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Effects of intramuscular injections of folic acid on serum folates, haematological status and growth performance of growing-finishing pigs

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Summary — In a first trial, 48 piglets aged 5 weeks, grouped into 6 blocks of 8 pigs each, were used to determine the effect of intramuscular injections of folic acid on serum folates, haematological status, growth performance and carcass characteristics. Each block consisted of 2 pens of 4 pigs; in one pen, pigs received, by weekly intramuscular injections, an increasing volume of a solution containing 5 mg/ml folic acid, while in the other the animals were non-injected controls. The concentration of serum folates in treated pigs was 19% higher ($P = 0.005$) than in controls. There was no effect of treatment ($P \geq 0.29$) on haemoglobin and haematocrit. During the starting period, (5-11 weeks) average daily gain was not influenced by folic acid injections but feed intake and feed conversion were decreased ($P = 0.07$ and $P = 0.05$ respectively). No effect of folic acid ($P \geq 0.23$) was noted from 11–23 weeks of age, suggesting that the supplement was suboptimal during the growing-finishing period. In a second trial, 72 piglets aged 9 weeks were assigned to 6 blocks of 12 animals each. The following treatments were randomly distributed in each block according to a 2 x 3 factorial design: level of feeding (restricted *vs ad libitum*) and weekly intramuscular injections of increasing volume of solutions containing either 0, 15 or 30 mg/ml of folic acid. The variables studied were the same as in Trial 1. Concentrations of serum folates varied quadratically ($P = 0.0001$) with the dose injected, a plateau being attained with injections of 15 mg/ml. There was no effect of treatment ($P = 0.043$) on haemoglobin and haematocrit. During the growing period (9-15 weeks), no effect ($P \geq 0.72$) of folic acid was noted on growth performance. However, during the finishing period (15–21 weeks), folic acid given at a dose of 30 mg/ml decreased ($P = 0.006$) feed intake while no effect ($P \geq 0.13$) of the vitamin supplementation was noted on average daily gain and feed conversion. In both trials, there was no effect ($P \geq 0.21$) of any treatments on carcass characteristics. These results indicate that a supplement of folic acid administered by intramuscular injections was effective in increasing concentration of serum folates of starting or growing-finishing pigs. This supplement may be associated with a decrease in feed intake.

folate / haematological status / growth performance / fattening pig

Résumé — Effet de différents apports d'acide folique sur l'évolution des folates sériques et les performances zootechniques des porcs d'abattage. *Dans un premier essai, 48 porcelets âgés de 8 semaines ont été répartis en 6 blocs de 8 animaux chacun afin de déterminer l'effet d'injections intramusculaires d'acide folique sur les folates sériques, le profil hématologique, les performances de croissance et les caractéristiques charcutières. Chaque bloc était constitué de deux en-*

