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SHORT COMMUNICATION

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Water kefir for weaned piglets: A pilot study on its farm-scale production, its palatability and its effects on growth performance

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Abstract

BACKGROUND: Water kefir is a beverage with potential benefits for weaned piglets, including increasing feed intake and being a source of probiotics and organic acids. However, experimental confirmation of this potential is lacking. The aim of this pilot experiment was to develop a method for producing and delivering water kefir to weaned piglets at farm scale, assess its palatability and identify potential side effects in piglets varying in weight.

RESULTS: The method described here is applicable to farm practice, as it could produce >50 L water kefir per day with little investment and time. The lactic acid concentration and pH in the water kefir remained stable between batches, while the acetic acid concentration was more variable. Piglets consumed the total quantity offered, which represented about 1 L per piglet per day. No clinical signs of disease were observed requiring medication administration to the piglets. However, a trend of reduced growth was observed when water kefir was offered, especially in the lighter piglets.

CONCLUSIONS: In addition to its applicability on farms, the water kefir production method facilitates experiments in young livestock under farm-scale conditions. The health-promoting potential of water kefir may be increased by elevating the nutrient supply in the culture medium. No critical issue was identified that would prevent the distribution of water kefir to weaned piglets. However, the reason for the trend toward reduced growth and whether this trend would be reversed in situations with greater disease pressure remain unclear.

KEYWORDS
acetic acid, lactic acid, post-weaning health, probiotic bacteria, yeast

INTRODUCTION

Weaning is one of the most critical periods for pig health, and gastrointestinal infections are especially prevalent. The weaning period is often associated with the greatest use of antibiotics,[1] and about 80% of these antibiotics are used to combat diarrhoea.[2] There is an urgent need to reduce antibiotic use in animal production, and thus the weaning period is particularly important. Different strategies related to feeding have been proposed to tackle gut health problems in piglets in the first days post-weaning. First, improving feed intake promotes the resilience of piglets against gastrointestinal problems.[3] In this context, Da Silva et al.[4] indirectly increased the feed intake of weaned piglets by up to +70% and weight gain by up to +87% by promoting water intake by up to +84% by flavouring and sweetening the drinking water. Second, as stomach pH is empirically known to increase after weaning, supplying organic acids to piglets may reduce the incidence and severity of post-weaning diarrhoea.[5,6] This effect has been attributed to the acidifying properties of organic acids supporting enzymatic activity in the stomach and the growth of endogenous gut bacteria over pathogenic bacteria.[5,6] Third, the use of probiotics to prevent gut infections via competitive exclusion of pathogens and through the immunomodulatory effects of the probiotics is of interest. Positive effects on intestinal health in weaned piglets have been reported from the live yeast *Saccharomyces cerevisiae* (villus length, development of the gut immune system)[7] and live lactic acid bacteria (abundance of endogenous bacteria over pathogenic bacteria).[8,9] Furthermore, the combination of live yeast and lactic acid bacteria has been shown to improve the growth rate and feed conversion ratio in weaned pigs.[10] Implementing these strategies in combination requires either accumulating feed supplements into complete piglet feed or finding a substitute product that would comprise all three strategies.

Water kefir is a fermented beverage, slightly sparkling and with a sweet–sour taste. It differs significantly from the better-known milk kefir.[11] Water kefir grains are composed of a symbiotic population of a variety of lactic acid and acetic acid bacteria and yeasts, mostly *S. cerevisiae*.[12–14] In the context of the problems occurring at weaning, water kefir could be a patent-free alternative to the individual products mentioned above, as it combines the three strategies. Indeed, the sweet–sour taste of water kefir could be appetising to piglets and have the same positive effect on overall nutrient intake as water flavouring found by Da Silva et al.[4] As the fermentation of water kefir grains produces organic acids,[13] it could support a low stomach pH and assist in preventing post-weaning diarrhoea. Water kefir also provides a combination of the probiotic microbes specified above. Its grains require (besides water) sucrose as a main substrate, and additional nutrients (vitamins, minerals, and amino acids) are commonly provided by dried figs, but other sources such as molasses may be used as well.[15] Production is possible via both aerobic and anaerobic fermentation. Aerobic fermentation promotes the growth of acetic acid bacteria more than anaerobic fermentation, whereas lactic acid bacteria and yeasts are not significantly affected by the type of fermentation.[13] Water kefir production only requires limited resources. Costs are restricted to the one-time purchase of water kefir grains, as subsequent cultivation occurs on-farm at room temperature, and the running costs of purchasing nutrient sources for continuous culturing. Although contamination of water kefir cultures cannot be totally excluded, the few available literature indicates little to no pathogen growth in water kefir cultures.[14,16] Therefore, water kefir could become an alternative to a wide range of patented commercial feed supplements. Yet, to our knowledge, no scientific report on the potential of water kefir in pigs is available. Furthermore, previous studies have only focused on the small-scale production of water kefir, limited to production levels of 100–1000 ml under laboratory conditions,[12–14] and this quantity is clearly too low for farm practice. To date, water kefir has not been produced at an industrial scale but only at the household level.[14]

