The stability research of different types of sodium butyrate in course of processing

CHENG Guo-shun[1] XU Zhen-fei[1] YU Rong[2] ZHAO Fang-fang[1,2] XU Xiao-fang[2]
1 College of Animal Science and Technology, Gansu Agricultural University, Gansu Lanzhou, 730070
2 National Engineering Laboratory of Biological Feed Security and Contamination Control, Test Base of Biological Coating Engineering, Zhejiang Hangzhou, 311107

Abstract: The objective of this study is to research the stability of sodium butyrate in feedstuff. The sodium butyrate with three types of microencapsulated, powdery and granular was produced at three different factories and sampled at different production links and determined the content. The results showed that with increasing of moisture in feedstuff, the loss rate of sodium butyrate had increased. The loss rates of microencapsulated sodium butyrate of different moisture in feedstuff were less than 2.0%. The optimized sequence of premix carrier determined by the stability research was Rice hull powder, Wheat-middlings, $Ca(HCO_3)_2$, Zeolite and $CaCO_3$. The trace elements with high dosage and choline chloride with strong hygroscopicity would consume and damage feed nutrients, but the effect would be improved after coating. Under the same pressure, the loss rate of sodium butyrate increased with increasing of pelleting temperature. The powdery and granular sodium butyrate were decomposed in artificial gastric juice and intestinal juice, but the microencapsulated sodium butyrate gradually released in gastrointestinal tract and made more sodium butyrate reach hindgut.

Key words: feed, sodium butyrate, coating, stability, retention rate

1. Introduction

The short chain fatty acids have kinds of functions in the body, and the butyrate plays a major role. Due to the volatility of butyrate, the effect cannot be guaranteed when added into the feed directly, therefore, sodium butyrate is now commonly used. The sodium butyrate is the main energy source for proliferation of intestinal mucosal epithelial cells, it can promote growth and development of intestinal mucosal, enhance mucosal permeability, thereby, enhance digestion and absorption of small intestine. The sodium butyrate can repair the impaired colonic epithelial cells, regulate water and salt balance and reduce diarrhea, and has good effects in practical application. However, sodium butyrate has a special odour and bad fluidity, and is easy to absorb moisture, these make it difficult to process, transport and use, and consequently limit its wide use. What people more worried about is that the sodium butyrate will be lost contribute to the storage, mixing, granulation, changes in ration formulas, and changes in temperature and humidity when producing and using products, at the same time, the effects of sodium butyrate will be decreased and some other instable raw materials will be lost. The objective of this study is to research the loss rates of microencapsulated sodium butyrate, powdery sodium butyrate and granular sodium butyrate during processing and provide references for application.

2. Materials and methods

2.1 Materials and experimental locations

Microencapsulated sodium butyrate (30% sodium butyrate) and powdery sodium butyrate (98% sodium butyrate) was provided by Hangzhou King Techina Feed Co., Ltd. Trade name of microencapsulated sodium butyrate is CM3000. Granular sodium (70% sodium butyrate) was purchased from the market. The production of feed was completed at three feed companies: Jiuquan Tianhe Feed Co., Ltd., Baiyin Yellow River Feed Co., Ltd., and Lanzhou Fuchang Feed Co., Ltd.

2.2 Experimental design

The three companies used the same feed formula (the feed ingredients except corn were supplied by the same supplier, and corn with similar moisture and unit weight was put into production after optimization of formula.). 1000 g/t of three types of sodium butyrate was added into feed respectively. Feed sample were packed and stored according to regular procedure. Each link of each factory produced 2 batches of feed, and 5 samples of each batch were sampled for determination. The contents of sodium butyrate were determined at 7, 14, 15 and 30 d. The experimental design was shown in table 1.

Table 1 Experimental design

Treatment	1	2	3
Type	CM3000	Powdery sodium butyrate	Granular sodium butyrate
Content of sodium butyrate	30%	98%	70%
Actual content per ton (g)	300	980	700

2.3 Experimental method

The samples were taken back to laboratory of Gansu Agricultural University within 2 d. The actual contents of sodium butyrate were determined. The dissolution rates of sodium butyrate in stomach and small intestine were determined by RC-8DS dissolution rate tester according to the determination of gradual and controlled-release of microcapsule. The moisture was determined by GB/T 6435-2005/ ISO6496:1999. The mixing uniformity of feed should be kept within 7% which was determined by GB/T 5918-2009.

