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Synthesis of acyl chitin derivatives and miscibility characterization of their blends with poly(ϵ -caprolactone)

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1 **Synthesis of acyl chitin derivatives and miscibility characterization of their blends with**
2 **poly(ϵ -caprolactone)**

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13

14 **ABSTRACT**

15 Chitin derivatives having a normal acyl group (side-chain carbon number, $N = 2-6$) at different
16 degrees of substitution (DS) were synthesized by a homogenous reaction of crab shell chitin with
17 various acyl chlorides in *N,N*-dimethylacetamide-LiCl solution. NMR analysis quantitatively
18 demonstrated the acylations not only for C3/C6 hydroxy protons but also for C2 amino proton(s).
19 Solution cast blend films of the acyl chitin products with poly(ϵ -caprolactone) (PCL) were provided
20 for the miscibility characterization by differential scanning calorimetry. The critical total-DS
21 required for attaining a miscibility of the blending polymer pair decreased with an increase in N .
22 The degree of miscibility was enhanced definitely with an increase in ester-DS, but it made less
23 correlation with amide-DS. In analogy with cellulose ester/PCL blend systems, a structural
24 affinity of the ester side-group of the chitinous component considered with a repeating unit of PCL
25 may be a crucial factor for the miscibility attainment.

26

27 **Key words:** acyl chitin; degree of substitution; poly(ϵ -caprolactone); polymer blend; miscibility

28

1. Introduction

Chitin, which is supplied in abundance by marine crustaceans, is a representative of a naturally occurring polysaccharide derived from animal sources. It is structurally similar to cellulose and can be regarded as cellulose with the hydroxyl group at position C2 replaced by an acetylamino group. However, the difference in property between chitin and cellulose arises due to the presence of amino groups in the former, which is advantageous in that it can provide distinctive biological functions, for example, bioassimilability and antibacteriability. Therefore, chitin has a large potential for developing as specially functionalized materials in medical, pharmaceutical, food, and textile industries.

Chitin has a highly ordered structure because of intra- and intermolecular hydrogen bondings. Its rigid crystalline structure with multiple hydrogen bonds causes serious problems based on its poor solubility and thermal-moldability. To improve the processibility for subsequent applications, acylation has been extensively studied as a modification reaction, where carboxylic acids (Teramoto et al., 2006; Yang et al., 2009), acid anhydrides (Hirano & Ohe, 1975; Kurita et al., 1977; Nishi et al., 1979; Shoruigin & Hait, 1935), and acyl chlorides (Fujii et al., 1980; Kaifu et al., 1981; Kurita et al., 1988) have been used as attacking reagents. However, it is still not so easy to thermally mold the product (Acyl-Ch) alone, because the glass transition temperature and melting or flowing point are fairly high; marked thermal decomposition can be a serious problem. In order to alter the thermal properties of Acyl-Ch, the utilization of plasticizer is an effective method. If we adopt flexible polymers as plasticizer, we can avoid possible problems in the use of low molecular weight plasticizers, such as fume generation in the molding process and bleeding out in long-term applications.

As is well established, polymer/polymer blending is an important method to modify the original physical properties of one or both of the components, or to obtain new polymeric materials having wide-ranging properties. Blend works of chitinous polymers including deacetylated product

(chitosan) have been facilitated (Honma et al., 2003; Ko et al., 1997; Kubota et al., 1998; Lee et al., 1996; Miyashita et al., 1995, 1996, 1997a, b; Nishio et al., 1999) and the number of examples is still increasing. Among them, chitinous blends with water-soluble or hydrophilic synthetic polymers (Aoi et al., 1995; Kubota et al., 1998; Lee et al., 1996; Miyashita et al., 1995, 1996; Nishio et al., 1999) may be of importance from a standpoint of application as biomedical materials.

Meanwhile, aliphatic polyesters, which provide good biocompatibility and environmental degradability, have also attracted considerable interest as candidate materials for a blending partner of chitinous (Honma et al., 2003; Ikejima & Inoue, 2000; Ikejima et al., 1999; Mi et al., 2002; Mi et al., 2003; Sarasam & Madhally, 2005; Senda et al., 2002; Wan et al., 2008) and other biomass polymers (Kusumi et al., 2008; Nishio et al., 1997; Teramoto et al., 2009). Several preparation methods of chitinous blends with aliphatic polyesters proved to be a potential route leading to biomaterials applicable for tissue engineering (Sarasam & Madhally, 2005; Wan et al., 2008) and drug delivery systems (Mi et al., 2002, 2003); these studies were based on the control of the inter-component morphology on the micrometer scale, however. Generally, it seems more difficult to obtain intimate mixtures of chitinous polymers and aliphatic polyesters on the scale of a few nanometers, which can be judged to be thermodynamically miscible in the amorphous fraction of the mixture.

