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# Effects of a synthetic growth hormone-releasing peptide on growth and carcass traits of pigs

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## SUMMARY

The current study was to determine the effects of growth hormone-releasing peptide ([His[1], Lys[6]]-GHRP) on growth performance and carcass traits in pigs, and to determine whether the results were comparable to those of recombinant porcine growth hormone (r-PGH) treatment. Thirty Landrace and Yorkshire pigs (40–45 kg) were randomly divided into three groups, each group consisted of two boars, five gilts and three barrows. Peptides were administered daily by intramuscular injection with the following schemes and dosages: Group A ([His[1], Lys[6]]-GHRP, 200 μg/day in 0·5 ml saline), Group B (r-PGH, 3 mg/day in 0·6 ml saline), and Group C (sterilized physiological saline, 0·5 ml/day). [His[1], Lys[6]]-GHRP caused significant improvements both in pig growth and carcass traits. The average daily gain (ADG) and feed efficiency of the [His[1], Lys[6]]-GHRP treated group were higher than those of the control, but less than with the r-PGH treatment. The net increase in the ADG by [His[1], Lys[6]]-GHRP was *c.* 14 %, and was 23 % by r-PGH (both $P < 0{\cdot}01$). The feed efficiency was improved 17 % by [His[1], Lys[6]]-GHRP ($P < 0{\cdot}01$) and 20 % by r-PGH ($P < 0{\cdot}01$). On the other hand, the back-fat thickness in both [His[1], Lys[6]]-GHRP and r-PGH treated groups ($1{\cdot}76 \pm 0{\cdot}041$ cm and $1{\cdot}72 \pm 0{\cdot}040$ cm, respectively) was considerably less ($P < 0{\cdot}05$) than that of the control group ($1{\cdot}81 \pm 0{\cdot}040$ cm). In addition, the lean cuts percentage and the rib longissimus muscle area were comparable in the [His[1], Lys[6]]-GHRP and r-PGH treatments (lean cuts percentage: $57 \pm 2{\cdot}0$ % and $57 \pm 1{\cdot}5$ %, respectively; rib longissimus muscle area: $32{\cdot}8 \pm 0{\cdot}91$ cm$^2$ and $32{\cdot}9 \pm 1{\cdot}10$ cm$^2$, respectively), and both were significantly ($P < 0{\cdot}05$) greater than those of the control group (lean cuts percentage: $53 \pm 1{\cdot}5$ %; rib longissimus muscle area: $28{\cdot}3 \pm 1{\cdot}61$ cm$^2$). In conclusion, [His[1], Lys[6]]-GHRP appeared to be a promising tool for controlling carcass fat in pigs.

## INTRODUCTION

The rate of liveweight gain and feed conversion efficiency have long been major determinants of efficiency in animal production systems. However, for reasons such as health and economic costs, carcass fatness has increasingly become a matter of concern. It is known that an increase in the concentration of circulating growth hormones can cause mammals to have faster growth rates and increased milk production (Baker *et al.* 1984; Croom *et al.* 1984); hence considerable attention has been directed toward the anabolic effects of growth hormone (GH) on growth performance of meat animals. For instance, previous reports have shown that the administration of exogenous porcine growth hormone (PGH) to pigs increases growth rate, improves feed efficiency and changes carcass traits markedly (Machlin 1972; Chung 1985; Rebhun *et al.* 1985; Etherton *et al.* 1987). These observations, with other recent advances in the availability of PGH synthesised through recombinant DNA technology, have promised the possibility of developing a PGH-based growth promoter for the hog raising industry.

In addition to a direct administration of exogenous growth hormone, various ways have been discovered which promote the release of growth hormone *in vivo*. For instance, the effects of growth hormone-releasing factor, known as [hpGRF-(1-44)-$NH_2$], which is a 44-amino acid peptide with carboxy terminus amino acid amidated (Guillemin *et al.* 1982), on growth hormone

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Table 1. *Effects of recombinant porcine growth hormone (r-PGH) and growth hormone-releasing peptide ([His¹, Lys⁶]-GHRP on pig growth performance*

| Trait | Treatment | | | | | |
|---|---|---|---|---|---|---|
| | [His¹, Lys⁶]-GHRP | | r-PGH | | Saline | |
| Number of pigs | 10 | | 10 | | 10 | |
| Mean initial weight (kg) | 42 ± 1·1 | (10)[f] | 45 ± 1·4 | (9) | 45 ± 1·4 | (10) |
| Mean finishing weight (kg)[a] | 97 ± 2·7 | (10) | 99 ± 1·8 | (9) | 97 ± 2·1 | (10) |
| ADG[b] | 647 ± 20 | (10) | 706 ± 20 | (9) | 557 ± 20 | (10) |
| ADFI[c] | 1·94 ± 0·021 | (10) | 2·04 ± 0·028 | (9) | 2·00 ± 0·022 | (10) |
| F/G[d] | 3·0 ± 0·11 | (10) | 2·9 ± 0·11 | (9) | 3·6 ± 0·11 | (10) |
| Backfat thickness (cm)[e] | 1·76 ± 0·041 | (4) | 1·72 ± 0·040 | (4) | 1·81 ± 0·042 | (4) |

