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## Stability of liquid lysine in feed processing

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**SUMMARY** - The stability of a liquid preparation of free base L-Lysine (L-Lysine concentration 50%) was tested under conditions of feed processing with expander at 90, 110 and 130°C in a growth trial with broilers from 8 to 35 days of age. Non-expanded feed resp. application of the liquid lysine post expanding served as control. Total dietary lysine content was set at 1.00% (90% of NRC 1994; 77% of Austic 1994 recommendation) to make lysine deficient diets. Two wheat-soyabean meal diets, without and with molasses addition have been studied. In diets without molasses addition the treatment at 90°C resulted in the same performance as with the untreated feed resp. the feed with post expanding application of the liquid lysine. The treatment at 110 and 130°C caused reduction in liveweight gain but no differences in feed conversion rate. In the diets with molasses addition the increase in processing temperature showed reduction in feed intake combined with a non-significant decrease in liveweight gain. Feed conversion rate demonstrated a high level of lysine efficiency even after high processing temperatures. Apparent faecal digestibility of total dietary lysine was high ( $89.01\% \pm 1.27$ ) and N-retention, calculated from carcass analyses did not differ significantly between treatments. Addition of molasses to the diet did not decrease availability of dietary lysine. The results lead to the conclusion that free base liquid L-Lysine is stable under the tested High-Temperature-Short-Term feed processing conditions.

**Key words:** Broiler, performance, lysine, feed processing.

**RESUME** - "Stabilité d'une préparation liquide de la L-Lysine base". La stabilité d'une préparation de L-Lysine base liquide (concentration de 50% de L-Lysine base) a été testée sur des poulets de chair entre 8 et 35 jours avec de l'aliment soumis à un expandeur à 90, 110 et 130°C. Un aliment non soumis au traitement de l'expandeur ainsi qu'un aliment avec addition de la préparation de lysine après passage par l'expandeur ont servi comme témoin. La teneur en lysine totale a été définie à 1,00% de l'aliment (90% de la recommandation NRC 1994 ou 77% de la recommandation de Austic 1994) afin d'obtenir un aliment limitant en lysine. Deux compositions d'aliment céréales-tourteaux de soja (avec ou sans mélasse) ont été étudiées. L'aliment sans mélasse expansé à 90°C donne le même résultat de performance que l'aliment non traité et l'aliment où la préparation de lysine liquide a été ajoutée après l'expandeur. Les aliments sans mélasse traités à 110 et 130°C montrent des réductions de la croissance journalière mais pas de différences de l'indice de consommation comparés avec les aliments de contrôle. Sur les performances avec l'aliment contenant de la mélasse nous observons avec l'augmentation de la température de traitement une réduction de la consommation allée à une croissance journalière moins élevée non significative. L'indice de consommation montre un haut niveau d'efficacité de lysine même avec le traitement à haute température. La digestibilité fécale apparente de la lysine totale a été de haut niveau ( $89,01\% \pm 1,27$ ) ainsi que la rétention d'azote évaluée au niveau carcasse et elle n'ont pas donné de différences significatives. L'addition de la mélasse n'a pas diminué la disponibilité de lysine. Les résultats mènent à la conclusion que cette préparation de L-Lysine base liquide 50% est stable dans les conditions de fabrication testées dans cet essai.

**Mots-clés :** Poulet de chair, performances, lysine, traitement de l'aliment.

### Introduction

High-Temperature-Short-Term (HTST) feed processing became common practice in the feed industry worldwide. Advantages coupled with this technology are multiple, e.g., improvements in pellet quality, plant capacity, flexible use of available raw materials, decontamination of pathogens and improvements in dietary metabolisable energy contents (Peisker, 1993). On the other hand concerns about nutrient stability have been constantly addressed by users of HTST, namely vitamins and amino acids. Some feed additives like enzymes or probiotic growth promoters are excluded from this treatment due to lack of stability. However, liquid forms of some of these substances are available for post-HTST application. The issue of vitamin stability is largely settled by the development of

formulation techniques withstanding higher temperatures, although for some vitamins (K<sub>3</sub>, A, E, D) the protection still is not 100% for expander/extruder processing (Coelho, 1996).

The issue of amino acid stability always has been given high attention by the feed industry due to the fact, that losses of amino acids show in a much more direct manner than losses of vitamins in livestock feeding and due to the common knowledge of the Maillard-reaction. The reaction of proteins with reducing sugars perhaps is the most common cause of nutritional damage to protein during food processing and storage (Hurrell and Finot, 1985). Though the Maillard reaction pertains mainly to food processing, excessive toasting of soybean meal may also cause damage of lysine leading to reduced growth performance (Araba and Dale, 1990).