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clos de 4 porcs chacun; dans l'un de ces enclos, les porcs ont reçu, par voie intramusculaire, un volume croissant d'une solution, contenant 5 mg/ml d'acide folique alors que dans l'autre les animaux, servant de groupe témoin, n'ont pas été injectés. Les concentrations de folates sériques ont été de 19% plus élevées (P = 0,005) ches les porcs traités comparativement aux porcs témoins. Aucun effet du traitement (P ≥ 0,29) n'a été observé sur l'hémoglobine ou l'hématocrite. Le gain de poids moyen quotidien n'a pas été influencé par les injections d'acide folique pendant la période de début (5–11 semaines) mais la prise alimentaire (P = 0,07) et le taux de conversion (P = 0,05) ont été diminués. Aucun effet de l'acide folique (P ≥ 0,23) n'ayant été observé entre l'âge de 11 et 23 semaines, il est possible que le supplément administré pendant la période de croissance et de finition ait été suboptimal. Lors d'un second essai, 72 porcelets âgés de 9 semaines ont été répartis en 6 blocs de 12 animaux chacun. A l'intérieur de chaque bloc, les traitements factoriels (2 x 3) suivants ont été distribués: niveau d'alimentation (restreint vs libéral) et injections intramusculaires d'un volume croissant d'une solution contenant 0, 15 ou 30 mg/ml d'acide folique. Les variables étudiées ont été les mêmes que dans l'essai précédent. Les concentrations de folates sériques ont varié de façon quadratique (P = 0,0001) avec la dose injectée, un plateau étant atteint à la dose de 15 mg/ml. Aucun effet du traitement (P ≥ 0,43) n'a été observé sur l'hémoglobine ou l'hématocrite. Pendant la période de croissance (9–15 semaines) les performances zootechniques n'ont pas été influencées par le supplément d'acide folique (P ≥ 0,72). Par contre, suite aux injections de 30 mg/ml d'acide folique en période de finition (15–21 semaines), la prise alimentaire a diminué (P = 0,006); aucun effet de l'acide folique (P ≥ 0,13) n'a été noté sur le gain de poids moyen quotidien ou le taux de conversion. Dans les deux essais, il n'y a pas eu d'effet (P > 0,21) de traitements sur les caractéristiques charcutières. Un supplément d'acide folique administré par injections intramusculaires augmente donc les concentrations de folates sériques et semble entraîner une réduction de la prise alimentaire.

folate / statut hématologique / performance de croissance / porc d'abattage

INTRODUCTION

Folic acid has an essential nutritional role in mammals through its involvement in synthesis of DNA, RNA (Herbert and Das, 1976) and proteins (Chang and Kaiser, 1972). Tissues with a high rate of cell turnover such as red blood cells are primarily dependent upon an adequate supply of folic acid (Davis and Nichols, 1988). The fast growth rate of pigs during the first 6 months after birth requires intense cellular hyperplasia and hypertrophy of tissues as well as accretion of protein (Whittemore *et al*, 1988). In growing-finishing pigs, the folates provided by ingredients of the diet, bacterial synthesis and coproghagy have usually been considered sufficient to meet the animal's requirements (ARC, 1981; INRA, 1984; NRC, 1988), despite the lack of data confirming it. Nevertheless, folates from the diet are likely to be the main reliable source of this vitamin since coprophagy, as recently suggested by Bilodeau *et al* (1989) and de Passillé *et al* (1989), only seems to play a minor role in the B-complex vitamin status of fattening pigs.

Easter *et al* (1983) reported no change in gain or feed utilization after dietary addition of 0.5 or 1.5 ppm of folic acid during the starting, growing and finishing periods. However, no data on the efficiency of the dietary folate supply by feed analysis and/or measurement of folate status of the animals were included in that study. Such information is critical considering the lability of folic acid to heat and light (Ek and Magnus, 1980; Ford *et al,* 1983). The stability of the vitamin in premixes or feeds is not well documented (Wornick, 1968) but it is known that pelleting of diets reduces the high levels of folic acid naturally present in lucerne meal (Frape, 1985). Therefore, it seems that supplying additional folic acid *via* intramuscular injections would be a more reliable research tool in order to overcome the potential problems associated with stability of the vitamin. A first experiment was undertaken to measure the changes in folate status as well as production and haematological variables of pigs following intramuscular injections of folic acid during the starting and growing-finishing periods. The results of this first trial suggested that the supplementary folic acid used was suboptimal during the growing-finishing periods. Therefore, a second experiment using higher doses of folic acid than in Trial 1 and measuring similar variables was carried out in order to study particularly the growing and finishing periods.

