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Continuous Tank Reactor Synthesis of Highly Substituted Sulphobutylether β-Cyclodextrins

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19   ABSTRACT

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21   Batch synthesis of Sulphobutyl ether β-cyclodextrin (also known as SBE-β-CD or

22   SBECD) is a process effectively divided into three main stages, i.e. initial reagent

23   dissolution, a sulphoalkylation reaction and final reaction quenching. This reaction is

24   followed by downstream processing and purification, and ultimate isolation of the

25   solid SBECD material. However, a feature associated with using this synthetic

26   method is that a high proportion of lower substituted SBECD is observed. There is

27   therefore a need to provide an improved synthetic method for producing higher

28   substituted cyclodextrins.

29   The authors here present a Continuous Tank Reactor (CTR) method for preparing

30   sulphobutyl ether-cyclodextrins. The method comprises first contacting cyclodextrin

31   with a base to form activated cyclodextrin. The method then involves separately

32   contacting the activated cyclodextrin with an 1,4-butane sultone to form sulphoalkyl

33   ether-cyclodextrin.

34   The activation reaction is carried out in batch synthesis mode and the

35   sulphoalkylation reaction is carried out under continuous flow conditions resulting in

36   a novel method for the synthesis of highly derivatised cyclodextrins.

37   The work is particularly concerned with producing controlled substitution in

38   sulphobutyl ether β-cyclodextrins and novel compositions of highly substituted

39   sulphoalkyl ether β-cyclodextrins are described.

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Abbreviations

ADS	Average Degree of Substitution
β-CD	β-Cyclodextrin
BS	1,4-butane sultone
CD	Cyclodextrin
CD-Screen-DAP	HPLC Stationary Phase for Analysis of Cyclodextrin-Derivatives
CTR	Continuous Tank Reactor
ELSD	Evaporative Light Scattering Detection
HPLC	High performance liquid chromatography
IDS	Individual Degree of Substitution
MPA	Mobile phase A
MPB	Mobile phase B
PTFE	Polytetrafluoroethylene
SBE-β-CD	Sulphobutyl ether β-cyclodextrin
SBECD	Sulphobutyl ether β-cyclodextrin
USP35/NF30	United States Pharmacopeia 35 and National Formulary 30
US FDA	US Food and Drug Administration

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Introduction

75 Sulphobutyl ether β-cyclodextrin (SBECD) is one of a class of polyanionic,

76 hydrophilic water soluble cyclodextrin derivatives. The parent β-cyclodextrin can

77 form an inclusion complex with certain active pharmaceutical ingredients (API) with

78 two benefits, the apparent aqueous solubility of the API increases and, if labile

79 functional groups are included, chemical stability is improved. However, the parent β-

80 cyclodextrin suffers from two problems, including lower aqueous solubility and

81 nephrotoxicity when given via injection, e.g. the intravenous route. Derivatisation of

82 β-cyclodextrin (and its variants α and γ-cyclodextrin) has been shown to be

83 beneficial with respect to both of these two defects. The first derivatised cyclodextrin

84 was the hydroxypropyl derivative, which was later followed by sulphobutyl ether (see

85 Figure 1). These two derivatised cyclodextrins are the most commercially significant.

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β-cyclodextrin   1,4-butane sultone   sulphobutylether β-cyclodextrin
                                                           SBECD

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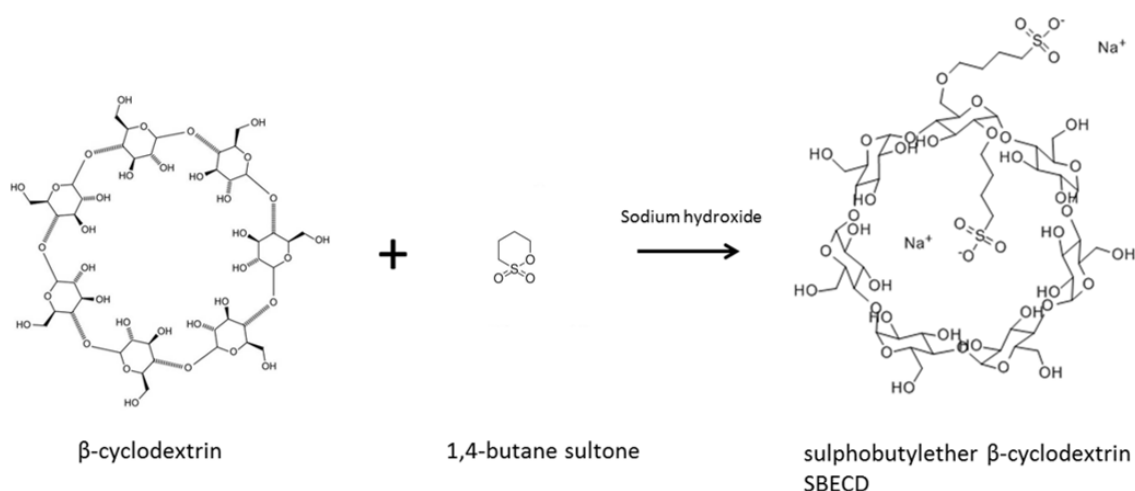
94 Figure 1: A general scheme for the synthesis of SBECD from the reagents β-cyclodextrin (β-CD)
95   and 1, 4-butane sultone (BS).