The aim of this pilot study was to establish a suitable method for producing sufficient amounts of water kefir and distribute it daily to weaned piglets on a farm, assess the piglets' voluntary intake of water kefir (here referred to as palatability) and observe whether water kefir creates any problems in the piglets. Accordingly, we formulated the following hypotheses: (1) The production of water kefir in a larger quantity is feasible using white sugar and molasses as nutrient sources in the culture medium, and this will result in a constant pH and concentration of organic acids in the end product over a longer period of time; (2) weaned piglets will willingly drink water kefir; (3) the growth of healthy weaned piglets will be at least as good with water kefir as without; and (4) lighter piglets may react differently from heavier piglets to water kefir.

MATERIALS AND METHODS

Water kefir production

There are various methods to cultivate water kefir.[11] The present cultivation method based on wet water kefir grains was adapted from previously published methods[12,13] in a way that it is easily applicable at the farm level. In the method tested, water kefir grains represented 10% (w/v) of the water volume, and the nutritive substrates were white sugar and sugar beet molasses purchased from grocery stores. The target white sugar content was adjusted to simplify the repeated preparation of 27 L kefir with two bags (1 kg) of white sugar, the standard package size in grocery stores. The culture medium consisted of warm tap water to dissolve the white sugar and molasses, adjusted with cold tap water to 26 ± 1°C (74 g/L of white sugar and 1 g/L of sugar beet molasses). This medium was complemented with 100 g/L water kefir grains. The culture was left at room temperature (here: 22°C) in the dark in two lid-covered but not airtight food-grade drums (Brau- und Rauchshop, Densburen, Switzerland; OBI, Schaffhausen, Switzerland). These conditions were chosen to make the cultivation as simple as possible, preventing the pressure that could occur in airtight containers while also protecting the culture from contact with flies, dust, or other contaminants.

To initiate the process, a starter culture consisting of 60 g water kefir grains intended for human consumption was purchased from a local pharmacy and cultured in two parallel batches in serial back-slopping cycles of 48–72 h each. With this technique, about 10% water kefir grain (fresh weight) was produced from each back-slopping. Every week, half of each batch of water kefir grains was exchanged with the other batch to ensure that the water kefir cultures remained similar in both batches. It took 7 months to obtain 2 × 2.7 kg fresh kefir grains to produce 2 × 27 L water kefir.

For the animal experiment, the culture conditions were maintained, but the back-slopping cycles were shortened to 24 h to have access to the same amount of water kefir every day. The culturing took place in two 30 L food-grade plastic drums (OBI, Schaffhausen, Switzerland) equipped with a tap (Figure 1). To facilitate the transfer of water kefir grains from one drum to the next at each back-slopping, the 2.7 kg grains were always retained in a 25 L mesh bag (mesh size <3 mm; Brau- und Rauchshop, Densburen, Switzerland). The weight of the water kefir grains was adjusted before the next back-slopping. During the 2-week break between experimental runs (see below), no water kefir was needed. During that time, water kefir grains were preserved at 4°C in two drums, each filled with 20 L tap water but without nutrients until the day before weaning of the next group of piglets, when the water kefir grains were cultivated again in fresh medium.

