Influence of the application methods of iron/anticoccidial products on the behaviour and associated stress factors of suckling piglets

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Abstract

The aim of the present study was to evaluate the effect of different treatment protocols for the control of iron-deficiency anaemia and coccidiosis, one combination injection versus injection of iron and oral application of toltrazuril, on the general behaviour of piglets. Piglets were divided into three experimental treatment groups: 24 piglets were left untreated; (control group, C); 24 piglets received an oral administration of generic anticoccidial agent (20 mg/kg BW) plus intramuscular administration of iron dextran (200 mg/ml; 1 ml/piglet) in the same handling (oral + parenteral group, O+P) and 24 piglets received an intramuscular application of the combination product (parenteral group, P). As expected, the handling time was higher in the oral + parenteral group than in the parenteral group. Regarding the behaviour of piglets, the time spent suckling after treatment was variable in the control and oral + parenteral groups, while piglets from the parenteral group spent more time suckling, and hence, their time spent resting was decreased. A statistical trend of lower concentration of cortisol was observed in P compared to O+P after processing. Regarding productivity indices, the average daily gain of piglets during lactation and, subsequently, their weaning weight were higher in P compared to O+P, and similar to the control. In conclusion, administering a combination product injection decreases the time of administration compared to individual treatments.

Welfare, iron deficiency anaemia, coccidiosis, route of administration

During the first days of life, newborn piglets are routinely subjected to manipulation and handling, together with several simultaneously invasive procedures. Ensuring adequate colostrum and milk intake during these early days is essential for the proper development of immunity and the optimum growth of piglets (Blavi et al. 2021). Iron-deficiency anaemia (IDA) is the most common mineral deficiency. A high percentage of piglets from hyper prolific sows is born already anaemic (Lipiński et al. 2010). Piglets require supplemental iron, as the already low levels will gradually decrease during the first week of life; this is due to the low iron stores at birth and the low iron content in milk (Rincker et al. 2004). At the same time, coccidiosis caused by *Cystoisospora suis* is one of the most frequent causes of diarrhoea in suckling piglets (Joachim et al. 2018). Within the first week of life, piglets on farms positive for coccidiosis are subjected to parenteral iron injection and routine oral metaphylactic administration of toltrazuril-based products, beside other procedures, like teeth clipping, tail docking, identification and castration of male piglets. These practices are invasive, require longer period of handling and can be stressful for piglets and sows (Brown et al. 1996), with a consequent effect on the behaviour and welfare of piglets (Noonan et al. 1994; Marchant-Forde et al. 2014). It is important to know not only the handling time of different treatment protocols, but also the time budget per piglet. A time budget is a log of the sequence and duration of activities engaged in by an individual over a specified period of time

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(Szalai 1966). According to Leslie et al. (2010) and Valenzuela et al. (2016), the time budget dedicated to each activity (e.g. suckling, resting, exploration, etc.) may be affected by the different protocols immediately after administration. The recently introduced, first injectable combination product for concomitant treatment of IDA and prevention of diarrhoea caused by coccidiosis (Forceris®, CEVA Santé Animale, Libourne, France) is labelled for use during the first 3 days of life. It combines in a single injection the 2 previous processing procedures: the iron injection and the oral administration by drenching of toltrazuril-based products. Early and simultaneous application of iron and toltrazuril within the first 3 days of life is more effective to control both conditions than later separate administrations (Sperling et al. 2018; Hiob et al. 2019). The use of a combination product reduces the number of handlings needed. Consequently, it is likely that the possible stress and negative impact on the behaviour, time budget and welfare in connection with multiple drug administrations will be reduced.

The aim of the present study was to evaluate and compare the effect of different treatment protocols for the control of IDA and coccidiosis on the general behaviour of piglets and time budget: one combination injection versus iron injection and oral application of toltrazuril.