In the present study, a representative aliphatic polyester, poly(ϵ -caprolactone) (PCL), was chosen as a counterpart for Acyl-Ch blends. In contrast to chitinous polymers, PCL is easily heat-moldable due to a rather low melting temperature (~ 60 °C); therefore this is an eligible partner for blending. Acyl-Ch derivatives having various normal acyl side groups ($-\text{COC}_n\text{H}_{2n+1}$; side-chain carbon number, $N = n + 1 = 2-6$), i.e., acetate (Ac-Ch), propionate (Pr-Ch), butyrate (Bu-Ch), valerate (Va-Ch), and caproate (Ca-Ch), were synthesized by a homogeneous reaction of crab shell chitin with different acyl chlorides in *N,N*-dimethylacetamide (DMAc)-LiCl solution. Formerly, one of the authors (Y. N.) has prepared several cellulose ester (CE) derivatives and investigated an effect of the side-chain length on the blend miscibility with PCL (Kusumi et al.,

2008; Nishio et al., 1997). It was reported that cellulose butyrate (CB) and valerate (CV) exhibited the highest miscibility. In that case, a dipole-dipole interaction between the carbonyl of PCL and that in the side groups of CB and CV appeared to be an important factor for the miscibility. A possibility of further contribution of a structural similarity was also pointed out; because CB and CV have the same structural unit as that of PCL, if the carbon atoms in a glucopyranose unit are taken into account. A main objective of the present study is to clarify the relationship between the ability of Acyl-Ch to form a miscible blend with PCL and the side-chain structure in terms of side-chain length (N) and degree of substitution (DS), through observations of the thermal behavior for the binary polymer mixtures. We also intend to compare the extent of blend miscibility between Acyl-Ch/PCL and CE/PCL systems, possibly, both of them being a biodegradation-controllable material of great promise based on a difference in the degradation behavior between the constituents (Kusumi et al., in press; Teramoto & Nishio, 2004).

2. Experimental section

2.1 Materials

An original material of chitin isolated from crab shells (Fluka 22720 Chitin Lot & Filling code: 405226/1 12900) was purified by treatment with aqueous hydrochloric acid and sodium hydroxide solutions according to a Hackman's method (Hackman, 1954). The nominal molecular weight of the chitin material is 400,000. The degree of deacetylation (DD) of the purified chitin (p-Ch) was 5.0 %, determined by infrared spectroscopy (Sannan et al., 1978). The viscosity average molecular weight (M_v) was evaluated for a deacetylated product of the p-Ch, treated with 50 wt% aqueous NaOH at 95 ± 5 °C, by measuring the intrinsic viscosity ($[\eta]$) in a solvent of 0.1:4.0:0.2 M NaCl/urea/acetic acid (Tokura, 1991); where the Mark-Houwink Sakurada equation, $[\eta] = KM_v^\alpha$,

was used with $K = 8.93 \times 10^{-4}$ dL/g and $\alpha = 0.71$. The corresponding viscosity average degree of polymerization (DP) was determined to be 380 for the p-Ch.

PCL with a nominal weight average molecular weight of 15,000 was purchased from Scientific Polymer Products, Inc., and it was used after purification by dissolution in tetrahydrofuran (THF; Wako Pure Chemical Industries, Ltd.) and reprecipitation into distilled water. Reagent-grade DMAc, *N,N*-dimethylformamide (DMF), and dimethyl sulfoxide (DMSO) were purchased from Nacalai Tesque, Inc. and stored over molecular sieves 4A before use. Lithium chloride (LiCl) was purchased from Nacalai Tesque, Inc., Japan and dried at 120 °C for 12 h in a vacuum oven. Triethylamine (TEA), propionyl chloride, *n*-butyryl chloride, *n*-valeryl chloride, and *n*-caproyl chloride were obtained from Aldrich or Wako Pure Chemical Industries, Ltd. and used without further purification. Other solvents were all guaranteed reagent-grade and used as received.

2.2 Preparation of chitin solution

The p-Ch sample was first treated with water, methanol, and DMAc in succession by a solvent exchange technique (Miyashita et al., 1997a; Nishio et al., 1987). The DMAc-wet chitin was then added to a solvent system DMAc-LiCl and stirred at room temperature for 5 days. The solvent DMAc-LiCl was used at a salt concentration of 7 wt% with respect to DMAc. The actual concentration of chitin in the clear solution was controlled at 0.5 wt%; this was determined precisely by weighing the solid film regenerated from a portion of the solution.

2.3 Synthesis of Acyl-Ch derivatives

Acyl-Ch derivatives were synthesized with acid chloride/base catalyst from the chitin solution in a way similar to that used in previous studies for cellulose (Kusumi et al., 2008; Nishio et al., 1997). Taking an example of synthesis for Va-Ch, the typical procedure can be summarized as

follows. A solution of 0.098 mol TEA (10 eq/glucopyranose unit) in DMAc ($\approx$ 15 mL) was slowly added dropwise to 400 g of chitin solution (containing 2 g chitin). After 30 min, a solution of 0.28 mol *n*-valeryl chloride (28 eq/glucopyranose unit) in DMAc ($\approx$ 36 mL) was added to the chitin solution. The reactive solution system was stirred continuously at 50 °C under a nitrogen atmosphere. After reaction over a prescribed time period (6–48 h), the solution was added dropwise into a vigorously stirred, large excess amount of distilled water. Then the product obtained as precipitate was filtered, dissolved in acetone, and reprecipitated in a large excess of distilled hexane, which was followed by standing with slow stirring and collection by filtration. Via further repetition of this purification procedure, the collected Va-Ch product was dried at 40 °C in vacuo for three days.

2.4 Preparation of Acyl-Ch/PCL blends

Acyl-Ch/PCL blends were cast in film form from mixed polymer solutions by solvent evaporation. A 1.0 wt% solution of Acyl-Ch and that of PCL were prepared in DMF separately and mixed with each other in the desired proportion which ranged from 10/90 to 95/5 in a weight percent ratio of Acyl-Ch:PCL. After stirring at room temperature for 24 h, each mixed solution (transparent) was poured into a Teflon tray and a film sheet was made by evaporating DMF at 50 °C under reduced pressure (<10 mmHg). The as-cast samples were washed with distilled water and dried at 40 °C in vacuo for 3 days.