Animals and experimental set-up

The Swiss cantonal authorities for animal experimentation approved the present experiment (licence: ZH182/19). The experiment took place on the pig farm of the research station AgroVet-Strickhof (Lindau, Switzerland) between March and July 2020. Due to the limited number of pens available and to observe the effect of water kefir over several production batches, the experiment consisted of five identical runs. The farm utilised an All-In All-Out system with a 3-week production rhythm. Therefore, a new experimental run started every third week. Four-week-old piglets (from a Landrace or Duroc boar) were separated from the sows (9–12 breeding sows, either Landrace or Landrace × Large White per run) and brought to the neighbouring rearing unit and kept in either of two barns, each with four pens. In total, 175 piglets were pure Landrace, 197 piglets were Duroc × Landrace, and 142 piglets were Duroc x (Landrace × Large White). The pens were 11 m^2 with solid concrete (3/4) or slatted (1/4) floors and included sawdust as a bedding material and a heated nest. Two pens in one of the two barns were also equipped with a water meter. For each run, piglets were distributed to the four pens in a randomised block design. The piglets were blocked by live weight at weaning (as routinely practiced on commercial farms) to establish group homogeneity and limit the stress for the lighter piglets. A threshold of 9 kg of live weight was used to allocate piglets to the lighter or heavier groups, as this was the empirical average live weight of weaning piglets on this farm. One pen of lighter piglets and one pen of heavier piglets received the liquid water kefir obtained from fermentation with water kefir grains, while lighter and heavier piglets in the other pens served as a control and only received commercial feed (Figure 2). For each water kefir group, and independently of the number of piglets, a fresh drum of water kefir (27 L) attached to a nipple drinker (Figure 1) was installed every morning and left available until the next morning. This quantity of water kefir was considered sufficient for ad libitum access based on an expected total liquid consumption of about 1 L per piglet per day.[17] The procedure was repeated for 5 days from weaning onwards. Water (two drinkers per pen; also in the water kefir group), commercial pre-starter feed (first week; 14.5 MJ/kg metabolisable energy and 160 g/kg crude protein) and starter feed (following weeks; 13.7 MJ/kg metabolisable energy and 165 g/kg crude protein) were offered ad libitum. Piglets were weighed on the 7th and 28th days following weaning. Water consumption over the 28 days of the experimental run was controlled by water meters installed in one control and one kefir pen ($n = 3$; Figure 2).

FIGURE 1 Drum attached to a nipple drinker with a piglet consuming water kefir from the drum

Water kefir analyses

To control the fermentation process of the water kefir, the pH and concentrations of lactic and acetic acid were measured. The pH of the water kefir was measured at the end of the fermentation after removing the water kefir grains before distributing it to the piglets in each drum, and the average pH value was calculated for each day. During the 5 days when the piglets received the water kefir, 1 ml was sampled every day from each drum before installing it above the pens. These samples from both drums were pooled across the 5-day periods (producing five pooled samples, one per run) and stored at −20°C.

