.80	0.83	15 000	1.19	13000
BMAA-1:1-H	80:80	33	0.85	0.88	17 000	1.16	14 000
BMAA-1:2-H	53:107	33	0.79	0.84	16 000	1.17	12 000

[a)] The code reflects the targeted BMA/MAA unit ratio in the final copolymer and also the targeted MW (L: low, M: medium, H: high); [b)] Molar ratios of the comonomers in the feed relative to the initiator (EBiB). Other conditions: [EBiB]:[CuCl]:[dNbpy] = 1:1:2, monomers/toluene = 1:1 (v/v), 60 °C; [c)] Conversion of the individual comonomers as determined by the ^{1}H NMR analysis of the crude polymerization mixture at the end of the experiment; [d)] Values for the poly(BMA-*co*-tBMA) copolymers determined by SEC with poly(MMA) calibration (apparent MW values); [e)] Values for the final BMAA copolymers calculated from $M_{n,BMA\text{-}tBMA}$ based on the feed composition and assuming the complete acidolysis of the tBMA units.

dolysis) ratio and also their relative MW (L: low, M: medium, H: high). Monomer conversions were calculated based on ^{1}H NMR analyses of crude polymerization mixtures (see a typical spectrum in Figure S1, Supporting Information, and the accompanying discussion for details). In all cases, this analysis revealed only marginal differences in the conversions of the individual comonomers irrespective of the feed composition and the reaction stage (e.g., both at ≈70% and ≈95% conversion), which appears to be consistent with our initial expectations about the random character of the copolymerization of BMA and tBMA. This can be seen as an advantage of the two-step preparation of the BMAA copolymers employed here because the possible alternative route, consisting in direct BMA and MAA copolymerization via RAFT, may yield less uniform ("blocky") copolymeric chains.[47] A size exclusion chromatography (SEC) analysis, performed using a poly(methyl methacrylate) calibration, revealed that the prepared poly(BMA-*co*-tBMA) copolymers were very well defined (dispersity under 1.25), with the relative M_n values approximately doubling in the S-M-L variant series (**Figure 1** and Table 1). Using the known feed compositions, we recalculated these values to obtain relative MWs of the final BMAA copolymers, arriving to M_n values of roughly 3000 for the L-variants, 7000 for the M-variants, and 13 000 for the H-variants (Table 1). Figure S2 (Supporting Information) shows a typical ^{1}H NMR spectrum of the final BMAA copolymer confirming the complete acidolysis of tBMA units as indicated by the absence of the intensive signal of *tert*-butyls at ≈1.45 ppm.

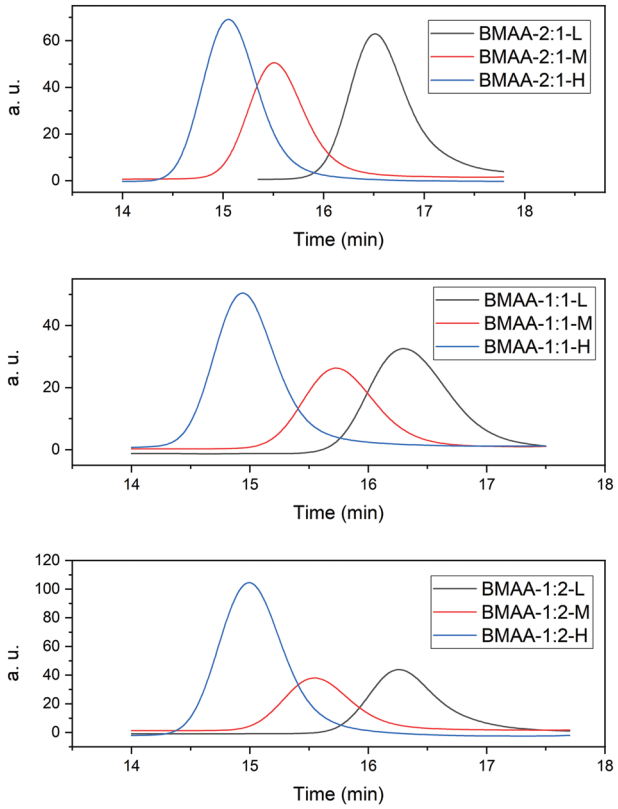


Figure 1. SEC elugrams of the synthesized poly(BMA-*co*-tBMA) copolymers. The copolymers of the same composition are grouped together to allow for relevant MW comparison. For clarity, the traces are denoted using the coding of the final BMAA copolymers.