2.5 Measurements

Gel permeation chromatography (GPC) was carried out with a Tosoh HLC-8220 GPC apparatus. The measuring conditions were as follows: column, two Tosoh TSK Super HZM-H columns connected with each other; flow rate, 0.25 mL/min; temperature 40 °C; eluent, THF; standard,

159 monodispersed polystyrene.

160 ^1H NMR spectra were measured at 25 °C for purified Acyl-Ch derivatives by using a Varian
161 INOVA300 apparatus. The solvent was CDCl_3 , pyridine- d_5 or CF_3COOD and the concentration
162 was 10 mg/mL. Tetramethylsilane was employed as an internal standard. 128 scans were
163 conducted.

164 FT-IR spectra were measured with a Shimadzu FT-IR 8600C apparatus. All the measurements
165 were carried out at 25 °C. Samples were dried at 80 °C in vacuo for 24 h and a standard KBr pellet
166 method was employed for all the measurements.

167 Differential scanning calorimetry (DSC) was carried out with a Seiko DSC6200/EXSTAR6000
168 apparatus. The temperature proof-readings were calibrated with an indium standard. The
169 calorimetry measurements were made on ca. 5-mg samples at a scanning rate of 20 °C/min under a
170 nitrogen atmosphere. The samples were first heated to 200 °C and immediately quenched to −140
171 °C. In this first cycle, the thermal histories of the respective samples were equalized completely.
172 Then the second scans were run from −140 °C to 200 °C to record stable thermograms. The glass
173 transition temperature (T_g) was determined from the midpoint of the discontinuity in heat flow.

174

175

176 3. Results and discussion

177

178 *Characterization of Acyl-Ch derivatives*

179

180 The degree of total acyl substitution (total-DS) of each of the Acyl-Ch products was determined
181 by ^1H NMR measurements. Figure 1 demonstrates a ^1H NMR spectrum obtained for a Bu-Ch
182 sample. In the spectrum, the intensity of the methyl-group proton signals (δ 0.8–1.1 ppm) of acyl
183 moiety was compared with that of the signals (δ 3.1–5.8 ppm) from the chitinous backbone protons
184 located in the anhydroglucosamine unit. We designated a resonance peak area derived from the

185 methyl protons of acyl groups as [A], and an area of the resonance signals from the protons of
186 glucopyranose as [B]. Then, for Ac-Ch, Pr-Ch, Bu-Ch, Va-Ch, and Ca-Ch, the respective total-DS
187 values were calculated by

$$188 \quad \text{total-DS} = ([A] / 3) / ([B] / 7)$$

189 Hereafter, Acyl-Ch of total-DS = x is encoded as Acyl _{x} -Ch.

190 In the present acylation system, for detailed molecular characterization, we should take into
191 consideration the reaction of acyl chlorides not only with protons in the hydroxyl groups but also
192 with C2-NH₂ and C2-NHCOR of chitin (DD = 5 % for p-Ch). This is supported by FT-IR
193 spectroscopy qualitatively. Figure 2 displays FT-IR spectra of p-Ch and selected Bu-Ch samples.
194 As can be seen from the spectral data, no absorption was present in the spectral region associated
195 with NH₂ (1625 cm⁻¹) after the acylation. Instead, for the Bu-Chs, a remarkable development in
196 absorption was observed in 1750 cm⁻¹ (ester C=O). With an increase in total-DS (≥ 2.50 in Fig. 2),
197 the amide I band (1660 cm⁻¹, C=O of NHCOR) shifted to the side of higher frequency, implying the
198 suppression of the intramolecular hydrogen bondings of the C=O with O6H and/or N-H (Focher et
199 al., 1992; Pearson et al., 1960). Further increasing total-DS gave rise to a suppression in the amide
200 I band, while a new absorption at 1710 cm⁻¹ (C=O of N(COR)₂) was present and it became
201 prominent with increasing total-DS. This observation indicates the occurrence of *N*-acylation of
202 the acetamide and/or *N,N*-diacyl substitution on the glucosamine residue. Similar *N,N*-diacyl
203 substitution was reported previously for the acylation of chitosan with acyl chloride in refluxed
204 pyridine and chloroform (Fujii et al., 1980).

205 Detailed quantitative molecular characterization for Acyl-Ch samples can be conducted by ¹H
206 NMR. A series of the spectra exhibited signals of the protons of acetamide methyl (1.8 ppm) and
207 amide N-H (6.1 ppm), other than those of the acyl methyl and chitinous backbone discussed above
208 for the estimation of total-DS. Since there is no NH₂ moiety in the chitin derivatives after the
209 acylation as has been shown in the FT-IR measurements (Fig. 2), the ¹H NMR data were used for
210 the determination of amide-DS, ester-DS, and DD, where amide-DS and ester-DS denote the

average number of acyl substitution associated with C2-NH₂/C2-NHCOR and C3-OH/C6-OH, respectively, per glucosamine residue of chitin. In Fig. 1, we designate a peak area of the resonance of the acetamide methyl protons as [C] and an area of the resonance signal from the amide N-H proton as [D]. Then, the values of DD, amide-DS, and ester-DS are able to be determined by following equations, respectively:

$$DD = (1 - ([C] / 3) / ([B] / 7)) \times 100 (\%)$$

$$\text{amide-DS} = \text{total-DS} - ([C] / 3 + [D])$$

$$\text{ester-DS} = \text{total-DS} - \text{amide-DS}$$

Table 1 tabulates the four parameters evaluated for the Acyl-Ch products prepared in this study. In a general trend, Acyl-Ch samples with higher total-DS incorporated higher ester-DS. Amide-DS was estimated to be >1 for most of the Acyl-Ch products, reflecting the replacement of *N*-acetate with another acyl group for these samples. Eventually, DD values were determined to be 10–80 %, raised noticeably from an initial value 5 % for p-Ch.