Materials and Methods

Animals and facilities

The study was conducted in the Swine Research Centre located in Aguilafuente (Segovia, Spain). A total of 36 male and 36 female piglets (Topigs TN70 sow × Pietrain boar) were included in the study. The six sows used were hybrid commercial pigs (Topigs TN70; third to fifth parturition) with the same litter size. Animals were healthy at the start of the study and no vaccinations were applied during the study. Piglets were three days old (48–72 h after birth) when the study started (initial body weight, BW: 1.5–2.0 kg), and their final age was 21 days old (final bodyweight: 5.0–6.0 kg). The lactation room had controlled environmental conditions (light, 16L:8D; temperature, from 23 °C at farrowing to 20 °C at weaning; relative humidity, 60 to 75% by a forced ventilation system) and the nests of piglets were equipped with infrared light sources that allowed a temperature between 32 °C and 28 °C to be maintained from farrowing to weaning. As a normal management on the farm, piglets were offered creep feed (milk replacer Healthy, Provimi B.V., Rotterdam, The Netherlands) from day 15 of lactation onwards.

All the experimental procedures were approved by the Animal Ethics Committee of the National Institute for Agricultural and Food Research and Technology (ORCEEA 2019-08) and were in compliance with the Spanish guidelines for care and use of animals in research (BOE 2013), in agreement with European Union Regulation 2010/63/UE.

Experimental design

Within the first 24 h after birth, piglets were individually identified with ear tags and weighed. The litters were equalized to 12 piglets/litter by cross-fostering between sows that farrowed on the same day. After day 1, piglets were kept in the same litters and piglet allocation to the treatments was carried out at random within each litter, by using computer-generated random allocation. Piglets were distributed into three experimental treatment groups: 24 piglets were left untreated (no treatment with iron/anticoccidial agents, no handling (control group, C); 24 piglets received an oral administration of generic anticoccidial agent (20 mg/kg BW; Baycox 5%, Elanco, Buenos Aires, Argentina) plus intramuscular administration of iron dextran (200 mg/ml; 1 ml/piglet; Calidex-G®; CALIER S.A., Barcelona, Spain) in the same handling (oral + parenteral group, O+P); 24 piglets received an intramuscular application of the combination product (1.5 ml; Forceris®, CEVA Santé Animale, Libourne, France) (parenteral group, P).

The procedure of application of each treatment (O+P or P) was performed by the same personnel. For its evaluation, the time of handling was considered, and it was described as the exact period (measured in seconds) between picking up the piglet and final release.

Two days before the probable farrowing date, 6 double infrared video cameras (Sricam® SP017, Shenzhen Sricctv Technology CO., Ltd, Shenzhen, China) were installed in each farrowing cage. The video information was captured and stored using a digital video recording system and an external memory drive. The cameras began with the recording mode 3 h before (−3 h, −2 h, and −1 h) iron/anticcocidial supplementation and continued up to 3 h after (+1 h, +2 h, and +3 h), as described by Leslie et al. (2010). All video images were analysed by two observers with the VLC software (version 3.0.12, VideoLAN Organization, Paris, France). Scan sampling every 3 min was used. Briefly, the observers recorded the behaviours of the instant at the 3 min mark. This allowed to capture the briefest behavioural state of interest. Once the repeatability of both observers was confirmed, each observation was considered as valid. The number of piglets performing each of the 7 behaviours described in Table 1 was registered at each sampling point (120 sampling points). Also, the 7 different behaviours were classified as active behaviours, which implied movement (suckling + positive interactions + normal locomotion + exploration) and neutral behaviours (resting + sitting + standing). The number of piglets and time spent out of sight was also registered.

Table 1. Description of behaviours of 2-days-old piglets according to Fraser and Broom (1997) and Leslie et al. (2010).