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97 SBECD is currently used as an effective pharmaceutical excipient, and has been

98 given the registered trade name Captisol. To date, there are five US FDA-approved,

99 Sulphobutyl ether β-cyclodextrin enabled drug products on the market: Nexterone

100 (Baxter International); Geodon and Cerenia (Pfizer); Kyprolis (Onyx); Abilify (Bristol

101 Myers Squibb).

102   Shah *et al* (1) has previously described a batch synthesis of SBECD, the process

103   being effectively divided into three main stages, i.e. initial reagent dissolution, a

104   sulphoalkylation reaction and final reaction quenching. The reaction is then followed

105   by downstream processing and purification, and ultimate isolation of the solid

106   SBECD material. However, a feature associated with using this synthetic method is

107   that a high proportion of lower substituted SBECD is observed. Antle (2) has also

108   described a continuous manufacturing process. However, there are significant

109   conceptual differences between our approach and that of Antle in that our approach

110   requires lower temperatures and operates at ambient pressure, and also allows for

111   controlled substitution in sulphobutyl ether β-cyclodextrins and the production of

112   novel compositions of highly substituted sulphoalkyl ether β-cyclodextrins.

113   It has been reported that the method of preparation of a cyclodextrin derivative can

114   have an impact upon the final structure (3). Previous studies have demonstrated

115   that, of the three types of hydroxyl groups present in CDs, those at the six position

116   (C6, primary hydroxyl) are the most nucleophilic, those at the two position (C2) are

117   the most acidic, and those at the three position (C3) are the most inaccessible(4,5).

118   It has also been reported that at high alkali concentration the primary hydroxyls have

119   higher reactivity than the secondary hydroxyls on C2 (6). Additionally, bulky

120   substituents prefer to react with the primary hydroxyl on C6 (6).

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122   **Methods**

123   The Continuous Tank Reactor (CTR) based Manufacturing Process.

124   The continuous flow experiments consisted of two Masterflex pumps connected to a

125   glass double 10 ml jacketed Continuous Tank Reactor (CTR). The two pumps were

126   connected to the CTR holding chamber via a three-way connector and PTFE tubing.

127   Non-return valves were fitted in line in the vicinity of the three-way connector to

128   prevent the reagent stream reverse flow as a result of the differential flow pressure in

129   either of the feed lines. The PTFE tubing was put in a water bath to maintain

130   temperature at approximately 60 ℃. In a typical experiment, a round bottom flask

131   containing a stock solution of β-cyclodextrin in NaOH solution was first prepared as

132   follows: 15 g of β-CD (1.32×10^{-2} mole) was added with stirring to an aqueous

133   solution composed of 6 g of NaOH in 30 ml water. This solution was maintained at

134   60 ℃ with a hotplate stirrer . The first pump (Figure 2) was then used to deliver stock

135   β-CD solution into the CTR where the substitution reaction takes place via the three

136   way connector, while the second pump was used to deliver neat 1,4-butane sultone

137   also held at 60 ℃ through the three way connector. An internal vortex circulation

138   was generated with the continuous flowing reaction stream and the reaction

139   proceeded in a continuous manner, i.e. once the pumps started they were not

140   switched off until completion of the reaction. The crude product was harvested in a

141   20 ml sample bottle.

142   <u>Analytical Methodology for the Analysis of High Substituted SBECD Species</u>

143   High performance liquid chromatography with evaporative light scattering detection

144   (ELSD) was used for the separation of sulphobutylether β-cyclodextrin into its

145   substituted constituents in order to determine the average degree of substitution.