2.3.1 Determination of content of sodium butyrate

2.3.1.1 Reagent and instrument

Reagent: glacial acetic acid (AR), acetic anhydride (AR), crystal violet indicator (took 0.5 g crystal violet, dissolved into 100 ml acetic acid), perchloric acid standard solution (0.1 mol/L).

Instrument: electronic balance, burette

2.3.1.2 Experimental procedure and computing method

Accurately weighed 0.1 g sodium butyrate and 20 ml glacial acetic acid, mixed with 5 ml acetic anhydride, after complete dissolution, titrated with perchloric acid standard solution until the solution turned green. A blank correction was made at the same time.

Formula: X=(V-V0)*C*0.11010/M*100

V: the volume of perchloric acid standard solution consumed by sample (ml)

V0: the volume of perchloric acid standard solution consumed by blank (ml)

C: the actual concentration of perchloric acid standard solution (mol/L)

M: the sample weight

0.11010: equivalent with 1.00 ml perchloric acid standard solution ($CHClO_4$=0.1 mol/L), i.e. the weight of sodium butyrate expressed as gram.

2.3.2 Determination of dissolution rate of sodium butyrate in gastric and intestinal juice

2.3.2.1 Preparation of artificial gastric juice and intestinal juice

Preparation of gastric juice: took 16.4 ml HCl, added about 800 ml water and 10 g pepsin, shook up, then added water to 1000 ml (Chinese pharmacopoeia, 2005, part II, page 72 of appendix).

Preparation of intestinal juice: took 6.8 g KH_2PO_4, dissolved in 500 ml water, then regulated pH value to 6.8 with 0.1 mol/L NaOH. Took 10 g pancreatin and dissolved with water. Mixed the two solutions and added water to 1000 ml (Chinese pharmacopoeia, 2005, part II, page 158 of appendix).

2.3.2.2 Determination of dissolution rate in gastric and intestinal juice

Determination of dissolution rate in gastric juice: 250 ml degassed artificial gastric juice was injected into each container and kept the temperature of solvent at 37±0.5℃ æ 60±1 rpm. Weighed 5 samples of 5 g each, put them into 5 containers respectively, rotated immediately. Samples were took at 0, 1, 2, 4 and 8 h respectively, filtrated with filter paper and dried under low temperature until reached constant weight. The content of sodium butyrate was determined by hydrochloric acid titration. The dissolution rate was calculated.

Determination of dissolution rate in intestinal juice: 250 ml degassed artificial intestinal juice was injected into each container and kept the temperature of solvent at 37±0.5℃ æand 60±1 rpm. Weighed 7 samples of 5 g each, put them into 7 containers respectively, rotated immediately. Samples were took at 0, 2, 4, 6, 8, 16, 32 and 48 h respectively, filtrated with filter paper and dried under low temperature until reached constant weight. The content of sodium butyrate was determined by hydrochloric acid titration. The dissolution rate was calculated.

2.4 Experimental setting

2.4.1 Effect of moisture on content of sodium butyrate

The moisture of feed was controlled by water addition equipment and corn. Water and mildew was added according to recommendations. Feed samples were packed and stored according to regular procedure. Each link of each factory produced 2 batches and 500 kg of each batch, 5 samples of each batch and 500 g of each sample were sampled for determination. The contents of sodium butyrate were determined at 15 and 30 d. The feed formula was shown in table 2.

Table 2 Feed formula of effect of moisture on content of sodium butyrate (kg/t)

Ingredient	Proportion	Ingredient	Proportion
Corn	400.0	Limestone	7.0
Extruded corn	250.0	CaHPO4	14.0
Soybean meal, 46%	150.0	NaCl	2.0
Fish meal (Peru)	30.0	Choline-50%	1.0
Extruded full fat soybean	80.0	Lys-98%	3.0
Glucose	45.0	Met	1.0
Acidifier	4.0	Thr	1.0
Sodium butyrate *	1.0	Try	0.0
Compound enzyme	1.0	Premix	10.0
Total			1000.0

Note : * stand for types of sodium butyrate can be changed according to need.