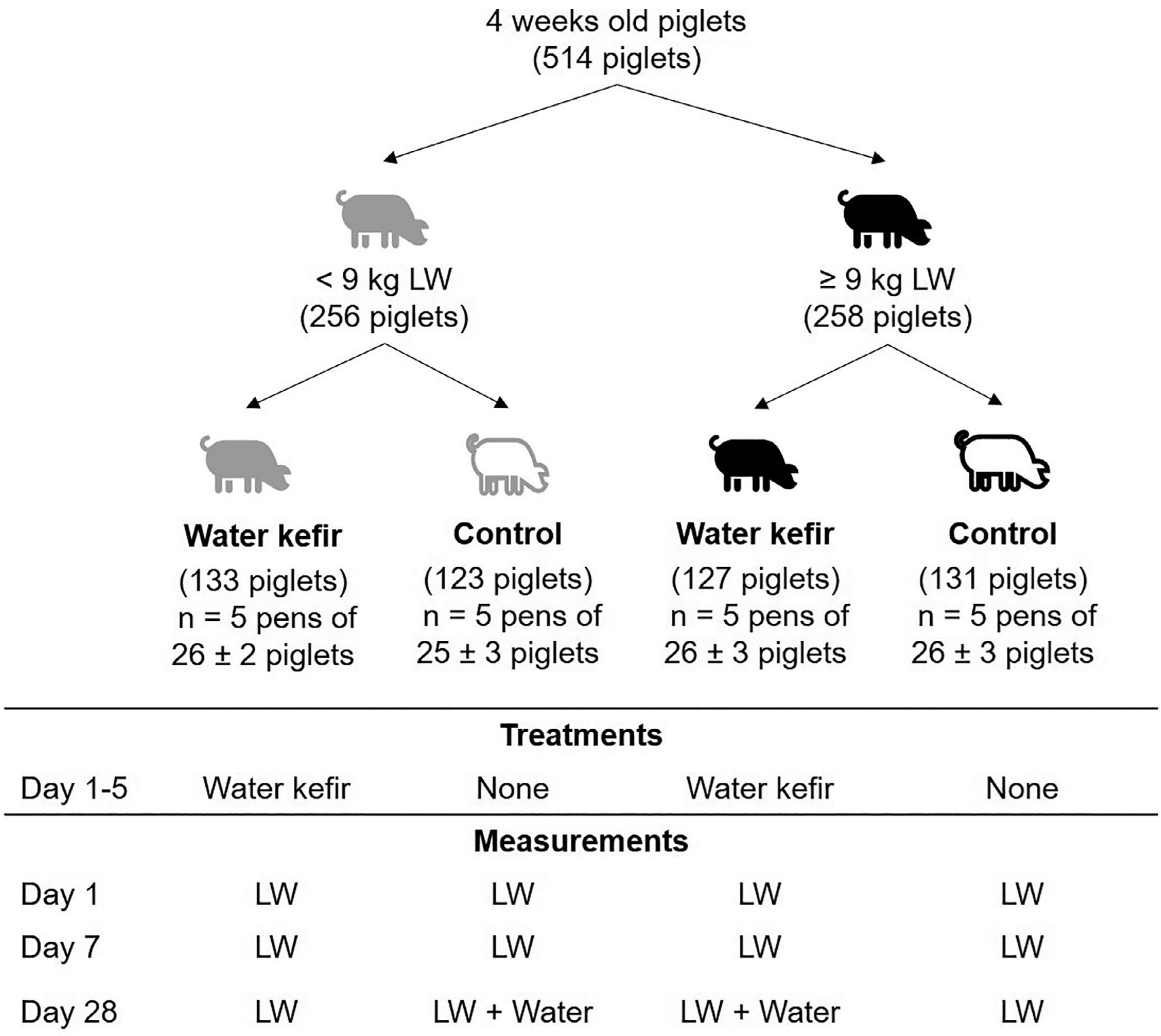

FIGURE 2 Overall experimental set up and measurements (LW, live weight, water, water consumption)

Lactic acid and acetic acid concentrations were quantified using high performance liquid chromatography (La Chrom, L-7000 series, Hitachi Ltd., Tokyo, Japan) according to Ehrlich et al.[18]

Statistical analysis

The statistical analysis was conducted with SAS 9.4 (SAS Institute, Cary, NC, USA). The effect of distributing water kefir on piglet live weight and weight gain was tested for each piglet weight class with an analysis of variance (general linear model procedure). Hence, the fixed effects in the model were kefir distribution, piglet weight class, and their interaction. The pen was considered the experimental unit, and the experimental run was included as a random factor. The least square means of the interaction were then compared using contrast analysis to test the effect of water kefir on lighter piglets, on heavier piglets and on both lighter and heavier piglets.

RESULTS AND DISCUSSION

Feasibility and repeatability of the water kefir production

Kefir grains are not available commercially in large quantities. The establishment of sufficient water kefir grains to produce water kefir at the farm level required a long period of time (7 months), but it is only necessary at the start of the process, and this period may be shortened by shortening the fermentation time to 24 h, as practiced during the animal experiment. The recipe and materials were chosen to simplify the production and daily distribution of two drums of water kefir to the weaned piglets. The boundary conditions were the limited number of ingredients, their easy dosage, their solubility in water (as opposed to dried figs), and the simple retaining of water kefir grains in mesh bags. The method of conserving water kefir in tap water and at 4°C during the experimental breaks would also be suitable on farms during periods when no weaning takes place. The pH and lactic acid concentrations remained stable across the runs, whereas the acetic acid concentrations were more variable between repetitions (Figure 3). This partly confirmed our first hypothesis. Our finding of a greater proportion of acetic acid compared to lactic acid is different from the results of Laureys and De Vuyst.[12] The technique without airtight containers used in the current experiment created semi-aerobic conditions, which might have promoted the growth of acetic acid bacteria over that of lactic acid bacteria.[13] Furthermore, the concentrations of both lactic and acetic acid were lower than those reported by Laureys and De Vuyst.[12] Greater concentrations of lactic acid and acetic acid could likely be achieved by increasing the nutrient (sugar and molasses) concentration in the medium and by prolonging the fermentation time to 72 h, as practiced by others.[12,13] However, the latter measure would require a threefold larger quantity of water kefir grains than in the present experiment for a daily availability of 54 L water kefir. It also would require more time for the initial cultivation phase to obtain the necessary quantity of kefir grains as well as more drums, but the procedure would not be more time-consuming afterwards.