2.2. Biochemical Studies

In literature, interactions of newly introduced copolymers with lipid membranes are often examined using artificial membranes or liposomes of a well-defined composition.[13] Nevertheless, it was shown that these model systems might not be necessarily representative of the copolymers' performance in assays using actual cell membranes.[12] As we believe that the results of works similar to the present one should be ultimately used in biological studies, we opted here for studying the relative performance of the BMAA copolymers by using the generally more relevant model of whole cells and cell membranes. For this purpose, we used human red blood cells (erythrocytes) and also the human

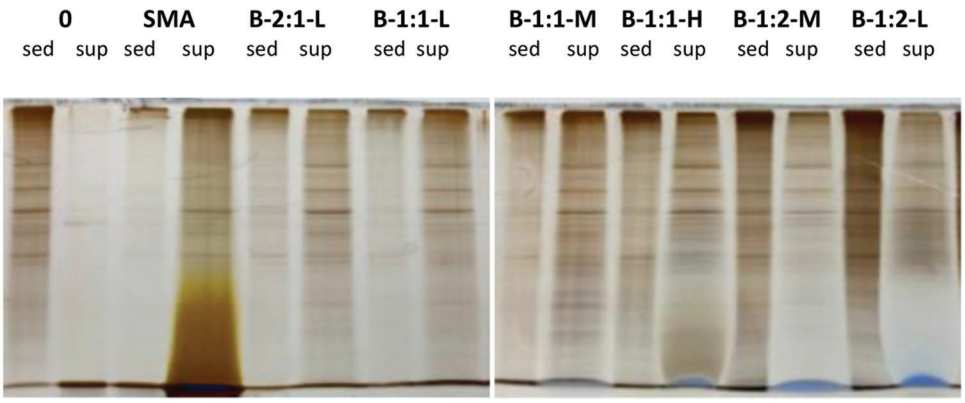


Figure 2. SDS-PAGE analysis of Jurkat T-cell membranes solubilized by selected BMAA copolymers. The cell membranes were treated with the lysis buffer containing 1% of a copolymer, and the insoluble materials were removed by centrifugation; the stained bands correspond to the most abundant proteins, separated approximately according to their MW. The broad intensely stained zone at the bottom of the SMA supernatant lane corresponds to free SMA molecules migrating with the front and fixed by the staining procedure. Sed: sediment, sup: supernatant; 0: negative control without any copolymer. The codes of the BMAA copolymers are shortened for clarity.

immortalized T-cell line Jurkat (derived from a T-cell leukemia) that has been one of the most widely used immunologically relevant cell lines for more than 40 years.

The prepared library of BMAA copolymers includes materials of rather different overall polarity and MW, with both these factors potentially impacting on the copolymer solubility in aqueous media. Therefore, before proceeding to the biochemical studies, we tested the copolymer solubility in the employed medium (0.02 M Tris-HCl buffer of pH 8.2, containing 0.1 M NaCl). We found most of the copolymers to be readily soluble at the target 1% concentration when pH was adjusted to 8.2, with the exception of BMAA-2:1-M and BMAA-2:1-H that were almost insoluble under the standard conditions and even after raising the pH over 9. This can be ascribed to the high content of the hydrophobic BMA units as well as the increased MW of the copolymers. Therefore, further screening was performed with the remaining seven BMAA copolymers.