Behaviours	Description
Suckling	Teat in the mouth, vigorous rhythmic movements
Positive interactions	Includes: grooming behaviour between piglets or between piglet and sow, play behaviour such as locomotor play (running, jumping, spinning) between piglets or directed towards the sow or parts of the crate by a piglet
Resting	Recumbent position, resting or sleeping with the head up or the legs and head outstretched
Sitting	Body weight supported by hindquarters and front legs
Standing	Body weight supported by all four legs
Normal locomotion	Forward movement in a four-time gait from point A to point B, all four limbs are involved
Exploration	Piglet extends neck towards part of the environment and looks at or sniffs at an object
Movement	Suckling + positive interactions + normal locomotion + exploration
Neutral	Resting + sitting + standing

Blood sampling was done as follows:

On day −1: blood samples (≥ 3 ml per piglet) were collected from 6 animals per litter (12 piglets/treatment). For that, piglets in each treatment were ordered by BW (Pig 1 – the heaviest, to Pig 12 – the lightest), and blood was sampled from piglets with alternate numbers (Pig 2, Pig 4, Pig 6, Pig 8, Pig 10 and Pig 12).

On day 0 (day of application treatment): blood samples (≥ 3 ml per piglet) were collected from 6 animals per litter (12 piglets/treatment). For that, pigs in each treatment were ordered by BW (Pig 1 – the heaviest, to Pig 12 – the lightest), and blood was sampled from piglets with alternate numbers (Pig 1, Pig 3, Pig 5, Pig 7, Pig 9 and Pig 11). Blood sampling was done after behaviour recording.

On day 21: blood samples (≥ 3 ml per piglet) were collected from 6 animals per litter (12 piglets/treatment). For that, pigs in each treatment were ordered by BW (Pig 1 – the heaviest, to Pig 12 – the lightest), and blood was sampled from piglets with alternate numbers (Pig 1, Pig 3, Pig 5, Pig 7, Pig 9 and Pig 11).

The concentration of cortisol was analysed in serum samples with a commercial ELISA kit (Invitrogen™ EIAHCOR, Fisher Scientific S.L., Madrid, Spain). The assay was ranged from 100 to 3200 pg/ml, sensitivity was determined to be 17.3 pg and the inter-assay and intra-assay variation coefficient were 8.1% and 8.8%, respectively.

Piglets were also weighed at weaning and the average daily gain (ADG) from birth to weaning was calculated.

Statistical analysis

Statistical analysis was performed using SAS software (version 9.4; SAS Inst. Inc., Cary, NC). The time budget, minutes and percentages of time allocated to each behaviour before and after treatment administration were calculated. A generalized linear mixed models (PROC GLIMMIX in SAS) was used to compare the groups. The growth of piglets was analysed using a linear mixed model (PROC MIXED in SAS) according to the completely randomized design. The model includes treatment as the fixed effect and the sow within the treatment. The BW of piglets at birth was used as a covariable in the analysis of ADG and BW of piglets at weaning.

Basal cortisol concentration was analysed using ANOVA test after initial normality check and identification of outliers (ROUT, Q = 1%) using GraphPadSoftware (San Diego, CA 92108, USA).

Least squares means (LS means) were computed for each effect with the Tukey adjustment. A significance level of $P < 0.05$ was applied.

Results

Handling time

As expected, the procedures of application of different iron/anticoccidial supplementation showed that group O+P had a numerically higher handling time (mean ± SEM) (198.5 ± 22.98 s) compared to group P (111.5 ± 22.98 s) but it was not significant ($P > 0.05$). As already mentioned, piglets from group C were not handled, therefore, no handling time was registered for this group.

Time budget

Regarding the time budget distribution of the different observed behaviours, Table 2 shows values before treatment administration, considering the 3 h evaluated. There were no differences in any of the behaviours studied ($P > 0.05$). It can be observed that animals from the three experimental groups spent most of the time with neutral behaviour (mean ± SEM) (70.2% ± 8.97%), and within it, they spent a mean of 64.2% ± 8.11% of the time resting. In terms of active behaviour, piglets spent 26.0% ± 8.95% in movement, and within this, they spent a mean of 17.3% ± 9.30% of the time suckling.