The series of Acyl-Ch was found to have improved solubility in conventional solvents, compared with chitin. As listed in Table 1, Acyl-Ch derivatives with higher DS and longer side chains showed better solubility in ordinary organic solvents. Particularly, Pr-Ch, Bu-Ch, Va-Ch, and Ca-Ch with higher total-DSs (3.06–3.77, 2.50–3.33, 2.32–3.09, and 2.64–2.94, respectively) were soluble in MeOH, differing from the corresponding CE samples with similar acyl DSs.

Values of the weight-average molecular weight (and the corresponding weight average DP) of representative Acyl-Ch samples were determined by GPC, as follows: 8.81×10^4 (210), 12.8×10^4 (267), 12.4×10^4 (244), and 12.3×10^4 (225), for Pr_{3.16}-Ch, Bu_{3.33}-Ch, Va_{3.48}-Ch, and Ca_{3.42}-Ch, respectively. Taking into consideration a viscosity average DP (380) of p-Ch, the DP of the Acyl-Ch products tended to slightly decrease, indicating some extent of scission of the chitinous backbone chain in the acylation reaction. The molecular weight distributions for the above four products were 2.89 (Pr_{3.16}-Ch), 2.51 (Bu_{3.33}-Ch), 1.94 (Va_{3.48}-Ch), and 2.25 (Ca_{3.42}-Ch).

237 *Miscibility characterization of Acyl-Ch/PCL blends*

238

239 Binary blend films of Acyl-Ch products with PCL were prepared in a wide range of
240 compositions by solution casting from DMF solution. Thus obtained blend films were visually
241 homogeneous, but the transparency decreased with an increase in PCL content. Higher
242 transparency was usually observed for the blends of Acyl-Ch samples of higher total-DSs.

243 Polymer-polymer miscibility is commonly estimated by the determination of the glass transition
244 temperature (T_g) of the blends. If any blend of a binary polymer system exhibits a single glass
245 transition between the T_g values of both components and a composition-dependent shift in T_g of the
246 blend is observed, then the system can be regarded as a highly miscible one on the T_g -detection
247 scale that is usually assumed to be less than a couple of tens of nanometers (Kaplan, 1976; Nishio,
248 1994; Utracki, 1990). If one component is crystalline, observation of a regression in the
249 development of crystallinity of the crystallizable component (PCL in the present study), caused by
250 addition of the second component (i.e. Acyl-Ch), is a common feature which is shared with other
251 crystalline/amorphous polymer pairs that are capable of forming a miscible phase in their blends
252 (Imken et al., 1976; Nishi & Wang, 1975).

253 In this work, the respective blend systems were classified into immiscible, partially miscible,
254 and miscible ones from their DSC measurements from the viewpoints of T_g -shift and crystallinity
255 regression. The PCL homopolymer gave a T_g signal centering -61.0 °C and besides a sharp
256 melting endotherm with a peak maximum (T_m) at 55.2 °C. The other component Acyl-Ch
257 derivatives were almost amorphous and no melting signal was observed in the present thermal
258 scanning. As listed in Table 1, a sharp depression of the T_g of chitin (> 200 °C) to the lower
259 temperature side was observed along with the increase in total-DS as well as in N . However, no
260 glass transition was observed for several samples of lower DSs, in which the acylation was
261 insufficient for internal plasticization of the carbohydrate backbone.

262 Figure 3a shows DSC thermograms obtained for Pr_{2.76}-Ch/PCL blends. It is judged that the

blending polymer pair is poorly miscible, from observation of the less-significant shift in T_g for both components. In particular, the compositions of 20/80–90/10 showed two independent glass transitions. In addition, the melting endotherm of PCL was observable for every composition with the peak area reducing in direct proportion to the PCL content. Thus, the thermal properties of the respective polymer components were never affected by the presence of the other component in the Pr_{2.76}-Ch/PCL blends.

On the other hand, Va_{2.86}-Ch indicated somewhat better miscibility with PCL. As shown in Fig. 3b, a series of Va_{2.86}-Ch/PCL blends gave rise to an appreciable degree of elevation in the PCL T_g . A similar shift in T_g was observed for the Acyl-Ch component at compositions of 95/5–70/30. Furthermore, a cold-crystallization phenomenon took place after onset of the glass transition of the blend on heating for the compositions of 20/80–60/40, reflecting a relatively slow kinetics of crystallization of the PCL component from the molten mixtures with Va_{2.86}-Ch. It should also be noted that the PCL crystallinity almost vanished when the Va_{2.86}-Ch content reached 90 wt%. These results supports that the polymer pair may be miscible at a certain level in the non-crystalline state of the mixture. Therefore, this type of blend system was judged to be partially miscible.

Figure 3c demonstrates an example of a miscible pair. Highly butyrated Bu_{3.08}-Ch can be taken as miscible with PCL in the amorphous mixing state, since a single T_g was detected and varied between the T_g values of the two constituent polymers, depending on the blend composition. The crystallinity of the PCL component disappeared completely when the Bu_{3.08}-Ch content increased to 85 wt%. Several other systems composed of a highly substituted Acyl-Ch/PCL pair provided similar thermogram data and they were determined to be miscible. In more careful inspection, however, the T_g shift was prominent only at compositions rich in Acyl-Ch (> 80 wt%). In addition, in such miscible Acyl-Ch/PCL systems, the crystallization habit of PCL remained clearly even at 80/20 composition. Contrary to this, in the previous CE/PCL series judged as miscible, the PCL crystallinity was no longer detectable at compositions containing > 60 wt% CE and the blend T_g shifted more conspicuously and smoothly in the composition range of 100/0–60/40

(Kusumi et al., 2008). These observations imply that the highly substituted Acyl-Chs generally show a somewhat lower degree of miscibility with PCL, in comparison with the previously used CEs.