146   Identification of each substituted cyclodextrin was determined by comparing the

147   retention times with materials produced by the method of Shah (1).

148   The chromatographic conditions are summarised as follows:

Instrument:	Agilent 1100 series
Software:	OpenLAB
Column:	CD-Screen-DAP, 3 µm, 150 × 4.0 mm, (CD-Screen -DAP-1504-03)
Column temperature:	25°C. ± 1°C.
Mobile phase A (MPA):	0.5% triethylamine-acetic acid buffer, pH = 5
Mobile phase B (MPB):	acetonitrile, HPLC grade
Flow rate:	1.0 ml/min

Gradient Ratio	Time (min)	0	6	15
	MPA (%)	100	50	50
	MPB (%)	0	50	50

Detection:	ELSD
Injection volume:	5 µl
Concentration:	10 mg/ml
Acquisition time:	15 minutes with post-time of 5 minutes
Needle wash:	none

149   ELSD Conditions

Instrument:	Alltech ELSD 2000
Tube temperature:	*115°C.*

Gas flow (nitrogen):   3.2 L/min
Gain: 2
Impactor: Off

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Results and Discussion

153   The authors carefully studied the batch sulphoalkyl ether β-cyclodextrin production

154   method that was described by Shah *et al* (1), and have then devised a Continuous

155   Tank Reactor Synthesis (CTR) method for producing SBECD and experimented with

156   the stoichiometry of the reaction. A significant modification to existing methods

157   comprised contacting cyclodextrin with a base to form activated cyclodextrin and

158   separately reacting the activated cyclodextrin with an 1,4-butane sultone to form

159   sulphoalkyl ether β-cyclodextrin. In our method the sulphoalkylation reaction is

160   carried out under continuous flow conditions. The resultant substituted sulphoalkyl

161   ether β-cyclodextrin is novel as it exhibits a higher degree of substitution, for a lower

162   input of 1,4-butane sultone and base than that which is produced using the known

163   batch process. The higher Average Degree of Substitution arises from the presence

164   of highly substituted species with an Individual Degree of Substitution in excess of

165   10.

166   By comparison, the batch method of preparing substituted sulphoalkyl ether β-

167   cyclodextrin produced a higher concentration of lower degrees of sulphoalkyl ether

168   β-cyclodextrin substitution than that produced using a Continuous Tank Reactor

169   method**.** Furthermore, it can be seen that material produced by the process

170   described in US 6,153,746 (1) has a range of substitution from 2 to 10, while material

171   produced in accordance with CTR processing has a range of substitution from 3 to

172   13. In addition the method does not produce any detectable di-substituted

173   sulphobutylether β-cyclodextrin and produces significant quantities of degree of

174   substitution of 11-13 not detected in the US 6,153,746 (1) material.

175   As described in the Method Section, the set-up for the continuous flow experiments

176   consisted of two pumps connected to a double Continuous Tank Reactor (CTR)

177   acting as a holding chamber/sight glass. The two pumps were connected to the CTR

178   holding chamber via a three-way connector. In a separate round bottom flask, a

179   stock solution of β-cyclodextrin in NaOH solution was first prepared and this solution

180   was maintained at 60 ℃ with a hotplate stirrer.   The sodium hydroxide was present

181   in an amount which was stoichiometrically controlled, relative to the amount of

182   cyclodextrin, to achieve a desired degree of substitution.

183   As β-cyclodextrin was added to the sodium hydroxide solution, a three stage

184   'activation' process occurred. Firstly, it takes a finite time to add the β-cyclodextrin

185   into the reservoir vessel containing aqueous sodium hydroxide. Next, the β-

186   cyclodextrin dissolves in the sodium hydroxide solution. Finally and more

187   significantly, an initial solution straw colouration progressively 'deepens' (the

188   activation process has typically taken 30 minutes) which is considered to be a visual

189   sign of reaction of the β-cyclodextrin by sodium hydroxide. With the deep

190   colouration present, and with both reagents at the specified temperature, mixing then

191   proceeded (see Figure 2).

192   Pump (1) was first turned on to feed β-CD until it reached the first chamber of the

193   CTR (4), after which pump (2) was turned on to feed heated BS into the CTR (4). An

194   internal vortex circulation was generated with the continuous flowing reaction stream

195   which ensured rapid mixing. It is important that both the aqueous, basic β-

196   cyclodextrin solution and the neat 1,4-butane sultone were heated within the range

197   50-60 °C prior to mixing.