Table 3 1% premix formula（kg/t）

Raw material	Nutrient element	Standard of raw material	Content of effective element	Proportion
$CuSO_4•5H_2O$	Cu	98.40%	25.00%	20.5
$FeSO_4$	Fe	91.30%	30.00%	30.5
$ZnSO_4$	Zn	96.00%	35.00%	26.2
$MnSO_4$	Mn	97.50%	31.70%	12.5
KI	I	5.00%	3.80%	1.3
$CoCl_2$	Co	5.00%	1.20%	15
Na_2SeO_3	Se	2.20%	1.00%	3.5
Pyridine carboxylic acid chromium	Cr	9.90%	1.23%	2.5
Quinocetone			100%	8.5
Compound vitamin Vm111			100%	30
Antioxidant		66.00%	66.00%	0.6
Carrier *				849
Total				1,000

Note: * stands for types of carrier can be changed according to need. The moisture of carrier is less than 8%.

2.4.2 Effect of premix carriers on sodium butyrate

The premix was produced by table 3. Each link of each factory produced 2 batches and 100 kg of each batch, 5 samples of each batch and 500 g of each sample were sampled for determination. Feed samples were packed and stored according to regular procedure. The contents of sodium butyrate were determined at 7 and 14 d.

2.4.3 Effect of specific raw material of premix on sodium butyrate

The premix was produced by table 3. The premix of high copper was produced according to table 3 by increasing the copper levels to 250 mg/kg of compound feed, and deducting redundant part from carrier. The production of high zinc diet and high copper and high zinc premix were the same as production of high copper premix. The premix of choline was produced according to table 2 by increasing the choline levels amount to 100% of compound feed and deducting redundant part from

Table 4 Parameter of granulation and conditioning

Granulation parameters			Conditioning parameters			Granulation
Diameter of die hole (mm)	Length to diameter ratio	Length of granule (mm)	Vapour pressure (Mpa)	Conditioning feed volume (r/m)	Conditioning temperature (℃)	Granulation current (A)
3.0	11	3-8	0.2-0.4Mpa	280-350	78-85	150-170

carrier. The copper chloride and zinc chloride substituted for copper sulfate and zinc sulfate of compound feed respectively, and remained copper and zinc levels unchanged. Each link of each factory produced 2 batches and 100 kg of each batch, 5 samples of each batch and 500 g of each sample were sampled for determination. Feed sample were packed and stored according to regular procedure. The contents of sodium butyrate were determined at 7 and 14 d.

2.4.4 Effect of productive technology on sodium butyrate

The productive technology and technological parameter were shown in table 2 and 4 respectively.

2.4.5 Dissolution rates of sodium butyrate in gastric and intestinal juice

The premix was produced by table 2 and the carrier was rice hull powder. Each link of each factory produces 2 batches and 500 kg of each batch, 5 samples of each batch and 500 g of each sample were sampled for determination. Feed sample were packed and stored according to regular procedure. The dissolution rates of sodium butyrate in gastric and intestinal juice were determined at 7 and 14 d respectively.

Table 5 Effect of moisture on retention rate of different types of sodium butyrate

Moisture content	10~12%		12~14%		14~16%		Above 16%	
	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)
Powdery sodium butyrate	828.1±59.22	92.0	810.5±60.14	90.1	783.3±56.02	87.0	612.0±18.45	68.0
Granular sodium butyrate	651.4±37.22	93.1	644.6±37.89	92.1	616.6±47.16	88.1	504.3±37.24	72.0
CM3000	297.4±222.31	99.1	297.5±23.44	99.2	294.9±23.69	98.3	294.3±24.12	98.1

3. Results

3.1 Effect of moisture on different types of sodium butyrate

From the table 5 we know that the loss rates of powdery, granular and CM3000 were 8.0%, 6.9% and 0.9% respectively when the moisture of feed was 10~12%; those were 9.9%, 7.9% and 0.8% respectively when the moisture of feed was 12~14%; those were 13.0%, 11.9% and 1.7% respectively when the moisture of feed was 14~16%; those were 32.0%, 28.0% and 1.9% respectively when the moisture of feed was above 14~16%. The retention rate of CM3000 was the highest and loss rate was less than 2.0% under the different moisture.