Insight into factors for miscibility attainment and comparison with cellulosic systems

Figure 4 summarizes the result of the miscibility estimation for Acyl-Ch blends with PCL as a function of the number N of carbons in the normal acyl substituent as well as of the substitution parameters, total DS, ester DS, and amide DS. For comparison, the corresponding result for CE blends with PCL is shown in Fig. 5, compiling the previous data (Kusumi et al., 2008; Nishio et al., 1997) and a complementary one for cellulose caproate (DS = 2.79 for $N = 6$) which was prepared especially for the present study. Ac-Ch ($N = 2$) was immiscible with PCL even in the highly acetylated state of DS ≈ 3.8 , as in the situation with cellulose acetate (Higeshiro et al., 2009; Nishio et al., 1997). Other Acyl-Ch products, Pr-Ch ($N = 3$), Bu-Ch ($N = 4$), Va-Ch ($N = 5$), and Ca-Ch ($N = 6$) showed miscibility with PCL when totally high-substituted grades of them were used. A critical total-DS value for the miscibility attainment, total-DS_{cr}, decreased with an increase in N . However, for the blend series of Acyl-Ch of $N = 3$ –6, total-DS_{cr} value was generally higher than the critical (ester-)DS for the corresponding CE/PCL system. It should also be recalled that the series of miscible Acyl-Ch/PCL combinations generally showed a lower degree of miscibility with PCL, in comparison with the miscible CE blends, as deduced already from the inspection of T_g shift and PCL-crystallinity regression. Such a lower degree of the miscibility can be attributed partly to a steric hindrance of the bulky planar acetamide structure on the chitinous backbone, to the accessibility of oxycaproyl segments of the PCL component. For the miscible CE/PCL blends, we inferred a possible contribution of dipole-dipole interaction between the carbonyl of PCL and that in the side groups of CE (Kusumi et al., 2008); in the Acyl-Ch/PCL systems, however, similar contribution would be suppressed by the steric hindrance mentioned above.

315 It was also reported for the CE/PCL system that a structural affinity of the ester side-group of
316 the cellulosic component considered with a repeating unit of PCL may be a crucial factor for
317 miscibility attainment (Kusumi et al., 2008; Nishio et al., 1997). Namely, as shown in Fig. 6, for
318 instance, the butyryl side-group is structurally identical with the repeating unit of PCL, if the
319 carbons in the glucopyranose ring are taken into account. For the present Acyl-Ch series, however,
320 the *N*-acyl substitution at C2 position can not contribute to the improvement of such an affinity,
321 although the esterification at C3/C6 positions can do. Therefore, higher total-DS_{cr} values would
322 be required for the miscibility attainment. Actually, the miscibility increased with an increase in
323 ester-DS (Fig. 4b), but there is less correlation between the miscibility and amide-DS (Fig. 4c).

324 Any of CEs with *N* = 3–5 showed good miscibility with PCL at DSs of ≥ 2.2 , and particularly
325 butyrate of *N* = 4 did even at a comparatively low DS of 1.9 (Fig. 5). However, cellulose caproate
326 (*N* = 6) and enanthate (*N* = 7) having fairly longer side-chains were estimated to show a relatively
327 lower degree of miscibility with PCL (Kusumi et al., 2008). It was assumed, exceptionally, that
328 these CEs would be able to aggregate to form a specific, mesomorphic ordered assembly, as was
329 also suggested for some cellulose triester (Takada et al., 1994) and chitin 3,6-*O*-diester (Teramoto et
330 al., 2006) derivatives having a longer acy substituent. On the other hand, the *N*-dependence of the
331 advent of miscibility for the Acyl-Ch blend systems differs from that for the CE systems, namely,
332 the miscibility of the former systems increased with an increase in *N* so far as the side-chain lengths
333 of ≤ 6 . In particular, Ca-Ch of *N* = 6 exhibited the lowest total-DS_{cr}, signifying the highest
334 miscibility with PCL. This may be because the caproyl side-chains virtually identical to the
335 repeating unit of PCL can contribute to the enhancement of the miscibility even if it is introduced
336 by “*N*-acylation”.

337 Further investigation of the side-chain length effect on the miscibility state may be made in
338 terms of isothermal crystallization behavior of the PCL component in miscible Acyl-Ch/PCL blend
339 systems. The topic including crystallization kinetics and spherulitic morphology will be reported
340 in no distant future.

341

342

343 Conclusions

344

345 We prepared chitin derivatives having various normal acyl groups ($-\text{OCOC}_n\text{H}_{2n+1}$; side-chain
346 carbon number, $N = n + 1 = 2-6$) by homogeneous reaction of crab-shell chitin with different acyl
347 chlorides in DMAc-LiCl solution. The degrees of *O*- and *N*-acyl substitutions were separately
348 evaluated by NMR analysis successfully. It was thus shown that the acylating reaction took place
349 not only to the protons in the C3/C6-hydroxyl groups but also to the protons in C2-NH₂ and
350 C2-NHCOR of the starting chitin (DD = 5 %). The replacement of *N*-acetate with another acyl
351 group also arose in these samples.