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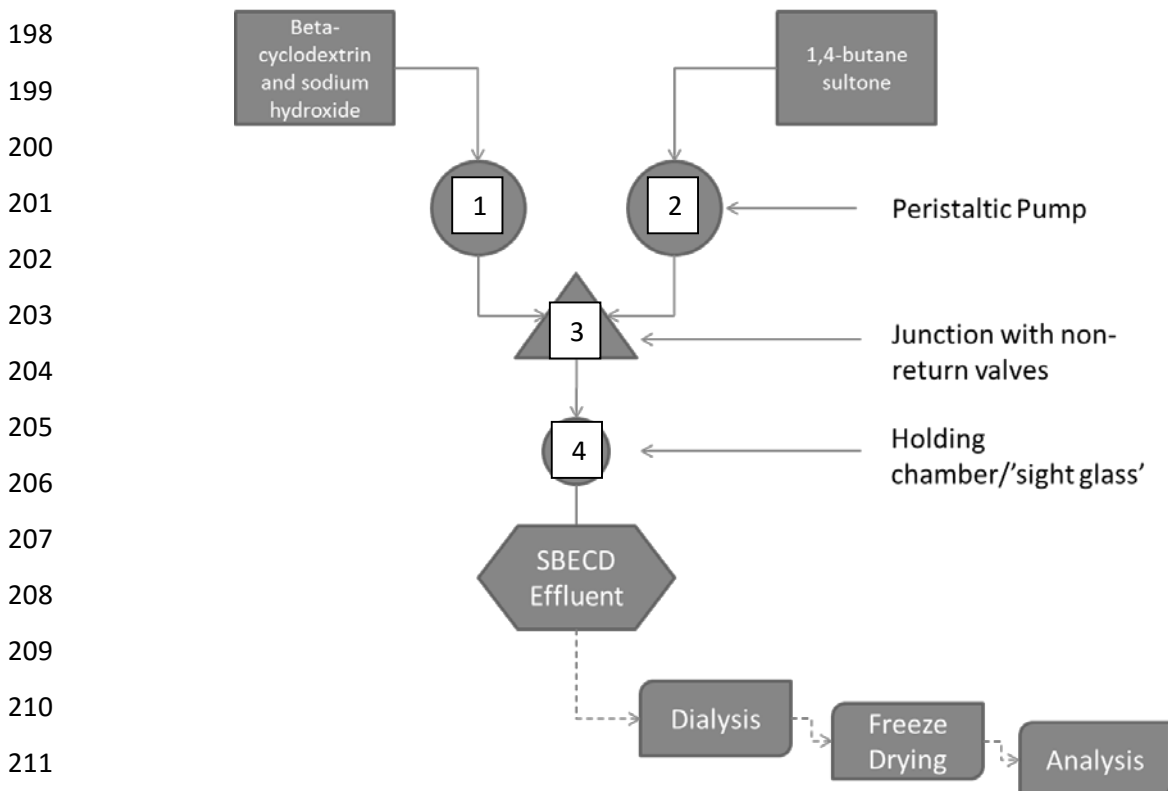
212   Figure 2: Continuous Tank Reactor Synthesis (CTR) method for producing SBECD

213 The total amount of 1,4-butane sultone was reacted to the extent that less than

214 0.1% by weight, of unreacted cyclodextrin was left. The entire initial charge of

215 cyclodextrin is thus reacted by being partially substituted. Residual cyclodextrin

216 can be monitored throughout this initial phase, for example by HPLC as

217 described below, until a desired endpoint of less than 0.1%, of residual

218 cyclodextrin starting material, has been achieved.

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221 Typical Flow rates and cyclodextrin to 1,4-butane sultone ratios are shown in

222 Table 1.

223

224 Table 1: The relationship between pump drive speed and flow rate giving rise to different butane
225 sultone-β-cyclodextrin molar ratios – constant 1,4-butane sultone flow rate.
226

	CD		BS
Drive speed(rpm)	11	15	5
Flow rate(ml/min)	0.99	1.35	0.45
Concentration Mol.min x10^{-4}	4.36	5.94	4.4×10^{-3}
[BS:CD] Mole ratio	10:1	7:1	—

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232 The reaction proceeds in a continuous manner, i.e. once the pumps have started

233 they are not switched off until completion of the reaction. The reaction takes place in

234 a temperature range of 50-60 ℃, in contrast to Antle (2) where high temperatures

235 and pressures were used. The CTR process handles the β-cyclodextrin-sodium

236 hydroxide solutions and 1,4-butane sultone as an immiscible, two phase system.