MATERIALS AND METHODS

Trial 1

Forty-eight 5-week-old piglets (Yorkshire x Landrace) weighing approximately 7.8 kg (CV = 10.3%) were assigned by weight to 6 blocks of 8 pigs each. Piglets were weaned at 4 weeks of age and, after one week of adaptation to the experimental diet, were moved into experimental pens. Each block consisted of 2 pens of 4 pigs (2 castrates and 2 females) which were raised throughout the experimental period on a slatted floor. In one of the 2 pens, pigs received weekly intramuscular injections of a solution containing 5 mg/ml of folic acid [1]. The volume of injection was 1 ml until 20 kg bodyweight, 2 ml until 45 kg bodyweight and 3 ml until 90 kg bodyweight. This increase in injected volumes was designed to simulate the progression of folic acid intake when supplied by the diet. In the other pen, pigs were non-injected controls. It was impossible under the conditions of the present experiment to inject these animals with a placebo. However, this non-injected group was judged as a valid control, since treated pigs were injected only once a week; the stress associated with blood sampling or injections is known to have very short-term effects (maximum of 4 h) on haematological and biochemical variables (Dubreuil *et al*, 1989). Haematological and serum folate measurements as well as growth performances and carcass characteristics from this control group have been previously presented in a study on the effect of floor type on the evolution of some B-vitamins during growth of pigs (Bilodeau *et al*, 1989).

Feed intake in each pen and individual weight of pigs were measured every fortnight. The animals were fed, *ad libitum*, diets computed to contain respectively for the starting and growing-finishing period, 3.33 and 3.22 Mcal DE, 19.4 and 16.9% crude protein, 0.96 and 0.83% lysine, 1.15 and 0.98% Ca and 0.77 and 0.69% total P. There was no addition of folic acid in any of the 2 diets; the calculated and measured levels of folates in the starting diet were 0.62 and 0.47 mg/kg, respectively while the corresponding values were 0.65 and 0.46 mg/kg in the growing-finishing diet. Detailed composition is given by Bilodeau *et al* (1989). Blood samples taken by jugular venipuncture were collected every two weeks immediately before the injection of folic acid. After 18 weeks of experimentation, pigs were slaughtered and the carcasses graded according to the Canadian hog grading system (Anonymous, 1979).

Dietary folates were analysed in duplicate on 3 hydrolysates of the same sample with commercial radioassay kits using [^{125}I]PGA as described by Tremblay *et al* (1986). Preparation of samples before the assay was carried out according to a method adapted from Cerna and Kas (1983). In a conical 50 ml tube, 0.1 g of feed was mixed with 12 ml of McIlvain buffer (for 100 ml: 0.2 M Na_2HPO_4, 50 mg ascorbic acid, distilled water was added, pH adjusted to 4.6 with NaOH 3.3 M and completed with distilled water up to 100 ml) and transferred to an autoclave for 10 min at 121 °C. The pH was adjusted to 7.0 with NaOH 3.3 M and the volume brought to 20 ml with distilled water. The solution was vortexed and then centrifuged at 3 000 *g* for 10 min. The supernatant was used for folate determination. The effect of chicken pancreas conjugase (transformation of poly- to monoglutamates) on concentrations of folates was tested using the method described by Cerna and Kas

[1] Folvite ®, parenteral solution no 4154, Cyanamid Canada Inc, Montréal, Quebec, Canada

(1983). No effect of conjugase was noted on concentrations of dietary folates, and subsequently all assays were run without pretreatment with conjugase. The results seem to confirm previous observations (Rothenberg *et al*, 1974; Schreiber and Waxman, 1974) on the versatility of the radioassay technique for mono- and polyglutamates. Results of parallelism tests were satisfactory (CV = 10%) between 0 and 5 mg/kg and inter-assay CV was 9.1%. Recovery tests from a simulated mixing in the laboratory gave a mean of 94.2%.