[a] Average finishing weight was determined when the pigs reached *c*. 95–100 kg. The time spent was 11 weeks for the r-PGH treated group, 12 weeks for the [His¹, Lys⁶]-GHRP treated group, and 13 weeks for the control group.
[b] ADG: average daily gain (g/day).
[c] ADFI: average daily feed intake (kg/day).
[d] F/G: feed to gain (kg/kg).
[e] Back fat thickness was estimated using real-time ultrasound measurements.
[f] Numbers in parentheses indicate number of values contributing to the mean.

Table 2. *Effects of recombinant porcine growth hormone (r-PGH) and growth hormone-releasing peptide ([His¹, Lys⁶]-GHRP) on carcass traits*

| Trait | Treatment | | | | | |
|---|---|---|---|---|---|---|
| | [His¹, Lys⁶]-GHRP | | r-PGH | | Saline | |
| Number of pigs | 4 | | 4 | | 4 | |
| Live body weight (kg) | 101 ± 1·6 | (4)† | 98 ± 1·9 | (4) | 100 ± 2·6 | (4) |
| Carcass weight (kg) | 80 ± 1·5 | (4) | 77 ± 0·3 | (4) | 81 ± 2·5 | (4) |
| Dressing percentage (%) | 79·7 ± 0·81 | (4) | 79·1 ± 1·11 | (4) | 79·1 ± 0·62 | (4) |
| Carcass length (cm) | 89·3 ± 0·50 | (4) | 89·5 ± 0·90 | (4) | 86·8 ± 1·60 | (4) |
| Lean cuts percentage (%) | 57 ± 2·0 | (4) | 57 ± 1·5 | (4) | 53 ± 1·5 | (4) |
| TRLMA* | 32·8 ± 0·91 | (4) | 32·9 ± 1·10 | (4) | 28·3 ± 1·61 | (4) |
| Back fat thickness (cm) | 1·76 ± 0·041 | (4) | 1·72 ± 0·040 | (4) | 1·81 ± 0·040 | (4) |

* TRLMA = 10th rib longissimus muscle area ($cm^2$).
† Numbers in parentheses indicate number of values contributing to the mean.

(GH) release *in vivo* and consequently on growth performance in pigs have been established (Etherton *et al*. 1986).

On the other hand, His-DTrp-Ala-Trp-DPhe-$LysNH_2$ ([His¹, Lys⁶]-GHRP), a synthetic Met⁵-enkephalin analogue which was designed through computer modelling techniques (Momany *et al*. 1981), is a new synthetic hexapeptide which specifically elicits a dosage-related release of GH *in vitro* and *in vivo* and in a variety of animal species including pigs without a concomitant release of LH, FSH, TSH or PRL (Bowers *et al*. 1984; Ilson *et al*. 1989; Yellin *et al*. 1990). In addition, several reports have shown that the administration of this peptide results in an increased rate of weight gain in rats (Baker *et al*. 1984) and increased milk production in lactating dairy cattle (Croom *et al*. 1984).

The objective of the current study was to determine the effects of [His¹, Lys⁶]-GHRP on growth performance and carcass traits in pigs, and to determine whether the results were comparable to those of r-PGH treatment.

## MATERIALS AND METHODS

The peptide was synthesised by solid-phase Fmoc chemistry (Stewart & Young 1984) utilizing an Applied Biosystems (ABI) 430A peptide synthesizer (Applied Biosystems, Foster City, CA) and synthesis cycles supplied by ABI. The crude peptide was purified by high performance liquid chromatography (HPLC) on the Beckman HPLC system equipped with a 10·7 × 250 mm Inertsil-ODS-2 reversed-phase semi-preparative column (Gasukuro Kogyo Inc, Tokyo).

The purity of the peptide was checked by analytical HPLC, thin layer chromatography, and amino acid analysis (6 N HCl, 110 °C, 24 h). The purified peptide was also characterized by N-terminal sequence analysis on an ABI Model 473A protein sequencer, and by Fast Atom Bombardment Mass Spectrometry (FABMS).