237 We have calculated that Antle's conditions, on the other hand, seem to create the

238 conditions where 1,4-butane sultone and the aqueous β-cyclodextrin-sodium

239 hydroxide streams become miscible, an enabler of flow chemistry processing.

240 Judging by the average degree of substitution achieved by Antle, the goal of

241 miscibility appears to have been achieved at the expense of 1,4-butane sultone

242 stability leading to very low degrees of substitution.

243

244 The crude product was harvested in a 20 ml sample bottle. Reaction products were

245 dialysed and lyophilized to obtain the sulphobutyl ether of β-CD as a white solid. The

246 product was initially analysed using capillary electrophoresis as described by United
247 States Pharmacopoeia 35/National Formulary 30 (7), in order to show the degree of
248 substitution. Mass spectroscopy was then carried out to show the absence of
249 unreacted β-CD and levels of 1, 4-butane sultone were analysed by gas
250 chromatography as described by Shah. The lyophilised product was weighed to give
251 the yield.

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253 The electropherogram in Figure 3 compares SBECD manufactured using our flow
254 synthesis process and a standard sample manufactured using the batch
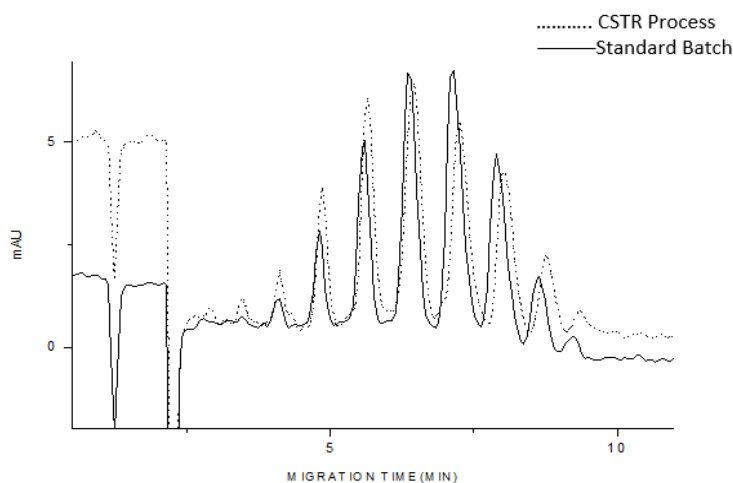255 manufacture method according to Shah (1).

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267 Figure 3: Electropherogram showing a standard sample of batch-produced SBECD according to Shah,
268 (solid line) and an SBECD Sample produced by a Flow Synthesis Process (dotted line).

269 Coincidence of the two electropherograms indicates an equivalent 'substitution
270 envelope', however it is remarkable that the flow synthesis process only requires 50
271 % of the sodium hydroxide used in the batch process and a 7:1 molar ratio of 1,4-
272 butane sultone to β-cyclodextrin instead of 10:1 used in the batch process. This
273 finding was unexpected as our entering bias was equivalent synthetic efficiency. It
274 would appear that the shielding of sodium hydroxide from 1,4-butane sultone up to

275 the point where the reactions streams mix and the reaction takes place allows for an

276 efficient activation of β-cyclodextrin hydroxyl groups at the point of the reaction with

277 minimal degradation of 1,4-butane sultone to low molecular weight by-products. In

278 short, more 1,4-butane sultone can react with β-cyclodextrin more efficiently to

279 generate higher degrees of substitution resulting in more efficient use of the starting

280 materials.

281 To test this hypothesis further, we attempted to increase the ratio of sodium

282 hydroxide to β-cyclodextrin ratio as outlined in Table 2. In a batch process,

283 according to Shah, this would have no beneficial effect on the degree of substitution,

284 i.e. a change in the substitution envelope, because the sodium hydroxide would

285 simply destroy the 1,4-butane sultone before reaction with cyclodextrin could take

286 place. In essence there is a kinetic limit to the degree of substitution under batch

287 processing conditions. Shah exploits this to reduce the residual concentration of

288 reactants upon batch reaction completion.

289

290 Table 2: Composition of sulphobutylether β-cyclodextrin determined by different reagent stoichiometries
291 produced by the CTR method.