Data were analysed as a complete random block design using the General Linear Models procedure of SAS (1985). The following model was used: $Y_{ijk} = \mu + B_i + F_j + BF_{ij} + T_k + BT_{ik} + FT_{jk} + BFT_{ijk} + e_{ijk}$, where Y_{ijk} indicates the following dependent variables: serum folates, haemoglobin or haematocrit. The overall mean is μ; B_i is the effect of block; F_j is the effect of folic acid administration and T_k is the effect of age. Least square means were compared using orthogonal contrasts when appropriate. Analysis of repeated measurements was made according to the procedure of Gill and Hafs (1971). Average daily gain, feed intake and feed conversion were analysed for each feeding period: weeks 5–11 for the starting period, and weeks 11–23 for the growing-finishing one. Data were submitted to a model similar to that described previously but the independent variable T_k was omitted. Carcass characteristics were also analysed using this last model.

Trial 2

Seventy-two 8-week-old piglets (Yorkshire x Landrace) weighing approximately 19.8 kg (CV = 12.3%) were assigned by weight to 6 blocks of 12 pigs each. After 1 week of adaptation to the experimental diet, animals were grouped 2 per pen (1 castrate and 1 female) and raised on slatted floors throughout the experimental period. The following treatments were randomly distributed within a block according to a 2 x 3 factorial design: level of feeding (restricted *vs ad libitum*) and weekly intramuscular injections of folic acid (0,15 or 30 mg/ml). Pigs in the restricted feeding group were fed twice daily and received between 90–95% of the intake recorded in the *ad libitum* group where animals were fed as described in Trial 1. The composition of each diet is presented in table I. In each feeding group, pigs were injected with 1 ml of one of the solutions of folic acid up to 45 kg of bodyweight, 2 ml up to 65 kg and 3 ml up to 85 kg. In the control (0 mg/ml) pen, pigs were injected with a corresponding volume of a saline solution (NaCl, 0.9%). At the end of the experiment, pigs were slaughtered and carcass graded according to the new Canadian hog grading system (Anonymous, 1986). Dressing percentage was also measured.

Feed intake per pen was recorded twice a week and individual weights were taken every fortnight. Blood sampling procedure, haematological and serum folate measurements were performed as in the first trial.

Data were analysed as a complete random block design using the General Linear Models procedure of SAS (1985). The following model was used: $Y_{ijkl} = \mu + B_i + A_j + F_k + AF_{jk} + BA_{ij} + BF_{ik} + BAF_{ijk} + T_l + BT_{il} + AT_{jl} + FT_{kl} + BAT_{ijl} + BFT_{ikl} + BAFT_{ijkl} + e_{ijkl}$, where Y_{ijkl} indicates the following dependent variables: serum folates, haemoglobin or haematocrit. The overall mean is μ; B_i is the effect of block; A_j is the effect of type of feeding; F_k is the effect of folic acid administration and T_l is the effect of age. Least square means were compared using orthogonal contrasts when appropriate. Analysis of repeated measurements were made according to the method of Gill and Hafs (1971). The concentration of serum folates before the initiation of treatments (9 weeks) was used as a covariate in the analysis of the dependent variable serum folates. Average daily gain, feed intake and feed conversion were analysed for each feeding period: weeks 9–15 for growing period and weeks 15–21 for the finishing one. Data were submitted to a model similar to that described above, the independent variable T_l being omitted. Carcass characteristics were also analysed using this last model.

RESULTS

Trial 1

The concentration of serum folates (fig 1) in pigs injected with folic acid was approximately 19% higher (P = 0.005) than in those injected with saline. The effect of

Table I. Composition of the experimental diets (Trial 2). * Provided per kg of diet a minimum of: Mn, 30 mg; Zn, 100 mg; Fe, 100 mg; Cu, 25 mg; I, 300 µg; vitamin A, 80 000 IU; vitamin D_3, 1 500 IU; vitamin E, 30 IU; menadione, 2.2 mg; thiamine, 1 mg; riboflavin, 4 mg; niacin, 20 mg; pantothenic acid, 13 mg; folic acid, 500 µg; pyridoxine, 1.5 mg; biotin, 200 µg; vitamin B_{12}, 15 µg; choline, 300 mg.