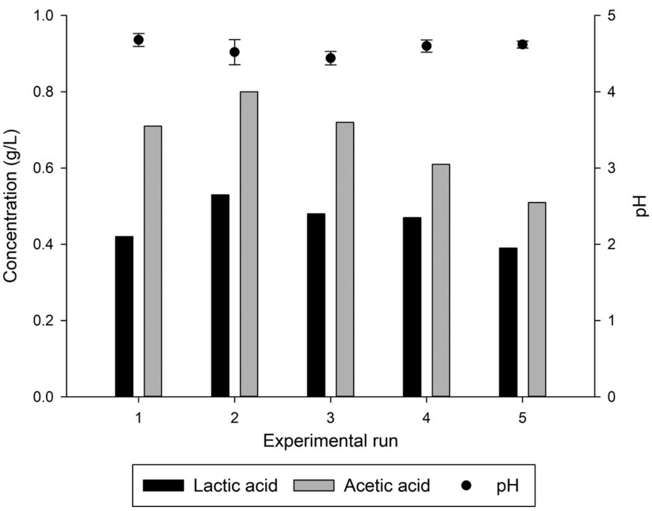

FIGURE 3 Concentrations of lactic and acetic acid in in the five experimental runs (one pooled sample per experimental run; left axis) and average end pH value and standard deviation ($n = 5$ per experimental run; right axis) as measured in the water kefir (liquid without grains)

Acceptance of water kefir and its effects on weaned piglets

Piglets started using the nipple drinker attached to the water kefir as soon as they entered the pen. From the third day onward, fights between piglets were observed when installing a fresh drum of water kefir. This may be relevant regarding piglet stress and may be resolved by distributing water kefir from more than one nipple per pen. However, based on the expected liquid consumption of piglets of this weight,[17] we assume that the quantity distributed (on average, approximately 1 L water kefir per piglet and day) allowed the consumption of water kefir by all piglets regardless of their hierarchy ranking in the group. All of the available water kefir was consumed by the piglets within 24 h, which confirms our second hypothesis about the palatability of water kefir. As observed visually, almost no spoilage occurred. The possibility that piglets had spoilt the water kefir by playing with the nipple was considered unlikely, as the bedding material remained mostly dry around the drinker. It was not expected that the 27 L drum would be emptied within 24 h because the piglets receiving water kefir consumed water as well (about 0.7 L/day per piglet), and the average water consumption of piglets of this size is limited to about 1 L/day.[17] To determine the full intake capacity of piglets, water kefir should be offered in even greater amounts. Even so, producing the amount required per day per pen of 20–30 piglets seems manageable on a farm, and using the drum technique would make applying the feeding of water kefir possible on larger pig breeding farms with limited effort.

No piglets died during the present experiment, and no piglet presented clinical signs of disease. Water kefir had no significant effect on piglet live weight at any time point (Table 1). However, average daily

TABLE 1 Least square means of the average live weight (LW) and average daily gains (ADG) of the piglets in each group ($n = 5$ pens) and standard error of the mean (SEM)

	Lighter (L)		Heavier (H)			Contrasts *p*-values		
Group	Control (C)	Kefir (K)	Control	Kefir	SEM	LC vs. LK	HC vs. HK	C vs. K
LW (kg)								
Weaning (day 0)	8.07	8.36	10.14	9.95	0.621	0.744	0.831	0.936
Day 7	9.14	9.04	11.04	10.61	0.695	0.928	0.665	0.711
Day 28	20.2	20.1	22.9	22.4	1.06	0.963	0.763	0.806
ADG (g)								
Day 0–7	152	97	129	94	21.5	0.097	0.274	0.059
Day 0–28	433	420	454	445	17.6	0.615	0.705	0.532

gains tended ($p < 0.10$) to be negatively affected by the access to water kefir, and this was particularly true for the lighter piglets. This only partly confirms our third hypothesis, and it opposes the fourth hypothesis. A possible explanation for this is that piglets consuming such large amounts of water kefir (on average 1 L per piglet and day), which has low nutrient density, reduced their solid feed intake to an amount that caused their total nutrient intake to decline. In contrast, Da Silva et al.[4] reported increased feed intake and weight gain as a consequence of increased water intake in the case of flavoured water. Another explanation could be that the time the piglets invested in drinking water kefir and fighting for it reduced the time spent eating. Offering the feed together with water kefir might reduce the eventual substitution of feed for water kefir, thus supporting total feed and nutrient intake.

CONCLUSIONS

A simple method to produce sufficient water kefir for piglet feeding from one purchased mother cultivation batch was developed, and the quality of the kefir remained quite stable over an extended period of cultivation. This method could also be used for animals other than piglets, such as calves. The piglets' voluntary water kefir consumption was as high as the total liquid consumption expected for this live weight. Hence, the palatability of water kefir is sufficient to conduct further research. It must be clarified whether better management of the supply of water kefir and solid feed, higher nutrient density of the water kefir or a restriction of the water kefir supply would eventually lead to the highest total nutrient intake. The consumption of water kefir did not lead to clinical health issues in piglets. The next step will be to test the effectiveness of water kefir supplementation in a situation with a great prevalence of diarrhoea after weaning to demonstrate its presumed ability to support the health of weaned piglets.

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