1,4-butane sultone to β-CD molar ratio	NaOH to β-CD molar ratio	NaOH relative to Stella(11)	Average Degree of Substitution	IDS_n present in Shaw(1) and not in the CTR-produced SBECD	IDS_n present in the CTR-produced SBECD and not in Shaw(1)
7:1	9:1	-25%	6.9	None	IDS_1, IDS_{11}
7:1	11:1	0%	8.7	None	$IDS_{11} - IDS_{13}$
7:1	14:1	+25%	12.1	$IDS_2 - IDS_6$	$IDS_{11}-IDS_{14}$
10:1	6:1	-50%	6.0	None	None
10:1	9:1	-25%	6.8	None	IDS_1, IDS_{11}
10:1	11:1	0%	8.4	None	IDS_1, IDS_{11}, IDS_{12}
10:1	14:1	+25%	10.4	IDS_2	$IDS_{11} - IDS_{13}$

292

293 It can be seen from Table 2 that in general, an increase in the content of sodium

294 hydroxide will increase the Average Degree of Substitution of sulphobutylether β-

295 cyclodextrin. This observation is in agreement with an earlier report of batch type

296 systhesis by Stella (11). However, the CTR reactions produce material with an

297 Average Degree of Substitution at levels not previously seen using batch or

298 continuous flow reactions. The higher Average Degree of Substitution arises from

299 the presence of highly substituted species with an Individual Degree of Substitution
300 in excess of 10.

301 The process used to produce the material in Figure 4 requires 25% more sodium
302 hydroxide than the batch process with an increase in the molar ratio of 1,4-butane
303 sultone to β-cyclodextrin from 7:1 to 10:1. This material produced by CTR synthesis
304 is unprecedented and demonstrates a positive skew in the substitution envelope with
305 a smaller population of lower degrees of substitution (electropherogram migration
306 time range 2-7 minutes) and an increase in higher degree of substitution products
307 with migration times ranging from 6 minutes to 9 minutes. It is concluded that an
308 increase in efficiency (more efficient activation of β-cyclodextrin hydroxyl groups by
309 sodium hydroxide and less consumption of 1,4-butane sultone resulted in a higher
310 degree of substitution. It is not possible to produce highly substituted SBECD using
311 the batch process. Figure 4 shows an electropherogram of this procedure versus
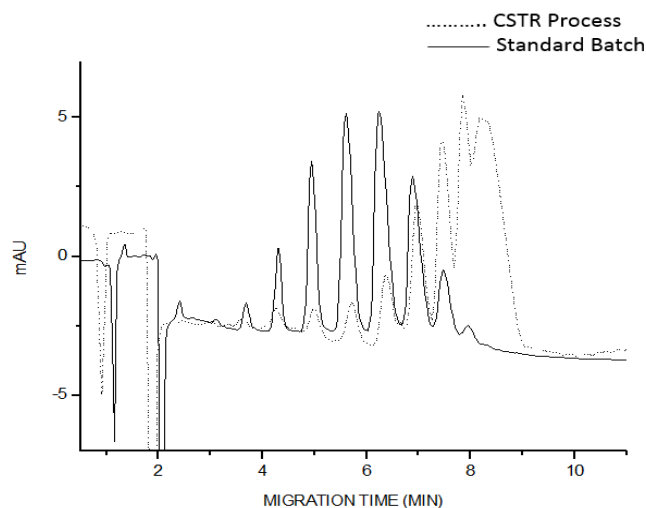312 standard batch SBECD (1).

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324 Figure 4: Electropherogram Showing a SBECD Sample produced by a Flow Synthesis Process (dotted
325 line) compared to a Standard Sample of Batch-Produced SBECD according to Shah (solid line).

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327 This method of carefully reacting sodium hydroxide with β-cyclodextrin to activate it

328 in advance of a two-phase continuous flow reaction seems to be the key to creating

329 a highly efficient reaction and a controllable average degree of substitution. The

330 activation process must be conducted at controlled temperature and for a specified

331 time after the β-cyclodextrin has dissolved in the aqueous sodium hydroxide solution.

332 The activation process has typically taken 30 minutes at this scale; the major

333 indicator of completion is the colour change which could be measured

334 colorimetrically but we have not verified this experimentally.