Ingredients	*Diets*			
	Growing		*Finishing*	
	Ad libitum	*Restricted*	Ad libitum	*Restricted*
Corn	595	659	400	580
Barley	120	–	215	185
Oat	–	–	207	–
Soybean meal (47.5%)	230	265	140	175
Animal fat	10	28	–	20
Limestone	15.5	16.5	12.5	13.5
Dical/phosphate	20	21	17	18
Salt	5	6	5	5
Mineral premix *	1.5	1.5	1.0	1.0
Vitamin premix *	3.0	3.0	2.5	2.5
Composition				
Energy (Mcal)				
calculated (DE)	3.29	3.42	3.08	3.32
analysis (GE)	3.87	3.97	3.90	3.82
Protein (%)				
calculated	17.8	18.7	15.2	15.7
analysis	18.3	18.7	16.2	17.2
Lysine (g/kg)				
calculated	9.2	10.0	7.1	7.7
Ca %				
calculated	1.11	1.17	0.92	0.98
P (%)				
calculated	0.72	0.74	0.65	0.67
Folates (mg/kg)				
calculated	1.11	1.12	1.03	1.04
analysis	1.0	1.07	0.64	0.72

age on concentration of serum folates was variable (age quintic, $P = 0.0001$). There was a quintic increase with age ($P = 0.007$) of haemoglobin and haematocrit but no effect ($P \geq 0.29$) of treatments was noted. Mean haemoglobin and haematocrit values were 11.5 g/dl (CV = 7.3%) and 39.1% (CV = 6.6%), respectively.

During the starting period (5–11 weeks), injections of folic acid improved ($P = 0.05$) feed conversion (table II). There was no effect of injection ($P = 0.56$) on average daily gain, but feed intake tended ($P = 0.07$) to be lower in treated animals. During growing-finishing period (11–23 weeks), there was no effect ($P > 0.23$) of treatment on

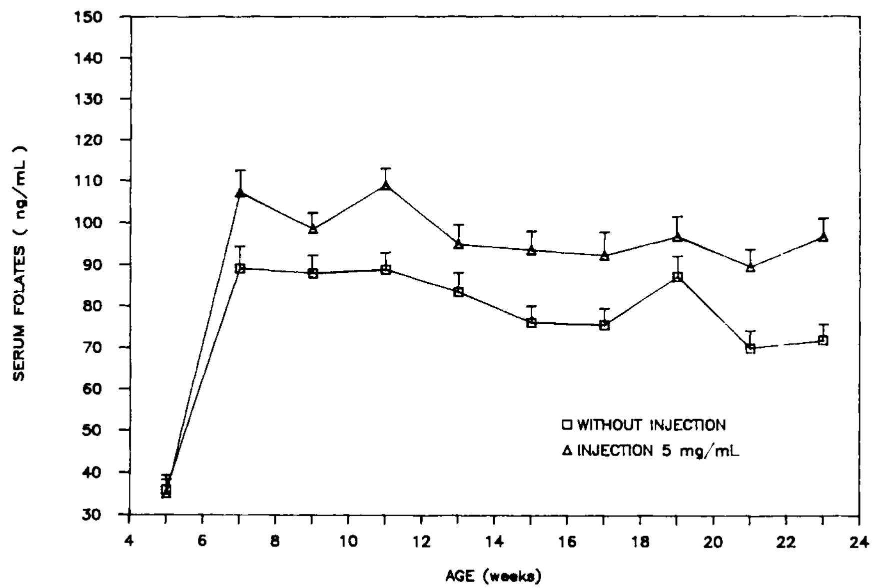

Fig 1. Concentrations (means ± SE) of serum folates during starting, growing and finishing periods of pigs injected or not with folic acid (Trial 1).

average daily gain, feed intake or feed conversion. Weight of warm carcasses, backfat thickness (sum of maximum shoulder and loin backfat) and carcass index were similar ($P \geq 0.34$) between treatments, the mean values being respectively 77.38 kg (CV = 8.3%), 54.9 mm (CV = 31.9%) and 105.0 (CV = 5.0%).