Table 6 Effect of premix carrier on retention rate of different types of sodium butyrate

Type of carrier	$CaCO_3$		Zeolite		$Ca(HCO_3)_2$		Wheat-middlings		Rice hull powder	
	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)
Powdery sodium butyrate	801.0±65.03	89.0	846.0±102.31	94.0	864.9±78.13	96.1	870.0±78.00	96.7	870.0±45.39	97.9
Granular sodium butyrate	647.5±50.12	92.5	665.0±34.27	95.0	672.9±34.20	96.1	681.7±54.12	97.4	686.3±56.42	98.0
CM3000	297.0±12.13	99.0	298.8±2.14	99.6	298.2±23.44	99.4	298.8±8.44	99.6	300.0±0.00	100.0

3.2 Effect of premix carrier on retention rate of different types of sodium butyrate

From the table 6 we know that the retention rates of different types of sodium butyrate changed with the carriers changed. The loss rates of powdery, granular and CM3000 were 11.0%, 7.5% and 1.0% respectively when the carrier was $CaCO_3$; those were 6.0%, 5.0% and 0.4% respectively when the carrier was limestone; those were 3.9%, 3.9% and 0.6% respectively when the carrier was $Ca(HCO_3)_2$; those were 3.3%, 2.6% and 0.4% respectively when the carrier was wheat-middlings; those were 2.1%, 2.0% and 0.0% respectively when the carrier was rice hull powder. From the above, the retention rates of different types of sodium butyrate were the highest when the carrier was rice hull powder and the stability of CM3000 was best.

Table 7 Effect of premix on retention rate of different types of sodium butyrate

Type of raw material	High-Cu formula		High-Zn formula		High-Cu-Zn formula		Formula mixed with choline		Formula with copper chloride and zinc chloride	
	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)
Powdery sodium butyrate	801.0±32.77	89.0	855.9±79.12	95.1	798.3±99.32	88.7	852.3±27.79	94.7	862.2.0±54.27	95.8
Granular sodium butyrate	646.7±45.63	92.3	662.2±44.36	94.6	639.8±47.89	91.4	667.8±48.13	95.4	673.4±66.00	96.2
CM3000	297.3±17.14	98.9	297.3±12.57	99.1	298.5±33.01	99.5	289.8±6.99	99.6	298.8±14.12	99.6

Note: the Cu levels in high-Cu formula is 250ppm; the Zn levels in high-Zn formula is 2250ppm; the Cu and Zn levels in high-Cu-Zn formula are 250ppm and 2250ppm respectively; the Cu and Zn levels in other feed are negligible in this experiment.

3.3 Effect of premix on different types of sodium butyrate

From the table 7 we know that the retention rate of sodium butyrate changed when raw material increased or supplemented with interactional material. The loss rates of powdery, granular and CM3000 in high-Cu formula were 11.0%, 7.7% and 1.1% respectively; those in high-Zn formula were 4.9%, 5.4% and 0.9% respectively; those in high-Cu-Zn formula were 11.3%, 8.6% and 0.5% respectively; those in formula with choline were 5.3%, 4.6% and 0.4% respectively; those in formula with copper chloride and zinc chloride were 4.2%, 3.8% and 0.4% respectively. The retention rates of CM3000 were the highest and related to high additive amount of raw material and mix with interactional material. The loss rate of CM3000 was about 1.0%.

Table 8 Effect of pelleting technology on retention rate of different types of sodium butyrate

Types of sodium butyrate	Pelleting temperature 78℃		Pelleting temperature 81℃		Pelleting temperature 85℃	
	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)
Powdery sodium butyrate	295.7±33.12	32.9	288.7±12.69	32.1	281.4±17.98	31.3
Granular sodium butyrate	444.6±27.98	63.5	429.5±45.34	61.4	422.0±18.99	60.3
CM3000	270.1±32.14	90.0	269.0±20.12	89.7	268.6±21.04	89.5

3.4 Effect of pelleting technology on different types of sodium butyrate

From the table 8 we know that the pelleting technology was the biggest factor to affect nutrient content. The loss rates of powdery, granular and CM3000 were 67.1%, 36.5% and 10.0% respectively when the average pelleting temperature was 78℃ æ those were 67.9%, 38.6% and 10.3% respectively when the average pelleting temperature was 81℃ æ those were 68.7%, 39.7% and 10.5% respectively when the average pelleting temperature was 85℃ æIn conclusion, the pelleting temperature affected retention rate of different types of sodium butyrate and the retention rate was the highest at 78℃ æThe retention rate of CM3000 was the highest and about 10.0%.