Stability of supplemented crystalline amino acids has not been studied yet extensively. However, unpublished data from Amandus Kahl Co. show good stability of L-Lysine-HCl, DL-Methionine and L-Threonine.

The use of free base lysine in a liquid form presents technological advantages in feed manufacturing. It saves the investment of a micro-dosing unit, because the liquid can be added directly into the main mixer by simple volumetric proportioning and spraying devices. Thus the feed manufacturer is less dependent on pre-mixed amino acid preparations and can easily adjust the dietary lysine concentration according to nutritional and customer demands. However, the presence of the unprotected  $\alpha$ -amino group in the free base lysine may give reason for further concern of heat damage of this important essential amino acid.

Thus the stability of liquid free base lysine in HTST feed processing was tested with growing broilers.

## Material and methods

### Treatments, diets and feed processing

8 treatments with two types of diets as illustrated in Fig. 1, have been tested, i.e.,

#### Diet A

- A 0 - untreated control
- A 90 - expander treatment with 90°C head temperature
- A 110 - expander treatment with 110°C head temperature
- A 130 - expander treatment with 130°C head temperature
- A 110pp - expander treatment with 110°C head temperature, liquid lysine added after expansion

#### Diet B

- B 90 - expander treatment with 90°C head temperature
- B 110 - expander treatment with 110°C head temperature
- B 130 - expander treatment with 130°C head temperature

Diet composition of the wheat-soyabean meal based diet is shown in Table 1.

Although cereals and oilseeds themselves contain little reducing sugar, damaging reaction can occur in fabricated feeds when sugars or sugar containing ingredients, such as molasses are added. For the purpose of creating favourable conditions for Maillard-reaction, in diet B molasses was added mainly at the expense of wheat. Diets have been made isocaloric by addition of different levels of soya oil. Liquid lysine with 50% L-Lysine content was added into the main mixer in all treatments except A 110pp. To make lysine deficient diets, total dietary lysine content was set at 1.00% which is 90% of NRC (1994) or 77% of Austic (1994) recommendation. The added lysine made up 20% of the total dietary lysine in all test diets.

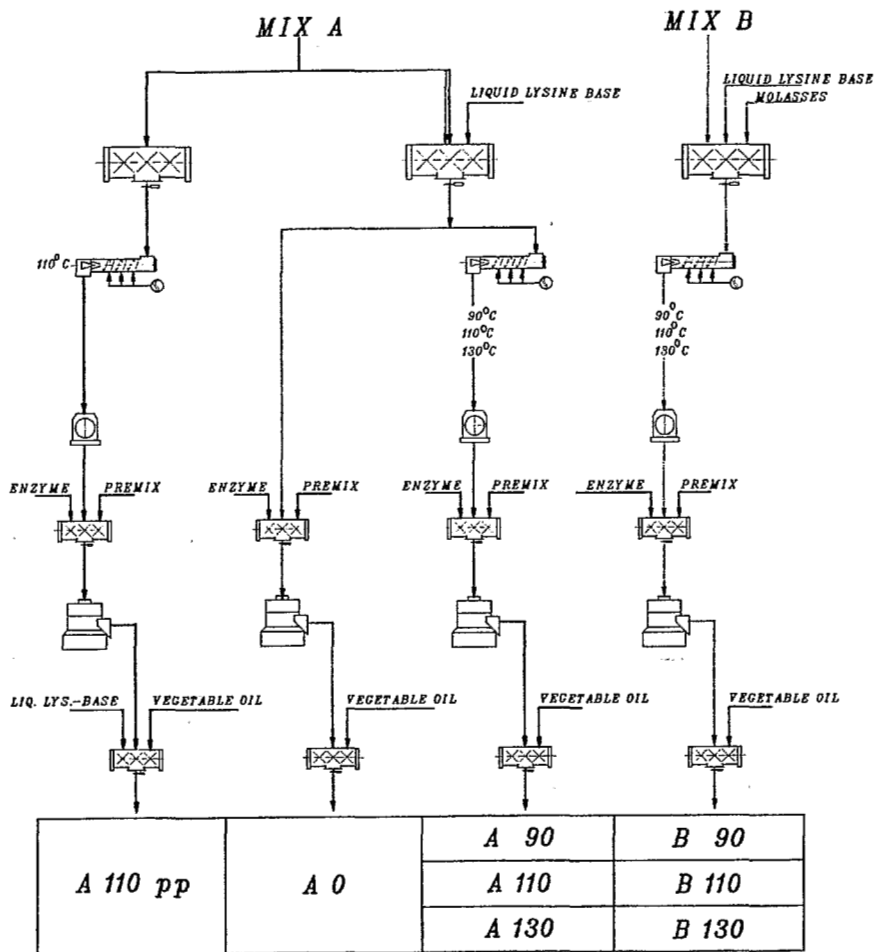


Fig. 1. Processing of broiler feed with liquid lysine treatments.