Trial 2

The concentration of serum folates (fig 2) increased in pigs injected with folic acid (quadratic effect of dose; $P = 0.0001$) as compared to those injected with saline, a plateau being attained with injections of 15 and 30 mg/ml. The effect of age on con-

Table II. Average daily gain (ADG), feed intake (I) and feed conversion (I/ADG) of pigs injected or not with folic acid (Trial 1).

Treatment	*Age (weeks)*					
	5–11 (Starting)			*11–23 (Growing-finishing)*		
	ADG	*I*	*I/ADG*	*ADG*	*I*	*I/ADG*
Without injection	0.47	0.96	2.07	0.78	2.45	3.15
Injection (5 mg/ml)	0.46	0.91	1.99	0.78	2.39	3.05
Probabilities	0.56	0.07	0.05	0.83	0.37	0.23
CV	5.51	4.38	2.43	5.40	3.88	4.02

centration of serum folates was significantly different between types of feeding (feeding x age cubic; $P = 0.0002$); the overall difference between *ad libitum* and restricted feeding, however, was small with values of 105.0 ng/ml (CV = 34.6%) *vs* 99.7 ng/ml (CV = 32.9%), respectively. There was a linear increase with age ($P = 0.0001$) of haemoglobin and haematocrit but no effect of treatments ($P \geq 0.43$). Mean haemoglobin and haematocrit values were 12.9 g/dl (CV = 5.4%) and 39.5% (CV = 5.0%), respectively.

Feed conversion was lower in restricted than in *ad libitum* feeding during both the growing ($P = 0.05$) and finishing ($P = 0.0001$) periods (table III). During the growing period (9–15 weeks), no effect of folic acid ($P \geq 0.72$) was noted on growth performances. However, the injection of a solution of 30 mg/ml folic acid decreased (injection linear; $P = 0.006$) feed intake during the finishing period (15–21 weeks); a 5% decrease was observed in the *ad libitum* group between 0 and 30 mg/ml (feeding injection linear; $P = 0.03$). Some changes were also noted in weight gain and feed conversion but the differences between levels of injected folic acid were not significant ($P = 0.13$ and $P = 0.20$, respectively). Weight of warm carcasses, lean and dressing percentage as well as index were similar ($P \geq 0.21$) between treatments, the mean values being respectively 71.0 kg (CV = 6.3%), 50.4% (CV = 2.0%), 81.7% (CV = 2.3%) and 102.0 (CV = 5.71%).

DISCUSSION

Serum folates

In humans, the concentration of serum folates is not considered as a reliable indicator of folate status because of its depen-