Table 2. Percentage of time (%) dedicated to each of the observed behaviours before the treatment administration in the control (C), the oral + parenteral (O+P), and the parenteral (P) groups.

Task	C	O+P	P	SEM	*P* value
Suckling	21.61	17.80	12.63	9.30	0.80
Positive interactions	7.88	5.31	5.03	2.57	0.72
Resting	54.52	66.17	71.85	8.11	0.42
Sitting	0.77	1.38	0.90	0.33	0.42
Standing	6.61	3.17	5.35	1.33	0.24
Normal locomotion	1.11	1.04	1.39	0.40	0.81
Exploration	0.07	0.69	0.49	0.38	0.53
Out of sight	7.44	4.44	2.36	1.69	0.25
Movement[1]	30.67	24.83	19.54	8.95	0.71
Neutral[2]	61.89	70.73	78.10	8.97	0.52

[1] Movement: suckling + positive interactions + normal locomotion + exploration
[2] Neutral: resting + sitting + standing

Values after treatment administration are shown in Table 3. Positive interactions were greater ($P = 0.06$) immediately after treatment administration (+1 h) due to the fact that the piglets were handled. Moreover, although there were no differences due to the treatment, the percentage of time spent with positive interactions was far higher in groups O+P and P than in group C. On the other hand, although there were some differences because of the time in the sitting ($P = 0.06$) and standing ($P = 0.04$) behaviour, both fall under the category of neutral behaviour, and considering the neutral conduct, the time was not affected. It should be noted that in group C more piglets were out of range ($P < 0.01$) than in the other two groups. This is probably because after treatment administration, the piglets spent increased time suckling, and therefore were closer to the mother, whereas C piglets, not being manipulated, were able to move freely throughout the pen.

As already mentioned, the main movement and neutral behaviours are suckling and resting, respectively, therefore, the analysis was focused on them. Figure 1 shows the variation in the percentage of time that piglets dedicated to suckling 1, 2, and 3 h after treatment administration, and taking as a reference value the percentage of time spent suckling 3 h before the treatment. It can be observed that variations in the behaviour of C piglets are appropriate for normal behaviour of animals due to the fact that these animals did not undergo any handling and consequent stress. However, some differences were observed in the treated animals. Immediately after treatment administration (+1 h), piglets from groups O+P and P spent an increased time suckling; the increase was practically equal for both groups. Nevertheless, at two hours after treatment administration (+2 h), meanwhile piglets from group O+P decreased the time spent suckling, those from the parenteral group increased it more than 20%. Finally, at three hours after treatment administration (+3 h), animals from group O+P increased again the time spent suckling,

Table 3. Percentage of time (%) dedicated hourly after the treatment administration to each of the behaviours observed in the control (C), the oral + parenteral (O+P), and the parenteral (P) groups.

Task	+1 h			+2 h			+3 h			SEM	*P* value		
	C	O+P	P	C	O+P	P	C	O+P	P		P_{treat}	P_{time}	$P_{treat \times time}$
Suckling	13.54	23.90	20.46	31.25	15.45	33.75	8.57	19.86	18.75	12.20	0.80	0.55	0.78
Positive interactions[1]	2.29	9.21	18.60	4.38	0.00	2.71	4.59	8.56	5.94	3.31	0.20	0.06	0.12
Rest	61.46	44.52	52.16	53.33	75.76	52.92	70.13	55.14	68.96	15.07	0.95	0.63	0.63
Sitting[2]	1.25	0.88	0.86	0.00	2.51	5.63	1.46	1.60	1.23	2.06	0.47	0.06	0.20
Standing[3]	11.04	8.11	4.13	1.25	2.72	2.08	4.49	4.61	2.86	2.36	0.40	0.04	0.61
Normal locomotion	1.88	0.22	1.88	1.25	0.00	0.63	0.52	3.91	1.63	1.51	0.99	0.55	0.49
Exploration	0.63	2.19	0.66	0.63	2.09	1.25	1.25	1.32	0.43	0.95	0.33	0.92	0.90
Out of sight	7.92	10.96	1.26	7.92	1.46	1.04	8.98	5.00	0.21	2.31	0.01	0.27	0.29
Movement[4]	18.33	35.53	41.59	17.55	17.55	38.33	33.66	33.66	26.74	11.36	0.65	0.64	0.29
Neutral[5]	73.75	53.51	57.15	54.58	80.99	60.63	76.08	61.64	73.05	12.63	0.93	0.67	0.35