352 Subsequently, miscibility characterization was carried out for blends of Acyl-Ch products with
353 PCL. DSC thermal analysis demonstrated that the critical total-DS required for the miscibility
354 attainment, total-DS_{cr}, decreased with an increase in side-chain carbon number *N*. However, for
355 the series of Acyl-Ch blends, the respective total-DS_{cr} values were higher than the critical
356 (ester-)DS for the corresponding CE/PCL system. It was also perceived that the miscibility found
357 for several Acyl-Ch/PCL combinations was generally of a lower degree, in comparison with that for
358 the CE/PCL blends.

359 In analogy with the CE/PCL systems, the structural affinity of the ester side-group of the
360 chitinous component considered with a repeating unit of PCL may be a crucial factor. However,
361 the *N*-acyl substitution at C2 position seems not to contribute so greatly to the improvement of such
362 an affinity. In support of this, the blend miscibility of Acyl-Chs ($N = 3-6$) with PCL was enhanced
363 with an increase in ester-DS, but there is less correlation between the miscibility and amide-DS.

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366 References

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368 Aoi, K., Takasu, A., & Okada, M. (1995). New chitin-based polymer hybrids. 1. Miscibility of
369 poly(vinyl alcohol) with chitin derivatives. *Macromolecular Rapid Communications*, 16,
370 757-761.

371 Focher, B., Naggi, A., Torri, G., Cosani, A., & Terbojevich, M. (1992). Structural differences
372 between chitin polymorphs and their precipitates from solutions: Evidence from CP-MAS
373 ¹³C-NMR, FT-IR and FT-Raman spectroscopy. *Carbohydrate Polymers*, 17, 97-102.

374 Fujii, S., Kumagai, H., & Noda, M. (1980). Preparation of poly(acyl)chitosans. *Carbohydrate*
375 *Research*, 83, 389-393.

376 Hackman, R. (1954). Studies on chitin. 1. Enzymatic degradation of chitin and chitin esters.
377 *Australian Journal of Biological Sciences*, 7, 168-178.

378 Higeshiro, T., Teramoto, Y., & Nishio, Y. (2009). Poly(vinyl pyrrolidone-co-vinyl
379 acetate)-graft-poly(ε-caprolactone) as a compatibilizer for cellulose
380 acetate/poly(ε-caprolactone) blends. *Journal of Applied Polymer Science*, 113, 2945-2954.

381 Hirano, S., & Ohe, Y. (1975). Facile *n*-acylation of chitosan with carboxylic anhydrides in acidic
382 solutions. *Carbohydrate Research*, 41, C1-C2.

383 Honma, T., Senda, T., & Inoue, Y. (2003). Thermal properties and crystallization behaviour of
384 blends of poly(ε-caprolactone) with chitin and chitosan. *Polymer International*, 52,
385 1839-1846.

386 Ikejima, T., & Inoue, Y. (2000). Crystallization behavior and environmental biodegradability of the
387 blend films of poly(3-hydroxybutyric acid) with chitin and chitosan. *Carbohydrate Polymers*,
388 41, 351-356.

389 Ikejima, T., Yagi, K., & Inoue, Y. (1999). Thermal properties and crystallization behavior of
390 poly(3-hydroxybutyric acid) in blends with chitin and chitosan. *Macromolecular Chemistry*
391 *and Physics*, 200, 413-421.

392 Imken, R. L., Paul, D. R., & Barlow, J. W. (1976). Transition behavior of poly(vinylidene

- 393 fluoride)/poly(ethyl methacrylate) blends. *Polymer Engineering and Science*, 16, 593-601.
- 394 Kaifu, K., Nishi, N., & Komai, T. (1981). Preparation of hexanoyl, decanoyl, and dodecanoylchitin.
- 395 *Journal of Polymer Science, Polymer Chemistry Edition*, 19, 2361-2363.
- 396 Kaplan, D. S. (1976). Structure-property relationships in copolymers to composites: Molecular
- 397 interpretations of the glass transition phenomemon. *Journal of Applied Polymer Science*, 20,
- 398 2615-2629.
- 399 Ko, M. J., Jo, W. H., Kim, H. C., & Lee, S. C. (1997). Miscibility of chitosans polyamide 6 blends.
- 400 *Polymer Journal*, 29, 997-1001.
- 401 Kubota, N., Konaka, G., & Eguchi, Y. (1998). Characterization of blend films of chitin regenerated
- 402 from porous chitosan with poly(vinyl alcohol). *Sen'I Gakkaishi*, 54, 212-218.
- 403 Kurita, K., Chikaoka, S., Kamiya, M., & Koyama, Y. (1988). Studies on chitin. 14. *N*-Acetylation
- 404 behavior of chitosan with acetyl chloride and acetic anhydride in a highly swelled state.
- 405 *Bulletin of the Chemical Society of Japan*, 61, 927-930.
- 406 Kurita, K., Sannan, T., & Iwakura, Y. (1977). Studies on chitin. 3. Preparation of pure chitin,
- 407 poly(*n*-acetyl-D-glucosamine), from water-soluble chitin. *Die Makromolekulare Chemie*, 178,
- 408 2595-2602.
- 409 Kusumi, R., Inoue, Y., Shirakawa, M., Miyashita, Y., & Nishio, Y. (2008). Cellulose alkyl
- 410 ester/poly(ϵ -caprolactone) blends: Characterization of miscibility and crystallization
- 411 behaviour. *Cellulose*, 15, 1-16.
- 412 Kusumi, R., Lee, S. H., Teramoto, Y., & Nishio, Y. (in press). Cellulose
- 413 ester-graft-poly(ϵ -caprolactone): Effects of copolymer composition and intercomponent
- 414 miscibility on the enzymatic hydrolysis behavior. *Biomacromolecules*, 10.1021/bm900666y.
- 415 Lee, Y. M., Kim, S. H., & Kim, S. J. (1996). Preparation and characteristics of β -chitin and
- 416 poly(vinyl alcohol) blend. *Polymer*, 37, 5897-5905.
- 417 Mi, F. L., Lin, Y. M., Wu, Y. B., Shyu, S. S., & Tsai, Y. H. (2002). Chitin/PLGA blend microspheres
- 418 as a biodegradable drug-delivery system: Phase-separation, degradation and release behavior.