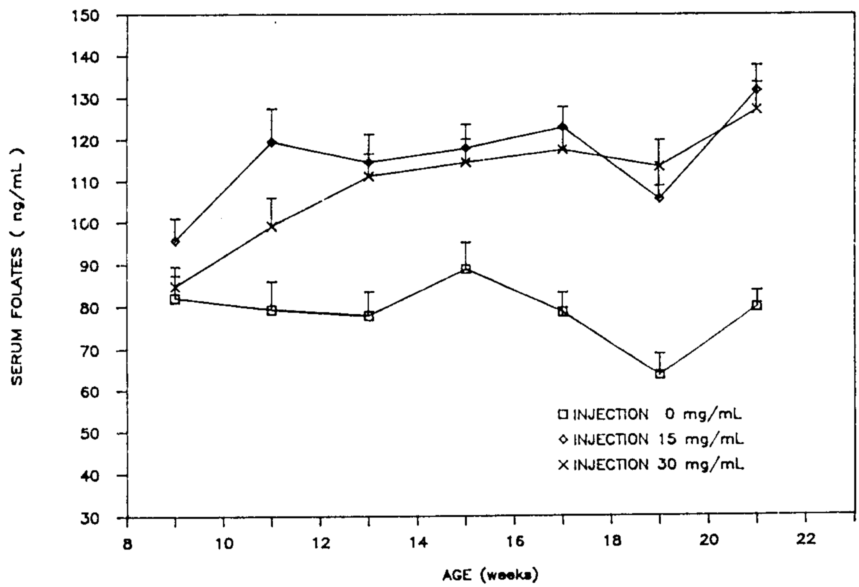

Fig 2. Concentrations (means ± SE) of serum folates during the growing and finishing periods of pigs treated or not with folic acid (Trial 2).

Table III. Average daily gain (ADG), feed intake (I) and feed conversion (I/ADG) of pigs according to level of feeding and folic acid treatments (Trial 2). [1] Injection linear, $P = 0.006$; [2] Feeding * Injection linear, $P = 0.03$.

Feeding regimen	*Injection of folic acid*	*Age (weeks)* *9–15 (Growing)*			*15–21 (Finishing)*		
		ADG	*I*	*I/ADG*	*ADG*	*I*	*I/ADG*
Ad libitum							
	saline	0.73	1.58	2.17	0.76	2.45	3.24
	15 mg/ml	0.73	1.60	2.19	0.71	2.48	3.49
	30 mg/ml	0.74	1.59	2.14	0.71	2.33	3.29
Restricted							
	saline	0.69	1.42	2.05	0.75	2.26	3.03
	15 mg/ml	0.69	1.40	2.02	0.73	2.21	2.97
	30 mg/ml	0.70	1.40	2.01	0.75	2.22	2.97
Probabilities							
Feeding		0.09	0.0001	0.05	0.23	0.0001	0.0001
Injection		0.72	0.99	0.74	0.13	0.01[1]	0.20
Feeding * Injection		0.97	0.76	0.86	0.39	0.04[2]	0.49
CV		4.8	3.9	5.3	5.2	2.8	5.9

dency upon short-term dietary changes (Rothenberg *et al,* 1974; Sauberlich, 1975). However, as noted previously by Matte *et al* (1984) with gestating sows, circulating serum folates in swine are likely to properly reflect the status in folates because of the uniformity of the feeding regimen within a herd. Moreover, in order to avoid any immediate interference due to feeding, blood samples were taken, in the present experiment, at the same time of the day (0830 to 1000) after a fasting period of at least 12 h.

Serum folates increased by over 100% in pigs from 5–7 weeks of age. This increase is most likely due to the difference in amount and type of feed consumed during this period (dry feed) compared to before weaning (milk). Indeed, it is known that the concentration of folates in sow's milk (Matte and Girard, 1989) represents, on a dry matter basis, about 10 times less folates than in cereal-based diets. In both trials, the concentration of serum folates in control animals varied between 70–90 ng/ml, from 9 weeks of age to the end of the finishing period. These fluctuations as well as the interaction feeding x age cubic (Trial 2) might be associated with variations in availability of folic acid between the different diets; the type of ingredients varied between feeding treatments and between periods. Considering the control groups in both trials, it appears that the higher levels of folates provided by diets in Trial 2 as compared to Trial 1 has a limited impact on the evolution of the concentration of folates as compared to weekly intramuscular injections during the growing-finishing period. Animals injected with folic acid had approximately 19% more circulating serum folates than control animals in Trial 1 and over 40% in Trial 2. It therefore seems that the injection of a solution of 5 mg/ml folic acid (Trial 1) was not sufficient to maximize the concentration of folates in serum. However, the quadratic effect of injection (P = 0.0001) in Trial 2 indicated that a plateau in concentration of serum folates was attained after the injection of a solution of 15 mg/ml folic acid. Such a plateau might be explained by the presence of serum folate-binding proteins. In humans, serum folates are either free or protein-bound (Markkanen *et al,* 1972; Wagner, 1985; Zamorano *et al,* 1985). In pigs, Mantzos *et al* (1974) noted the presence of a great quantity of unsaturated "avid specific binders" of folic acid in blood plasma of animals between 2 days and 3 years of age. The capacity of these "specific binders" to link folates in serum is not known, but the plateau in concentrations of serum folates suggests that injections of 15 and 30 mg/ml saturated the folate-binding capacity.