[1] There was a trend for a significant time-effect: 6.02%[a], 1.42%[b] and 3.82%[ab] for +1 h, +2 h and +3 h, respectively ($P = 0.0563$)
[2] There was a trend for a significant time-effect: 0.60%[b], 1.63%[a] and 0.86%[ab] for +1 h, +2 h and +3 h, respectively ($P = 0.0592$)
[3] There was a trend for a significant time-effect: 4.66%[a], 1.21%[b] and 2.39%[ab] for +1 h, +2 h and +3 h, respectively ($P = 0.0426$).
[4] Movement: suckling + positive interactions + normal locomotion + exploration
[5] Neutral: resting + sitting + standing

although they did not reach the values of time dedicated to suckling of piglets from group P. Overall, it can be observed that while groups C and O+P had fluctuations in the time spent suckling after treatment administration, piglets from group P spent more time suckling, to a greater or lesser extent, over the 3 h.

Similar to Fig. 1, Fig. 2 shows the variation in the percentage of time that piglets dedicated to resting at 1, 2, and 3 h after treatment administration, taking as a reference value the percentage of time spent resting 3 h before the treatment. As mentioned, the time that piglets are not in movement, is considered neutral behaviour. In addition, most of the time that the piglets spent in neutral corresponded to resting behaviour, while most of the time that piglets spent in movement corresponded to suckling behaviour. Therefore, it makes sense that the variation in time represented in Fig. 2 is almost opposite to that represented in Fig. 1. In this regard, the fluctuations that were seen in suckling were also found in the resting behaviour for groups C and O+P. Meanwhile, the variation in the time spent resting in piglets of group P was decreased and, therefore, in accordance with the increase in the suckling time (Fig. 1).

Growth performance

The growth performance of piglets is reported in Table 4. The animals had comparable BWs at the beginning of the trial ($P > 0.05$). However, the ADG during lactation was higher in piglets of group P (similar to C) compared to group O+P ($P < 0.01$). As a consequence, the BW of piglets at weaning was also higher in groups C and P than in group O+P ($P < 0.01$).

Table 4. Body weight at birth and at weaning and average daily gain during lactation (28 days) period of piglets from different experimental groups: negative control, oral + parenteral, and parenteral group.

Task	Negative control	Oral + Parenteral	Parenteral	SEM	*P* value
Body weight at birth (kg)	1.66	1.62	1.40	0.071	0.15
Body weight at weaning (kg)	7.50[a]	7.04[b]	7.77[a]	0.163	0.01
ADG (g/day)	282.9[a]	261.2[b]	295.9[a]	7.753	0.01

[a,b] Values in the same row with no common superscript are significantly different ($P \leq 0.05$); SEM - standard error of the mean; ADG - average daily gain.