3.5 Retention rate of different types of sodium butyrate in gastric and intestinal juice

The retention rates and loss rates of different types of sodium butyrate in gastric and intestinal juice were determined by dissolution tester. The result was shown in table 9.

Table 9 Retention rate and loss rate of sodium butyrate in gastric and intestinal juice

Types of sodium butyrate	Gastric juice (4h)Intestinal		juice (8h)	
	Retention quantity (g)	Retention rate (%)	Retention quantity (g)	Retention rate (%)
Powdery sodium butyrate	487.8±39.17	54.2	322.2±26.19	35.8
Granular sodium butyrate	373.8±27.844	53.4	211.4±17.42	30.2
CM3000	295.2±33.12	98.4	175.2±13.13	58.4

From the table 9 we know that the retention rates and loss rates of sodium butyrate in gastric and intestinal juice were determined by simulation of gastrointestinal tract of piglet. The loss rates of powdery, granular and CM3000 in gastric juice were 45.8%, 46.6% and 1.6% respectively; those in intestinal juice were 64.2%, 69.8% and 41.6% respectively. The loss rates of powdery and granular sodium butyrate in gastric juice were relatively higher and closed to 50%, the loss rates in intestinal rate were closed to 70%. But the loss rate of CM3000 in gastric juice was just 1.6%, the absorptivity of CM3000 in intestinal juice was 41.6%.

4. Discussion

The moisture of feed will indirectly reflect feedstuff quality. The moisture was different because the type and source of raw materials were different, thus the moisture in productive process and products were different, and the nutritive value and storage time were affected[4]. The high-moisture would soften the nutrients and thus cause damages to oxide and micro-elements, and would also lead to pH reaction, oxidizing reaction and catalytic reaction. When the moisture was 10-12%, 12-14%, 14-16% and above 16%, the loss rate of powdery sodium butyrate was 8.0%, 9.9%, 13.0% and 32.0% respectively; the loss rate of granular sodium butyrate was 6.9%, 7.9%, 11.9% and 28.0% respectively; and the loss rate of CM3000 was 0.9%, 0.8%, 1.7% and 1.9% respectively. The loss rare of sodium butyrate increased with increasing of moisture, so we need pay attention to the moisture in feed when using sodium butyrate.

Carrier was one of the most important conditions in productive technology of premix. Nowadays, the carriers of premix included organic carrier (rice hull powder, wheat bran, wheat-middlings, corn gluten meal, etc.) and inorganic carrier (limestone, zeolite, maifanitum, etc.). Although carrier itself was not the active part of premix, it would considerablely affect quality of premix. The carrier would cover characteristics of other materials after mixing, each property of carriers would affect other composition in premix or whole quality of premix. The destructive degree to premix from strong to weak was carrier, micro-element, choline chloride[5, 6]. When the carriers were $CaCO_3$, zeolite powder, $Ca(HCO_3)_2$, wheat-middlings and rice hull powder, the loss rates of powdery sodium butyrate were 11.0%, 6.0%, 3.9%, 3.3% and 2.1% respectively; the loss rates of granular sodium butyrate were 7.5%, 5.0%, 3.9%, 2.6% and 2.0% respectively; the loss rates of CM3000 were 1.0%, 0.4%, 0.6%, 0.4% and 0.0% respectively. The order of optimized carrier was rice hull powder, wheat-middlings, $Ca(HCO_3)_2$, zeolite, and $CaCO_3$. The effect of neutral carrier was better than effect of inorganic carrier. The stability of $CaCO_3$ was the worst as it contained lots of carbonate ion. But as carrier of premix, it needs to consider the whole unit weight and cost, rice hull powder combined with zeolite was a better choice. The retention rate of CM3000 was highest in this experiment. The loss rate of CM3000 was less than 1.0%.