335 Initial pH control assures the reduction of certain by-products. It is noted that

336 acid is produced as a result of the sulphoalkylation and that the pH tends to

337 decrease as the reaction proceeds. The reaction must be maintained in basic

338 conditions because if the reaction medium is allowed to become too acidic the

339 reaction will stop and so it is important to maintain the pH of the reaction medium

340 at a level of at least 8 by adding aqueous hydroxide as needed. If the pH is

341 allowed to exceed pH 11, then the reaction starts to produce a high level of the

342 by-products 4-hydroxyalkylsulphonate and bis-sulphoalkyl ether, thus consuming

343 1,4-butane sultone. By initially monitoring pH and maintaining it within the range

344 of 8 to 11, as opposed to simply providing the full charge of hydroxide at the start

345 of the reaction, the reaction proceeds while producing a relatively low level of by-

346 products. The total amount of hydroxide added throughout the reaction was typically

347 on the order of the amount stoichiometrically required plus a 10-20 % molar excess

348 relative to the amount of 1,4-butane sultone employed. Once the sulphoalkylation

349 reaction was complete and the low residual cyclodextrin end point reached,

350 additional hydroxide can be added to destroy any residual sultone.

351

352 Although the recommended method for determination of substitution SBECD species

353 is based on a capillary electrophoresis method (Figures 3 and 4), it can be seen that

354 whilst a qualitative idea of the substitution pattern is possible, it is difficult to integrate

355 the areas under the peaks reliably due to the shifting baseline. It is also evident from

356 Figure 4 that peak resolution deteriorates with increasing substitution. Peaks appear

357 to merge after approximately 8 minutes resulting in an inability to quantify the pattern

358 of substitution. It was therefore concluded that it would not be possible to quantify

359 the degree of substitution for the new CTR process using the USP35/NF30 capillary
360 electrophoresis method.

361 Alternative methods have been proposed for the analysis of cyclodextrin derivatives
362 using high performance liquid chromatography by Szeman (8). This has been
363 recently applied to sulphobutylether β-cyclodextrin (9). The method is based on a
364 specialized ion-exchange HPLC column, CD-Screen-DAP, using a bonded dimethyl
365 amino phenyl function to improve the selectivity of the analytical method. The
366 analysis of sulfobutyl ether-beta-cyclodextrin mixtures by ion-spray mass
367 spectrometry and liquid chromatography-ion-spray mass spectrometry has also been
368 reported by Grard et al (10).

369 High performance liquid chromatography with evaporative light scattering detection
370 (ELSD) was used for the detection of the separation of sulphobutylether β-
371 cyclodextrin into its substituted constituents in order to determine the average
372 degree of substitution. Identification of each substituted cyclodextrin was determined
373 by comparing the retention times of the standard, produced by the method described
374 in US 6,153,746 (1), with that of a material produced using our CTR processing
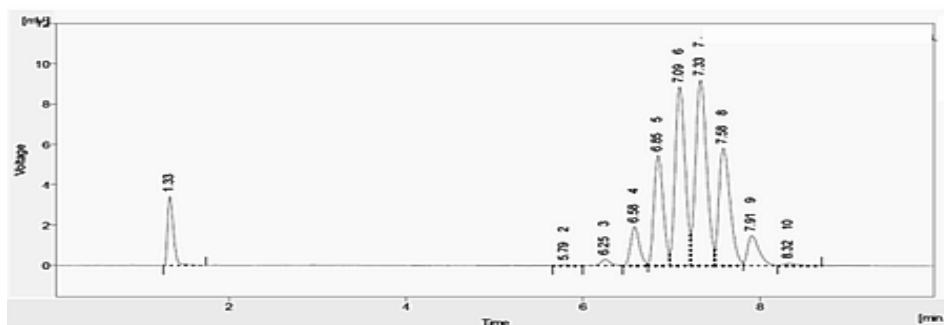375 method. A typical chromatogram for the standard material is given in Figure 5:

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382 Figure 5: Chromatogram of sulphobutylether β-cyclodextrin produced by the method described in
383 US 6,153,746 (Shah, 2000). HPLC conditions are based on a gradient separation with a CD-
384 Screen-DAP column and ELSD detection.

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389　The chromatogram for the material corresponding to CTR processing with ratios of
390　1,4-butane sultone to β-cyclodextrin of 10:1 are shown in Figure 6.