Haematological measurements

The increase in the serum concentration of folates induced by intramuscular injections of folic acid in both trials had no effect on the evolution of blood haematocrit and haemoglobin. It is known that prolonged folate deficiency may result in haematological disorders of pigs and poor performance (NRC, 1988). It appears that, in both trials, the lower concentrations of folates in control as compared to treated pigs were not indicative of any deficiency associated with haematocrit and haemoglobin.

Growth performance

In both trials, for some periods, feed intake decreased or tended to be reduced after folic acid treatments. The effect of a supplement of folic acid on feed conversion during the starting period in Trial 1 was mediated by an alteration in feed intake. This effect did not persist during the growing-finishing period in spite of similar differences, in absolute values, between treatments (table II). These results may be due to the fact that the total amount of folic acid given through both the diet and intramuscular injections was, on an ADG basis, higher, 3.0 mg/kg ADG, during the starting period as compared to a value of 2.6 mg/kg ADG during the growing-finishing period. It was concluded, therefore, that the supplement of folic acid might be suboptimal during the growing-finishing period. In Trial 2, during the growing period, the ratio was elevated to values of 4.7 and 7.1 mg/kg ADG in pigs injected with 15 and 30 mg/ml, respectively while the corresponding values were 6.2 and 10.0 during the finishing period. It seems that, in order to produce an effect on feed intake, a much higher quantity of folic acid is needed during the finishing period (Trial 2) than during the starting period (Trial 1).

The present results on appetite are similar to previous observations where feed intake tends to be reduced after addition of folic acid to a starting (Lindemann and Kornegay, 1986) and a growing-finishing diet (Easter *et al,* 1983). One could explain this effect by metabolism of folic acid in brain areas responsible for the control of feed intake. It is known that folates act as cofactors in the synthesis and catabolism of serotonin, norepinephrine and epinephrine (Laborit, 1970). Folic acid can also competitively inhibit the brain L-glutamate decarboxylase, an enzyme essential for the synthesis of gamma-aminobutyric acid (GABA) (Tunnicliff and Ngo, 1977), which is a neurotransmitter involved in the control of feed intake in rat (Grandison and Guidotti, 1977; Kelly and Grossman, 1979; Kelly *et al,* 1979) and sheep (Girard *et al,* 1985). Moreover, a recent study demonstrated that folic acid reverses the action of GABA by acting on the GABA a II-receptor in the brain (van Rijn *et al,* 1988).

The entry of folates in cerebrospinal fluid, and probably in the brain, is regulated by a saturable transport system. However, at high concentrations of folates, the transport system is saturated and folates enter the cerebrospinal fluid by diffusion (Spector and Lorenzo, 1975; Ordonez, 1977). Variations in the capacity of saturation of this transport system might explain the inconsistency of folic acid effect on feed intake between Trials 1 and 2. Moreover, assuming that folic acid affects feed intake *via* brain neurotransmitters, the responses observed in the present experiments would be in agreement with those reported by Hunter *et al* (1971) and Ch'ien *et al* (1975) showing wide individual variations in the physiological responses of brain to a supplement of folic acid. Therefore, it is possible that the tendency of feed intake to be reduced following intramuscular injections of folic acid administered during the starting, growing or finishing period was related to an effect of folic acid on the brain neurotransmitters involved in the control of feed intake.

In conclusion, the present results indicate that supplement of folic acid administered by intramuscular injections increased the serum concentrations of folates of starting, growing and finishing pigs but did not affect haematological status. The possible effect of folic acid on feed intake merits further evaluation.

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