In the experiment for study of the growth performance and carcass traits in pigs treated with [His$^1$, Lys$^6$]-GHRP or r-PGH, 30 Landrace and Yorkshire pigs (40–45 kg) were randomly divided into three groups, each group containing two boars, five gilts and three barrows. The pigs were individually housed and fed a corn–soyabean based diet formulated to contain 17·6% crude protein (CP), 0·5% lysine, and 3220 kcal/kg feed of digestible energy. [His$^1$, Lys$^6$]-GHRP or r-PGH (Pitman-Moore Inc, Mundelein, IL) was administered daily by intramuscular injection at 09.00 h for 11–14 weeks with the following dosages: Group A ([His$^1$, Lys$^6$]-GHRP, 200 μg/day in 05 ml saline); Group B (r-PGH, 3 mg/day in 0·6 ml saline); and Group C (sterilized physiological saline, 0·5 ml/day). To determine the growth traits, pigs were weighed at the beginning of the study, at 2-week intervals, and at the end of the study. Feed intake was also determined for the same periods. The data were used to determine average daily gain, average daily feed intake and feed utilization. When the body weights reached *c.* 95–100 kg, four animals from each group (two gilts and two barrows) were slaughtered and the carcass traits were measured. Carcass composition was evaluated on the basis of standard carcass measurements, including live pre-slaughter weight, hot carcass weight, carcass length, lean cuts percentage, longissimus muscle area at the 10th rib, and back-fat thickness at the 10th rib.

Data throughout were analysed by analysis of variance (Fisher 1949).

## RESULTS AND DISCUSSION

After the [His$^1$, Lys$^6$]-GHRP was synthesized and purified as described earlier, the purity of the peptide was found to be 96% by analytical HPLC. The molecular weight of the peptide as observed by FABMS, which was used to confirm that the synthetic peptide was of the correct composition and of high purity, had the expected $(M+H^+)$ peak at m/z 873·5 and was in good agreement with the calculated value (cal. 872·4, $\Delta = 1{\cdot}4$). The amino acid composition and sequence analyses were identical to those of actual samples.

In the study of the effects of [His$^1$, Lys$^6$]-GHRP and r-PGH treatment on growth and carcass traits in pigs, the pigs were divided into three groups: group A was treated with [His$^1$, Lys$^6$]-GHRP, Group B with r-PGH, and group C with sterilized saline. Throughout the growth period during the experiment, most of the pigs grew normally except for one in group B, which suffered from diarrhoea. The treatment of pigs with [His$^1$, Lys$^6$]-GHRP or r-PGH daily for 11–14 weeks positively affected growth performance (Table 1). The net increase in the average daily gain (ADG) by [His$^1$, Lys$^6$]-GHRP was *c.* 14% and 23% by r-PGH (both $P < 0{\cdot}01$). The feed efficiency was improved 17% by [His$^1$, Lys$^6$]-GHRP ($P < 0{\cdot}01$) and 20% by r-PGH ($P < 0{\cdot}01$). In both cases, the effects on the treated groups were higher than on the control. However, the effect of the [His$^1$, Lys$^6$]-GHRP treatment was less than with the r-PGH treated ones.

On the other hand, there was a trend for a positive effect of [His$^1$, Lys$^6$]-GHRP or r-PGH on carcass composition (Table 2). The back-fat thickness of carcass both in the [His$^1$, Lys$^6$]-GHRP and r-PGH treated groups was considerably less than that of the control ($P < 0{\cdot}05$). However, the effect of [His$^1$, Lys$^6$]-GHRP was intermediate between that of r-PGH and that of the control. Meanwhile, the lean cuts percentage and rib longissimus muscle area were comparable in [His$^1$, Lys$^6$]-GHRP and r-PGH treatments, and both were significantly greater than those of the control group ($P < 0{\cdot}05$).

The data presented in this report demonstrate that the administration of [His$^1$, Lys$^6$]-GHRP or r-PGH to pigs enhanced growth rate, improved feed efficiency and changed carcass traits. The results were consistent with those previously reported for GRF or recombinant GH-treated pigs (Baile *et al.* 1983; Etherton *et al.* 1986; Boyd 1987). Nevertheless, this is the first time that the effects of [His$^1$, Lys$^6$]-GHRP on growth and carcass traits of pigs have been demonstrated. However, to stimulate growth performance maximally, it is important to establish the optimal dose of [His$^1$, Lys$^6$]-GHRP, as well as the most appropriate pattern of delivery. These are currently being investigated.

Although [His$^1$, Lys$^6$]-GHRP and GRF stimulate GH release from the pituitary glands, there are differences between these two peptides in their action both *in vitro* and *in vivo*. There is evidence that [His$^1$, Lys$^6$]-GHRP and GRF act via different receptors and through a different mechanism to release GH (Badger *et al.* 1984). Thus, a synergistic effect on GH release in rats has been observed both *in vitro* and *in vivo* (Etherton *et al.* 1986; Cheng *et al.* 1989). We are currently also investigating whether the synergistic effect will be observed on the growth and carcass traits in pigs.

The people of developed countries have become more health conscious, particularly in the light of the decreased necessity for manual labour and hence decreased calorific requirements, together with the association of high intake of animal fat with human obesity, cardiovascular disease and premature death. Thus the development of products that will reduce

carcass fatness is consistent with current consumer preference and with medical advice favouring the consumption of leaner meat. The data shown in this report suggest that [His[1], Lys[6]]-GHRP will be a potential candidate for fulfilling this role in controlling carcass fatness in pigs.

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