The high-Cu could decrease the absorption of zinc and ferrum. The unstable materials (VA, VE, VD, riboflavin, lipid, etc.) in feed were oxidized, thus nutritive value and palatability of feed were decreased[7]. The compound with different microelement had different capacity of oxidation and reduction. The sulfate with high solubility in water is easy to absorb moisture and ionize, thus its destructive effect was far greater than carbonate and oxide, and the destructive effect of sulfate was often ignored in production. The greatest feature of choline chloride was hygroscopicity. If 20% choline chloride was added into 1% premix, the moisture in premix increased by about 3%, the free water in hydrated salt increased when choline chloride existed in premix[8]. The choline chloride could strongly absorb moisture and CO_2 in air which would cause damages to micro-element and carrier. Jaskiewicz (1998) reported that when premix with choline chloride, especially when carrier was limestone, would cause greater loss to vitamin. $CuCl_2$ with high-Cu, undissolved in water, easy to dissolve in neutral salt and acid solution, low hydroscopicity makes it rapidly dissolve in gastrointestinal tract, enhance absorption and utilization, and decrease caking and destruction to raw material and some vitamin during productive process[9]. The result showed that the loss rates of powdery, granular and CM3000 in high-Cu formula were 11.0%, 7.7% and 1.1% respectively; the loss rate of powdery, granular and CM3000 in premix with choline chloride were 5.3%, 4.6% and 0.4% respectively. The trace elements with high dosage and choline chloride with strong hydroscopicity would consume and damage feed nutrient, but the effect would be improved after coating.

The productivity and quality of feed production were related to characteristics of pelleting equipment, and to a larger extent depended on pelleting properties of raw material and pelleting technology, thus lead to large differences between productivity and quality of production[10]. When the pelleting temperature ranged from 78℃ to 81℃ to 85℃, the loss rate of powdery sodium butyrate ranged from 67.1% to 67.9% to 68.7% respectively, the loss rate of granular of sodium butyrate ranged from 36.5% to 38.6% to 39.7%, the loss rate of CM3000 ranged from 10.0% to 10.3% to 10.5%. Under the same pressure, the loss rate of sodium butyrate increased with increased pelleting temperature.

Sodium butyrate as an energy source for animal's intestinal tract could increase the intestinal immunity and balance of colonic microflora. The sodium butyrate was wasted by digesting in stomach. The results of dissolution rates in artificial gastric and intestinal juice showed that the powdery and granular sodium butyrate were decomposed in artificial gastric and intestinal juice, but CM3000 was gradually released. The loss rates of powdery and granular sodium butyrate in gastric juice were 45.8% and 46.6% respectively, it was relatively higher and closed to 50%, the loss rates in intestinal rate were 64.2% and 69.8% respectively, closed to 70%. But the loss rate of CM3000 in gastric juice was just 1.6%, the absorptivity of CM3000 in intestinal juice was 41.6%. The dissolution rate of sodium butyrate in gastric juice decreased after coating, most of sodium butyrate can reach intestinal tract and play its physiological action.

5. Conclusions

(1) The loss rate of sodium butyrate increases with increased moisture in feed. The effect of moisture in feed on sodium butyrate while using needs to be concerned. The retention rate of CM3000 is highest, and the loss rate is less than 2.0% under different moisture.

(2) The optimized order of premix carrier is rice hull powder, wheat-middlings, zeolite, and $CaCO_3$. The effect of neutral organic carrier is better than that of inorganic carrier. The stability of $CaCO_3$ is bad because it contains carbonate ion. The retention rate of CM3000 is highest and less than 1.0% in this experiment.

(3) The trace elements with high dosage and choline chloride with strong hygroscopicity will consume and damage feed nutrient, but the effect will be improved after coating.

(4) Under the same pressure, the loss rate of sodium butyrate increases with increasing of pelleting temperature.

(5) The powdery and granular sodium butyrate are decomposed in artificial gastric and intestinal juice, but CM3000 gradually releases and makes the most of sodium butyrate reach hindgut.

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HANGZHOU KING TECHINA FEED CO., LTD.
Add: No.8 Yingxin Road,Renhe Street,Yuhang District,Hangzhou,China
Tel: +86-571-88234535 Fax: +86-571-86039759 www.kdqtechina.com
Email: shirley@kdqchem.com export@kdqchem.com ljx@kdqchem.com