- 419 *Biomaterials*, 23, 3257-3267.
- 420 Mi, F. L., Shyu, S. S., Lin, Y. M., Wu, Y. B., Peng, C. K., & Tsai, Y. H. (2003). Chitin/PLGA blend
421 microspheres as a biodegradable drug delivery system: A new delivery system for protein.
422 *Biomaterials*, 24, 5023-5036.
- 423 Miyashita, Y., Kobayashi, R., Kimura, N., Suzuki, H., & Nishio, Y. (1997a). Transition behavior and
424 phase structure of chitin/poly(2-hydroxyethyl methacrylate) composites synthesized by a
425 solution coagulation bulk polymerization method. *Carbohydrate Polymers*, 34, 221-228.
- 426 Miyashita, Y., Sato, M., Kimura, N., Nishio, Y., & Suzuki, H. (1996). Effect of deacetylation of
427 chitin on the miscibility of chitin/poly(vinyl alcohol) blends (in Japanese, with English
428 abstract). *Kobunshi Ronbunshu*, 53, 149-154.
- 429 Miyashita, Y., Yamada, Y., Kimura, N., Nishio, Y., & Suzuki, H. (1995). Preparation and miscibility
430 characterization of chitin/poly(*n*-vinyl pyrrolidone) blends (in Japanese, with English
431 abstract). *Sen'I Gakkaishi*, 51, 396-399.
- 432 Miyashita, Y., Yamada, Y., Kimura, N., Suzuki, H., Iwata, M., & Nishio, Y. (1997b). Phase structure
433 of chitin poly(glycidyl methacrylate) composites synthesized by a solution coagulation bulk
434 polymerization method. *Polymer*, 38, 6181-6187.
- 435 Nishi, N., Noguchi, J., Tokura, S., & Shiota, H. (1979). Studies on chitin. 1. Acetylation of chitin.
436 *Polymer Journal*, 11, 27-32.
- 437 Nishi, T., & Wang, T. T. (1975). Melting point depression and kinetic effects of cooling on
438 crystallization in poly(vinylidene fluoride)-poly(methyl methacrylate) mixtures.
439 *Macromolecules*, 8, 909-915.
- 440 Nishio, Y. (1994). Hyperfine composites of cellulose with synthetic polymers. In R. D. Gilbert (Ed).
441 *Cellulose polymers, blends and composites* (pp. 95-113). Munich, New York: Hanser Gardner
442 Publications.
- 443 Nishio, Y., Koide, T., Miyashita, Y., Kimura, N., & Suzuki, H. (1999). Water-soluble polymer
444 blends with partially deacetylated chitin: A miscibility characterization. *Journal of Polymer*

- 445 *Science, Part B: Polymer Physics*, 37, 1533-1538.
- 446 Nishio, Y., & Manley, R. S. (1988). Cellulose-poly(vinyl alcohol) blends prepared from solutions in
447 *N,N*-dimethylacetamide-lithium chloride. *Macromolecules*, 21, 1270-1277.
- 448 Nishio, Y., Matsuda, K., Miyashita, Y., Kimura, N., & Suzuki, H. (1997). Blends of
449 poly(ϵ -caprolactone) with cellulose alkyl esters: Effect of the alkyl side-chain length and
450 degree of substitution on miscibility. *Cellulose*, 4, 131-145.
- 451 Nishio, Y., Roy, S. K., & Manley, R. S. (1987). Blends of cellulose with polyacrylonitrile prepared
452 from *N,N*-dimethylacetamide-lithium chloride solutions. *Polymer*, 28, 1385-1390.
- 453 Pearson, F. G., Marchessault, R. H., & Liang, C. (1960). Infrared spectra of crystalline
454 polysaccharides. 5. Chitin. *Journal of Polymer Science*, 43, 101-116.
- 455 Sannan, T., Kurita, K., Ogura, K., & Iwakura, Y. (1978). Studies on chitin. 7. IR spectroscopic
456 determination of degree of deacetylation. *Polymer*, 19, 458-459.
- 457 Sarasam, A., & Madhally, S. V. (2005). Characterization of chitosan-polycaprolactone blends for
458 tissue engineering applications. *Biomaterials*, 26, 5500-5508.
- 459 Senda, T., He, Y., & Inoue, Y. (2002). Biodegradable blends of poly(ϵ -caprolactone) with
460 alpha-chitin and chitosan: Specific interactions, thermal properties and crystallization
461 behavior. *Polymer International*, 51, 33-39.
- 462 Shorugin, P. P., & Hait, E. (1935). Acetylation of chitin. *Berichte der Deutschen Chemischen*
463 *Gesellschaft [Abteilung] B: Abhandlungen*, 68B, 971-973.
- 464 Takada, A., Fujii, K., Watanabe, J., Fukuda, T., & Miyamoto, T. (1994). Chain-length dependence of
465 the mesomorphic properties of fully decanoated cellulose and cellooligosaccharides.
466 *Macromolecules*, 27, 1651-1653.
- 467 Teramoto, Y., Lee, S. H., & Endo, T. (2009). Phase structure and mechanical property of blends of
468 organosolv lignin alkyl esters with poly(ϵ -caprolactone). *Polymer Journal*, 41, 219-227.
- 469 Teramoto, Y., Miyata, T., & Nishio, Y. (2006). Mesomorphic assemblage of chitin normal acylates
470 and rapid enthalpy relaxation of their side chains. *Biomacromolecules*, 7, 190-198.