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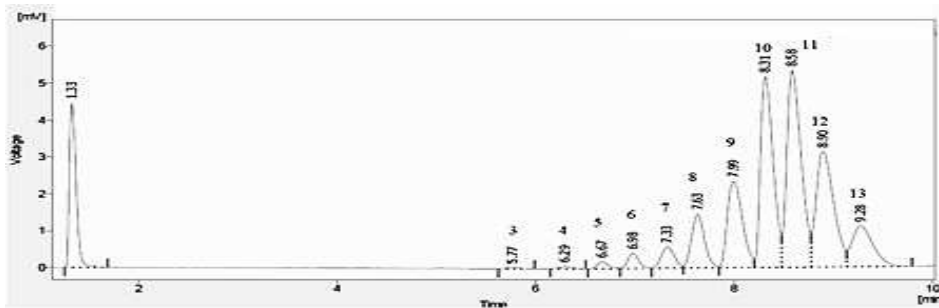397　Figure 6: Chromatogram of sulphobutylether β-cyclodextrin produced by the CTR method.

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400　Upon further examination of Figure 5, it can be seen that material produced by the

401　process described in US 6,153,746 (1) has a range of substitution from 2 to 10.

402　Figure 6 indicates that material produced in accordance with CTR processing has a

403　range of substitution from 3 to 13. In addition the method does not produce any

404　detectable di-substituted sulphobutylether β-cyclodextrin and produces significant

405　quantities of degree of substitution of 11-13 not detected in the US 6,153,746 (1)

406　material.

407

408　From the chromatographic data, an Individual Degree of Substitution (IDS_n) can be

409　calculated using the following formula:

410　　　　　　　　$IDS_n = (PA_n/\sum PA) \times 100$　　　　　　　Equation (1)

411

412　　　　　　　　where $\sum PA = \sum PA_L + PA_{L+1} \dots PA_H$　　　Equation (2)

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414　　　　n = Substitution Number

415　　　　PA = Peak area

416　　　　PA_L = Peak area corresponding to lowest degree of substitution seen on

417　　　　　　the chromatogram

418　　　　PA_H = Peak area corresponding to highest degree of substitution seen on

419　　　　　　the chormatogram

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421 These data can be used to describe an 'Envelope of Substitution' which is used

422 as the basis of a specification element in USP30/NF30 (7), where each IDS_n

423 should fall within a Proven Acceptable Range.

424 Table 3 shows calculated data based on the chromatogram shown in Figure 6.

425 This was processed using Equations 1-2 to give an Individual Degree of

426 Substitution (IDS_n).

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429 Table 3: Calculated Individual Degree of Substitution (IDS_n).

Substitution Number:	Retention Time:	Peak Area:	IDSn
3	5.77	0.271	0.133
4	6.29	0.507	0.248
5	6.67	1.455	0.712
6	6.98	3.142	1.537
7	7.33	5.221	2.553
8	7.63	13.283	6.496
9	7.99	24.842	12.148
10	8.31	46.056	22.528
11	8.58	53.920	26.368
12	8.90	39.220	19.180
13	9.28	16.570	8.103

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433 The Individual Degree of Substitution metrics are then used to calculate the

434 Average Degree of Substitution as follows:

435

436 $$ADS = \sum(IDS_n \times n)/ 100 \qquad \text{Equation (3)}$$

437

438 The material described in Figure 6 has an average degree of substitution (ADS) of

439 10.4 which is substantially higher than material produced by batch manufacture (1)

440 or Antle's flow process (2) which typically results in ADS values of 6 to 7.

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Conclusions

The Continuous Tank Reactor method of synthesis resulted in a lower concentration of lower substituted sulphoalkyl ether β-cyclodextrin (i.e. a degree of substitution value of 1-3) and surprisingly much higher concentrations of the higher substituted sulphoalkyl ether β- cyclodextrin (i.e. an average degree of substitution value of 3-13) than reported using more standard batch or flow techniques. The CTR process depends upon pre-activation of the β-cyclodextrin feedstock by sodium hydroxide where the extent of activation determines Average Degree of Substitution. The process allows greater control of the Average Degree of Substitution by varying sodium hydroxide concentration. The process can be used to produce material with a high Average Degree of Substitution, the utility of which is currently under investigation (12). It should also be possible to manufacture material compliant with the USP35/NF30 specification for sulphobutylether β-cyclodextrin, the current article of commerce, if that is desired. Using an improved HPLC analytical method, we have been able to validate these general observations. The technique has allowed us to produce descriptive statistics for highly substituted materials. The composition, produced by the CTR process, is novel in two respects: an unprecedented high average degree of substitution and the existence of highly substituted species with IDS_n values higher than 10.

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