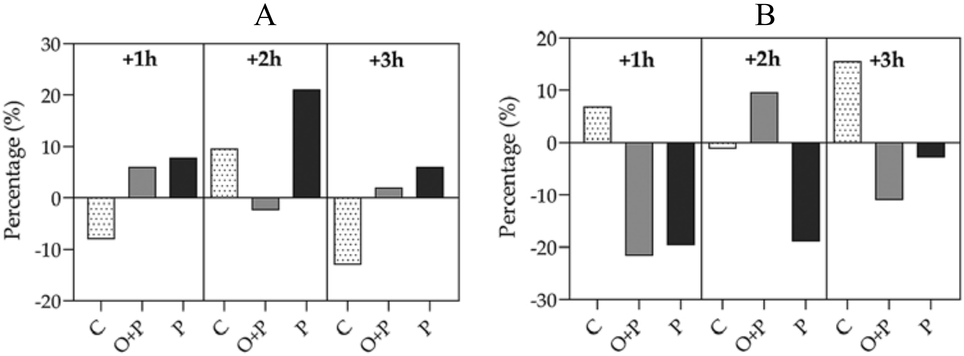

Fig. 1. Variation in the percentage of time (%) dedicated to: A) Suckling and B) Resting at one (+1 h), two (+2 h) and three (+3 h) hours after treatment administration, taking as reference value the percentage of time dedicated to each behaviour 3 h before the treatment administration, observed in the negative control (C), the oral + parenteral (O+P), and parenteral (P) groups.

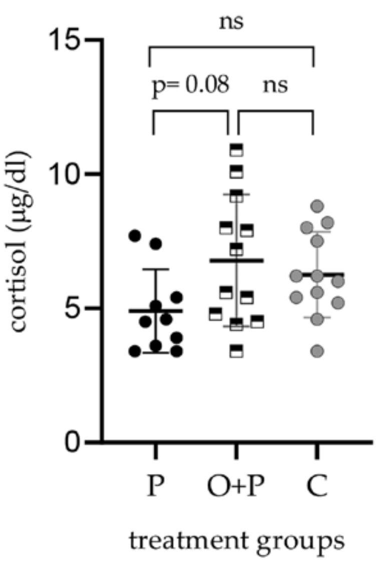

Fig. 2. Distribution of levels of basal concentration of cortisol observed on day 0 for injectable group compared to oral + parenteral one and control

Basal cortisol

There were no differences between the experimental groups ($P > 0.05$) in the basal cortisol concentration one day before the treatment administration (day − 1) and on day 28 (end of the trial). Statistical trend of lower cortisol concentration was observed on day 0 for group P compared to O+P ($P = 0.08$) with a mean value (± SD) of 4.9 µg/dl (± 1.557) vs. 6.78 (± 2.46) in groups P and O+P, respectively (Fig. 2).

Discussion

Neonatal piglets are routinely subjected to manipulation and handling during the first days of life. Within this management practice involving different invasive interventions, animals are subjected to parenteral iron injection and routine metaphylactic oral administration of toltrazuril-based products on farms positive for coccidiosis. The present work evaluated the effect of different treatment protocols for control of iron-deficiency anaemia and coccidiosis: injection with oral administration versus injection only, on the general behavioural effects on the time budget of piglets. The growth of piglets during lactation was also studied.

Although differences were not significant because of the low number of piglets used, injection with the combination product reduced the handling time compared to separate administration. A similar reduction of handling time was described previously (Carceles et al. 2021; Garza et al. 2021). Handling time per piglet was shorter in both scenarios: comparison of combination vs. oral administration + injection as well as part of full piglet management applied on farm (iron administration, toltrazuril treatment, ear tagging, tail docking).

Restraint and handling of the piglets are well known stressors with negative effects that remain after piglets are released (Noonan et al. 1994). Piglets that experienced an additional stressor to handling (such as the additional oral administration by drench in this study) showed non-specific behaviours, which became more frequent. Noonan et al. (1994) also observed differences in specific behaviours, especially a reduction in the period of suckling in the extra-handled piglets, similar to the combination product group observed in our study.

The route of administration itself can be considered as less or more stressful, as previously described for iron administration (Marchant-Forde et al. 2014). Administration of iron paste was considered as more stressful, taking into consideration the time needed to carry out the procedure compared to injection. Single procedures that can be performed more quickly are likely to have a lower impact on well-being (Marchant-Forde et al. 2009). Oral iron supplementation resulted in a higher behavioural disruption in neonatal pigs, probably associated with the increased handling time (Salmon-Legagneur and Fevrier 1956) and an unpleasant and persistent metallic taste of oral iron (Valenzuela et al. 2016).