To obtain an initial insight into the BMAA copolymers interactions with the two model membranes, we first used whole cells. Interestingly, none of the applied BMAA copolymers lysed erythrocytes; only SMA, that was used as a control, did so within seconds (data not shown). We ascribe this observation to the differences in the composition of the studied copolymers and also of the model membranes. Since both BMAA and SMA bear carboxylates as the polar (charged) components, the different performance of the two copolymer types may result from the character of the hydrophobic monomeric units. Presumably, the comparatively cholesterol-rich erythrocyte membrane is resistant to the attack by the flexible hydrophobic groups of BMAA copolymers (butyls of BMA units) while being susceptible to the more rigid phenyl groups of SMA.[15] On the other hand, visual inspection confirmed that six of the seven studied BMAA copolymers lysed the Jurkat T cells. The sole exception was the high-MW variant of the comparatively polar 1:2 series (BMAA-1:2-H). The six copolymers that passed the whole cell test (BMAA-2:1-L, BMAA-1:1-L, BMAA-1:1-M, BMAA-1:1-H, BMAA-1:2-L, and BMAA-1:2-M) were then applied to the solubilization of isolated Jurkat T-cell membranes. The isolated membranes represent a comparatively cleaner system where the interference with cytoplasmic and nuclear biopolymers is less probable. From the six tested copolymers, only BMAA-2:1-L, BMAA-1:1-L, BMAA-1:1-M, and BMAA-1:1-H solubilized the cell membranes in an extent comparable to SMA, as confirmed by a sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) analysis; the remaining two polar variants (BMAA-1:2-L and BMAA-1:2-M) were found to be inefficient (**Figure 2**). These results indicate that the equimolar ratio of BMA and MAA units in the BMAA copolymers represents the optimal balance between the hydrophobic and hydrophilic counterparts, which is seen as the key requirement for successful solubilization of cell membranes.[48] Notably, this ratio is the same as the ratio of the hydrophobic (phenyl) and hydrophilic (carboxylate) units within the most widely used SMA variant, SMA 2:1. Most importantly, the BMAA copolymers of the 1:1 composition maintain considerable membrane solubilization power up to the rather high MW of ≈14 000 while the high-MW variants of SMA were previously found to be significantly less effective than the low-MW counterparts,[49] and most of other polymers developed for cell membrane solubilization are also of low MW, with the zwitterionic zSMA (MW of up to 44 000) being a rare exception.[22] Surprisingly, the low-MW variant of the more hydrophobic BMAA composition (BMAA-2:1-L) solubilized the Jurkat cell membranes virtually to the same extent as the corresponding 1:1 variant (BMAA-1:1-L) whereas the higher-MW copolymers of the 2:1 composition (BMAA-2:1-M and BMAA-2:1-H) did not even pass the initial solubility test. The decrease in solubility with increasing MW is a well-known behavior of most polymers.[50] Our results indicate that this effect may be even more severe for polymers for which the used solvent is rather poor as is the case of the comparatively hydrophobic BMAA copolymers of the 2:1 BMA/MAA ratio when dissolved in an aqueous buffer. In other words, a low MW may be a prerequisite for the dissolution of the amphiphilic copolymers having relatively high content of hydrophobic units. In this context, it is noteworthy that sufficient hydrophobicity is needed for efficient copolymer insertion into a membrane.[51] We speculate that the MW-dependent polymer solubility effects might be one of the reasons why most of the (co)polymer types so far reported to be efficient cell membrane solubilizers are of low MW. It should