- 471 Teramoto, Y., & Nishio, Y. (2004). Biodegradable cellulose diacetate-*graft*-poly(L-lactide)s:
472 Enzymatic hydrolysis behavior and surface morphological characterization.
473 *Biomacromolecules*, 5, 407-414.
- 474 Tokura, S. (1991). Analysis of chitin and chitosan. In A. Ohtakara, & M. Yabuki (Eds.). *Chitin and*
475 *chitosan: Experimental manual* (in Japanese) (pp. 49-77). Tokyo: Gihoudo.
- 476 Utracki, L. A. (1990). *Polymer alloys and blends*. Munich: Hanser Gardner Publications.
- 477 Wan, Y., Wu, H., Cao, X., & Dalai, S. (2008). Compressive mechanical properties and
478 biodegradability of porous poly(caprolactone)/chitosan scaffolds. *Polymer Degradation and*
479 *Stability*, 93, 1736-1741.
- 480 Yang, B. Y., Ding, Q., & Montgomery, R. (2009). Preparation and physical properties of chitin fatty
481 acids esters. *Carbohydrate Research*, 344, 336-342.
- 482

483 **Figure captions**

484

485 **Fig. 1.** ^1H NMR spectrum of $\text{Bu}_{2.86}\text{-Ch}$ in CDCl_3 .

486

487 **Fig. 2.** FT-IR spectra of Bu-Ch and p-Ch .

488

489 **Fig. 3.** DSC thermograms obtained for (a) $\text{Pr}_{2.76}\text{-Ch/PCL}$, (b) $\text{Va}_{2.86}\text{-Ch/PCL}$, and (c)
490 $\text{Bu}_{3.08}\text{-Ch/PCL}$ blends. Arrows indicate a T_g position taken as the midpoint of a baseline shift.

491

492 **Fig. 4.** Miscibility maps for different Acyl-Ch/PCL blends, as a function of the number N of
493 carbons in the side-chain and the substitution parameters: (a) total-DS, (b) ester-DS, and (c)
494 amide-DS. Symbols indicate that a given pair of Acyl-Ch/PCL is miscible (circle), immiscible
495 (cross), or partially miscible (triange).

496

497 **Fig. 5.** Miscibility map for different CE/PCL blends, as a function of the number of carbons in the
498 side-chain and the acyl DS. Symbols indicate that a given pair of CE/PCL is miscible (circle),
499 immiscible (cross), or partially miscible (triange).

500

501 **Fig. 6.** Schematic representation of the similarity in chemical structure between the repeating unit
502 of PCL and the butyryl side-groups in CB and Bu-Ch .

Table 1

Substitution parameters, solubility data^a, and T_g for Acyl-Ch samples

	total-DS	amide-DS	ester-DS	DD (%)	Solubility								T_g /°C
					THF	chloroform	acetone	pyridine	DMF	DMAc	DMSO	methanol	
p-Ch					–	–	–	–	–	–	–	–	NE ^b
Ac-Ch	1.89	NE	NE	NE	–	–	–	–	–	–	–	–	NE
	2.65	NE	NE	NE	–	–	–	–	–	±	–	–	NE
	3.89	NE	NE	NE	–	–	–	–	±	+	±	–	196
Pr-Ch	2.76	1.09	1.67	36.5	–	–	–	+	+	+	+	±	167
	2.89	1.27	1.62	43.1	+	+	+	+	+	+	+	+	148
	3.06	1.24	1.82	44.2	+	+	+	+	+	+	+	+	111
	3.16	1.30	1.86	44.5	+	+	+	+	+	+	+	+	140
	3.77	1.77	2.00	80.0	+	+	+	+	+	+	+	+	133
Bu-Ch	1.65	1.07	0.58	10.2	–	–	–	+	–	–	–	–	NE
	1.84	1.50	0.34	61.3	–	–	–	+	+	+	+	–	190
	2.50	1.03	1.47	18.8	±	–	+	+	+	+	+	+	175
	2.66	0.99	1.67	14.9	+	+	+	+	+	+	+	+	140
	3.08	1.19	1.89	36.7	+	+	+	+	+	+	±	+	110
Va-Ch	3.33	1.33	2.00	48.9	+	+	+	+	+	+	±	+	103
	1.64	1.31	0.33	33.6	–	–	–	–	–	–	–	–	NE
	1.82	1.17	0.66	18.6	–	±	–	±	±	±	±	–	NE
	2.32	0.97	1.35	25.5	+	+	+	+	+	+	+	+	153
	2.86	1.49	1.37	70.7	+	+	+	+	+	+	+	+	107
Ca-Ch	3.09	1.30	1.79	49.5	+	+	+	+	+	+	+	+	115
	3.48	1.48	2.00	48.6	+	+	+	+	+	+	+	±	53
	2.64	1.04	1.60	37.2	+	+	+	+	+	+	+	+	131
	2.80	1.14	1.66	42.7	+	+	+	+	+	+	+	+	110
	2.94	1.09	1.85	36.8	+	+	+	+	+	+	+	+	110
	3.42	1.42	2.00	60.9	+	+	+	+	+	+	+	±	91

^a +, soluble; ±, partially soluble; –, insoluble; temperature, 25°C; concentration, 10 mg/mL. ^b not evaluable.

Fig. 1

Fig. 2

Fig. 3

Fig. 4

Fig. 5

Fig. 6