Table 1. Composition of diets A and B (%)

	Diet A	Diet B
Wheat	60.13	55.04
Cornglutenfeed	6.76	5.75
Soybeanmeal 44	21.76	22.65
Molasses	~	5.00
Soya oil	7.00	7.20
Limestone	1.07	1.07
Di-calc-phos	1.39	1.39
Salt	0.19	0.19
Vitamin-mineral-mix†	1.00	1.00
DL-Methionine	0.21	0.22
L-Threonine	0.09	0.09
Liquid Lysine 50%	0.40	0.40

†The vitamin-mineral-mix supplied per kg diet: 10,000 IU Vit. A; 2,000 IU Vit. D<sub>3</sub>; 15 mg Vit. E; 1.5 mg Vit. K<sub>3</sub>; 1 mg Vit. B<sub>1</sub>; 5 mg Vit. B<sub>2</sub>; 3 mg Vit. B<sub>6</sub>; 20 mcg Vit. B<sub>12</sub>; 30 mg Nicotinic acid; 10 mg Ca-Panthotenate; 0.5 mg Folic acid; 160 mg Cholin; 50 mg Zinc; 70 mg Manganese; 20 mg Iron; 10 mg Copper; 1 mg Iodine, 0.5 mg Cobalt; 0.15 mg Selenium; 50 mg Zincbacitracin; 105 mg Amprolium; 500 mg enzyme preparation

Calculated and analysed nutrient content of diets A and B is shown in Table 2. The analysed concentration of total and added lysine fairly matched the calculated amounts. Other essential amino acids were formulated close to current recommendations.

Diets A and B without enzyme and premix addition have been expanded at the aforementioned temperature levels. Enzyme preparation (Xylanase) and premix were added after grinding the cooled expandate. Then this mixture was cold pelleted and soya oil was sprayed onto the pellets according to the formula. In treatment A 110pp liquid lysine was sprayed onto the pellets before the soya oil.

Table 2. Calculated and analysed content of diet A and B (%)

	Diet A		Diet B	
	Calculated	Analysed	Calculated	Analysed
Dry matter	89.10	90.10	88.60	90.5
Crude protein	18.21	18.25	18.29	17.95
Digest. crude protein	16.05	15.90	15.50	15.25
Crude fat	8.70	9.90	9.00	10.01
Crude fiber	3.45	3.77	3.30	3.55
Ash	5.40	6.02	5.60	6.10
Ca	0.85		0.85	
P	0.63		0.63	
aP	0.40		0.40	
Na	0.12		0.12	
K	0.68		0.88	
Mg	0.16		0.17	
MEn-Poultry (kcal/kg)	3100	3055	3100	3089
Amino acids				
<i>Lysine total</i>	1.00	1.05	1.00	1.02
<i>Lysine added</i>	0.20	0.21	0.20	0.19
Methionine	0.47		0.48	
M + C	0.80		0.80	
Threonine	0.71		0.70	
Tryptophan	0.22		0.22	
Arginine	1.10		1.10	
Glycin + Serin	1.61		1.59	
Histidin	0.46		0.44	
Isoleucine	0.76		0.75	
Leucine	1.34		1.31	
Phenylalanine	0.85		0.83	
Phe + Tyrosine	1.43		1.41	
Valine	0.86		0.84	

### Broiler trial

240 male broiler chickens (ISA-4 line hybrids) have been randomly assigned to the 8 treatment groups (30 birds/treatment in 6 replicates with 5 birds/replicate). Broilers have been kept in the upper two floors of a three storey cage battery under 24 h light regime. Environmental temperature was set according to the breeders recommendation. Feed and water was supplied *ad libitum*. The growth trial went from 8-35 days (liveweight at start  $102 \pm 1.5$  g). Birds have been weighed and feed consumption has been recorded weekly. At the end of the trial birds were fasted for 20 h and 4 birds per treatment have been killed and autoclaved for 4 h at 2 bar. After homogenization dry matter, nitrogen and ash

has been analysed in each sample. N-retention has been calculated from N-deposition in the carcasses. 7 birds have been taken out of the experiment due to non-feed treatment related disease.

The apparent faecal digestibility of lysine and the apparent metabolisable energy of the diets (ME) was determined with 96 birds in metabolism cages (12 birds/treatment in 6 replicates with 2 birds each). The 2 birds per cage have been treated as one in calculating the results. After an adaptation period of 5 days the subsequent 5 days collection period started (days 19-24). In the excreta faeces and urine have been separated according to Ekman, modified according to Sandev (1979) and dry matter, nitrogen and lysine have been analysed. Dietary ME was corrected for zero nitrogen retention by taking into account 34.4 kJ/g N-deposition.