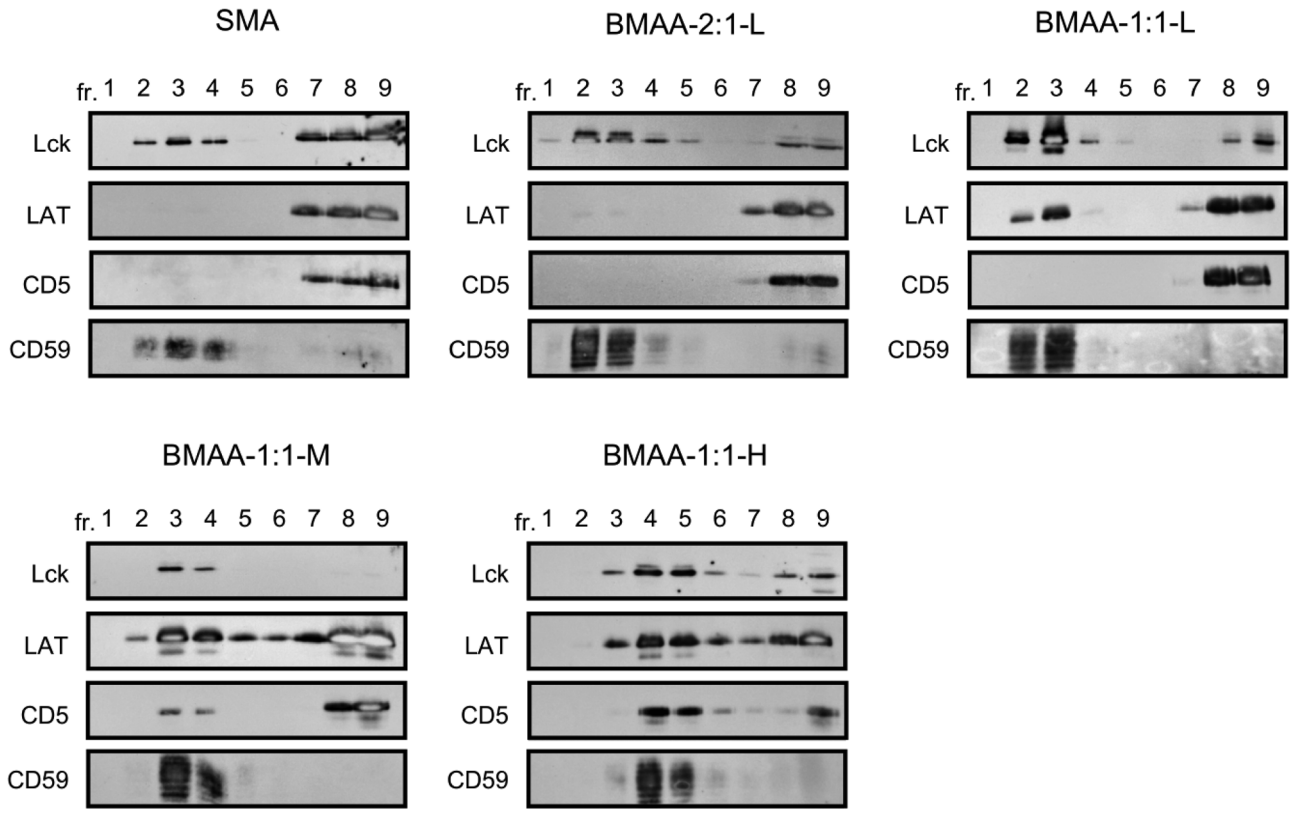


Figure 3. Distribution of the indicated Jurkat cell membrane proteins in the density gradient ultracentrifugation. Jurkat cell membranes were solubilized by the indicated copolymers, the lysates were fractionated by density gradient ultracentrifugation, and the indicated proteins in the fractions were detected by Western blotting. The fractions are numbered from the top of the gradient. Only the relevant parts of the blots are shown, corresponding to the area around the MW of the respective proteins (55 kDa for LCK, 45 kDa for LAT, 65 kDa for CD5, and 20 kDa for CD59).

be noted though that the situation can become rather complex when high-dispersity copolymers, such as the industrially produced SMA, are concerned because these materials involve polymeric chains of largely different MWs that may have very different solubility and self-aggregation behavior and hence also membrane solubilization properties. This highlights the necessity of using highly defined polymers when the key factors governing the process of cell membrane solubilization are investigated.

An important aspect of biological membranes is their lateral compositional heterogeneity, i.e., the presence of micro/nanodomains of different protein and lipid composition that may also differ in their susceptibility to detergent- or copolymer-mediated disintegration. The most studied type of such microdomains are so-called membrane rafts, enriched in cholesterol, lipids with saturated fatty acids, and lipid-modified membrane proteins.[52] Our previous results demonstrated that the SMA copolymer (used here as a comparative standard) apparently cuts most of the Jurkat cell membrane into small nanodiscs (SMALPs), containing membrane proteins surrounded by their native-like lipid environment.[53] However two types of raft proteins, GPI-anchored proteins and Src family kinases, were mostly present in much larger (>250 nm) SMA-resistant membrane fragments (SRMs), the composition of which resembled that of membrane rafts. The SRMs can be conveniently separated from the bulk of the membrane lysate by density gradient ultracentrifugation as they flotate to the top part of the gradient. Therefore, in the present study we examined to what extent the potential components of membrane rafts are also resistant to the disintegration by the newly developed BMAA copolymers. To this end, the four BMAA copolymers that showed the greatest solubilization power in the previous screening were, together with SMA as a positive control, used for the solubilization of Jurkat cell membranes, followed by density gradient fractionation. The fractions were then examined by SDS-PAGE, followed by Western blotting, with respect to the distribution of four membrane markers (CD59, LCK, LAT, CD5) differing in their affinity to lipid raft microdomains (**Figure 3**). As expected, the top (buoyant) gradient fractions of the sample solubilized by SMA contained copolymer-resistant membrane fragments (CRMs) characterized by the presence of membrane raft markers LCK (a palmitoylated and myristoylated protein tyrosine kinase) and CD59 (a GPI-anchored glycoprotein). The non-raft marker, the transmembrane protein CD5, was present in the bottom fractions of the gradient, presumably solubilized within small membrane nanodiscs, as reported in our previous study.[53] Also in agreement with our previous results, another potential membrane raft protein, the palmitoylated transmembrane adaptor protein LAT, was detectable only in the bottom fractions of the gradient (Figure 3). A similar pattern was observed in the case of the gradient fractions of membranes solubilized by the BMAA-2:1-L copolymer. However, in the case of membrane lysates produced by the other copolymers (BMAA-1:1-L, BMAA-1:1-M, BMAA-1:1-H), substan-

tial amounts of LAT were observed also in the top fractions of the gradient, indicating that the LAT-containing membrane rafts were more resistant to these copolymers. Therefore, these BMAA copolymers might represent a useful tool for biochemical studies on specific subsets of membrane microdomains differing in their composition and resistance to membrane attacking copolymers. It should be noted though that the CRMs are probably not a faithful biochemical equivalent of native membrane rafts, similarly to the situation with the detergent-resistant membrane fraction (DRM), as thoroughly discussed in literature, e.g., by Sezgin et al.[52]

The gradient fractions were analyzed also by dynamic light scattering (DLS) in order to compare the sizes of the membrane fragments produced by the individual copolymers (Figure S3, Supporting Information). The general pattern was similar in all the cases—the relatively large membrane fragments (hundreds of nm) were present mainly in the top fractions (No. 1–6) while the smaller ones essentially only in the bottom fractions (No. 7–9). It should be noted though that superficial inspection of the DLS curves may provide a misleading impression about the relative amounts of the large versus small membrane fragments as the signal intensity depends on the sixth power of the particle size and therefore is strongly biased towards the larger species. Indeed, the vast majority of membrane proteins are present in the relatively small membrane fragments obtained by SMA solubilization, as compared to the large SMA-resistant (presumably membrane raft derived) fragments.[53] Nevertheless, the data in Figure S3 (Supporting Information) indicate that the bottom fractions of the samples solubilized by the BMAA copolymers contain markedly more of larger fragments when compared to the fractions of the sample solubilized by the SMA control. This again indicates that these copolymers may be useful alternative tools for studies on the organization of biological membrane microdomains.

In comparison to SMA, the application of BMAA copolymers provides the obvious advantage of avoiding the strongly UV-absorbing and possibly overly perturbing styrene units. However, it is unclear to what extent the other disadvantages of the SMA-type copolymers, such as their sensitivity to acidic pH and to biologically relevant concentrations of Ca^{2+} or Mg^{2+} ions, are maintained also for BMAA copolymers that share the same charged group as SMA (carboxylate). Therefore, we briefly tested the four lead BMAA copolymers (BMAA-2:1-L, BMAA-1:1-L, BMAA-1:1-M, and BMAA-1:1-H) in this respect via turbidimetry. As is apparent from the data collected in Table S1 (Supporting Information), the BMAA copolymers precipitated significantly under the studied conditions, with the A_{490} values being the same or only marginally lower than for the SMA control. These results indicate that the sensitivity to the tested conditions stems largely from the presence of the carboxylic groups in the copolymer structure and that the more homogeneous distribution of these groups within the BMAA copolymeric chain (isolated COOH groups possible as compared to the pairwise arrangement in SMA) does not play a significant role.

3. Conclusions

In conclusion, we used here a straightforward two-step method to synthesize a comprehensive library of very well-defined amphiphilic BMAA copolymers differing in their MW and overall polarity. Using a biologically relevant model of Jurkat T-cell membranes, we identified four BMAA copolymers that solubilized the isolated Jurkat membranes to an extent comparable to the standard SMA copolymer. Surprisingly, the BMAA copolymers of the BMA/MAA = 1:1 composition were found to be effective in the whole studied MW range (≈from 3500 to 14 000). The more hydrophobic composition (BMA/MAA = 2:1) was also effective, but only the low-MW variant (BMAA-2:1-L) could be dissolved in the used aqueous medium. This observation highlights the importance of the low MW for the application of comparatively hydrophobic copolymers. Importantly, the results of the density gradient ultracentrifugation experiments demonstrated that three of the BMAA copolymers (BMAA-1:1-L, BMAA-1:1-M, and BMAA-1:1-H) disintegrated the model cell membranes in a manner different to SMA, as indicated by the different patterns of distribution of two of the tested membrane protein markers. This finding suggests that the BMAA copolymers may produce larger membrane fragments (CRMs), providing potentially information on the protein and lipid environment in membrane microdomains different from membrane rafts. The main limitation of the BMAA copolymers lies in their sensitivity toward low pH and biologically relevant Ca^{2+} or Mg^{2+} cations that is similar as for SMA. Finally, we envision that the library-based screening approach applied here can also be used for other amphiphilic copolymer designs, representing a straightforward route to expanding the still rather limited toolbox of cell membrane-solubilizing amphiphilic polymers and also helping to identify the key parameters characterizing such successful polymers. The knowledge of these parameters will help us advance toward the rational design approach in the future.

Supporting Information

Supporting Information is available from the Wiley Online Library or from the author.

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Conflict of Interest

The authors declare no conflict of interest.

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Keywords

amphiphilic copolymers, isolation, membrane proteins, screening, solubilization

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