## Results and discussion

### Growth trial

The zootechnical parameters of the growth trial with diets A and B are presented in Table 3. Feed intake with diet A did not differ significantly between the treatments although processing with 110°C and 130°C showed numerical decrease.

Table 3. Broiler performance with dietary addition of liquid lysine and different feed processing temperatures (8-35 days, treatments A and B)

	A0	A90	A110	A130	A110pp	B90	B110	B130
Feed intake (g)	2598	2607	2244	2371	2421	2815	2535	2414
Liveweight gain (g)	1199 <sup>a</sup>	1196 <sup>a</sup>	1071 <sup>b</sup>	1102 <sup>b</sup>	1166 <sup>a</sup>	1293	1243	1210
FCR	2.17	2.18	2.10	2.15	2.08	2.17 <sup>a</sup>	2.04 <sup>b</sup>	1.99 <sup>b</sup>

a,b: Values within one line and different superscript letters differ significantly ( $P \leq 0.05$ )

Liveweight gain was not different between A0, A90 and A110pp whereas A110 and A130 exhibited significantly lower liveweight gain ( $P \leq 0.05$ ). Feed conversion rate was not affected significantly by the treatments.

With diet B increasing processing temperature was combined with a pronounced numerical decrease in feed intake which in turn affected liveweight gain. However, feed conversion efficiency was improved significantly when the treatment temperature increased.

When comparing the same processing intensity between diets A and B it is mentioned, that feed intake and liveweight gain was higher with the B-diets containing molasses. The metabolisable energy content of diet A and B was the same (3055 resp. 3089 kcal/kg). This was confirmed in the balance trial. Thus the reason for higher growth performance with diets B is the higher feed intake.

However, when the expander temperature was increased this had a detrimental effect on feed intake in diets A and B. Reasons for this can only be speculated on. Both diet were wheat based and the effect of expander treatment on increasing gut viscosity in wheat based diets in broilers is known (Liebert *et al.*, 1993). But the addition of the appropriate enzyme preparation can offset this effect. Foregut viscosity was not determined in this study thus the question of its influence on feed intake remains unanswered.

Concerning the stability of dietary lysine, both feed-born and added it can be concluded from the data of feed conversion efficiency, that it had remained unaffected at all levels of processing intensity. FCR was either the same (diet A) or even improved (diet B) with increasing temperature.

## Lysine digestibility and N-retention

The apparent faecal digestibility of dietary lysine and N-retention for treatments A and B is exhibited in Table 4.

Table 4. Apparent faecal digestibility (AFD) of lysine and N-retention in broiler carcass with dietary addition of liquid lysine and different feed processing temperatures (8-35 days, treatments A and B)

	A0	A90	A110	A130	A110pp	B90	B110	B130
AFD-Lysine (%)	91.5	87.2	88.9	89.2	89.9	88.9	87.6	88.9
N-intake (g)	77.4	75.2	65.8	69.5	71.8	80.2	72.9	73.5
N-deposition (g)	32.8	33.3	28.7	29.5	30.0	36.2	33.4	31.9
N-retention (%)	42.4	44.2	43.6	42.5	41.9	45.1	45.8	43.4
s	2.71	0.52	1.42	3.05	2.31	1.99	1.91	5.10

Considering the fact that the diets were wheat based, the apparent faecal digestibility of dietary lysine was rather high. CVB table (1996) indicates an apparent faecal digestibility for wheat of 84%. The high digestibility can be explained by the significant portion of free crystalline dietary lysine, which made up 20% of the total dietary lysine. No differences in lysine digestibility could be observed between diets A and B and different levels of processing intensity.

N-retention expressed as relative amount of deposited N in relation to N-intake did not differ either between diets and treatments. The highest temperature level (130°C) exhibited the highest standard deviation (s) in N-retention, however the differences were not statistically significant. This is in accordance with earlier studies in broilers (Liebert, 1995), where body protein deposition was not affected by the expansion process. Data of N-utilization do not hint any lysine damage by processing under the given conditions.

In the case of clear lysine limitation as applied in the presented experiment, any impediment in lysine availability will show in feed conversion efficiency and at least in carcass protein deposition. Both parameters have not been influenced in a negative way. Thus it can be concluded that dietary lysine was not affected in its bioavailability for broilers. Deducted from this it can be stated that the liquid lysine preparation with 50% free base lysine is stable as well under the given conditions.

## Implications

Free base liquid lysine as a source for fortification of lysine deficient diets may offer technological advantages in compound feed production. However, concern was expressed on the processing stability of this lysine preparation at high temperatures. The data from this experiment with broilers do not support such concern. Post pelleting application as practiced with some heat labile feed additives is possible but not compulsory with liquid lysine.

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