In this regard, as expected, the percentage of time spent in each type of behaviour will be similar between groups before the treatment administration. Piglets spent most of their time (70%) with neutral behaviour, which includes resting, sitting, and standing positions. The rest of the time was spent in movement, i.e., suckling, exploration, normal locomotion and positive interactions with their mother or other piglets. It is worth noting that the values obtained in the present work are in accordance with previous literature, where it has been reported that considering resting and suckling together, can occupy over 70% of the daily time budget of newborn piglets (Salmon-Legagneur and Fevrier 1956; Leslie et al. 2010; Valenzuela et al. 2016).

After treatment administration, some changes occurred. Toltrazuril as the active substance of the oral anticoccidial administered to piglets of group O+P is known for its bitter taste which might be one of the reasons for behaviour disruption. However, there are two hedonic tastes in piglets, sweet and umami. These flavours have demonstrated increased consumption, and therefore no rejection, by the piglets (Salmon-Legagneur and Fevrier 1956; Kennedy and Baldwin 1972; Danilova et al. 1999; Tinti et al. 2000).

In a recent work, Valenzuela et al. (2016) administered oral iron supplementation, which is also known for its unpleasant metallic taste, and described an increase of standing, restless, gasping and tongue shaking, and biting and exploring behaviours after the treatment administration, behaviours which all may be associated with the taste. In the current study, similar results were obtained and could be also associated with the bitter taste of the oral anticoccidial administered. In this respect, although there were no significant differences, higher exploration behaviour was detected in piglets treated with the O+P protocol compared with the single parenteral administration.

Piglets from group O+P also spent more time out of sight, probably since the bad taste in their mouth provoked by the oral treatment did not make them want to suckle, and they were more restless and moved more freely in the pen. On the contrary, piglets treated only with the P protocol went directly to the teat after treatment, thereby increasing the time spent suckling. Moreover, the difference from baseline in the percentage of time spent suckling before and after treatment administration in piglets of group P was higher than in piglets of the other groups during the whole time of study.

Although there were no significant differences in the protocol of administration of the treatments in the present work, the handling time in group O+P was almost 50% higher than in group P. An increase in handling time means more stress for the piglets (Noonan et al. 1994).

It is important to keep in mind that not only the stress associated with the handling but also the stress connected to multiple drug administration, which is obviously reduced when a combined treatment is administered. Furthermore, this is an important point when considering the overall management of a farm, as the reduction in management time that we have seen in this work must be extrapolated to the time savings from the parenteral treatment administration protocol.

Cortisol is one of the most widely used biomarkers to detect stress and most stress stimuli tend to increase cortisol in pigs (Martínez-Miró et al. 2016). As expected, there were no differences between experimental groups in basal cortisol concentration one day before the treatment administration (day − 1) and day 28 (end of the trial). In contrast, statistical trend for lower concentration of cortisol was observed on day 0 for group P compared to group O+P, suggesting possible lower level of stress. It is assumed that acute stress due to extensive manipulation caused increased cortisol concentrations especially in group O+P; in contrast, the cortisol profile was similar in both groups P and C which remained without any manipulation.

Those joined elements may explain differences between the groups observed in this study. To better understand the impact, future assessment of stress markers like cortisol should be considered.

In relation to growth, piglets that were given parenteral treatment only, i.e those that were less handled, showed the same growth as C piglets that were not handled at all. This could be due to the lower stress group P endured during the treatment administration compared to group O+P. A negative effect on productivity in addition to the potential welfare aspects of piglets was previously described, especially in a shorter period after processing, causing stress (Marchant-Forde et al. 2009). More stressed piglets were affected by depression of the growth rate between day 2 and 7 post procedure (Marchant-Forde et al. 2014). Oral treatment not only generated a bad taste but also a higher satiety sensation, and thus the piglets suckled less than those that were not given any oral treatment. Besides triggering satiety signals, bitter tasting ingredients can lead to pigs having a lower feed acceptability and negative effect on performance.

Drenching is defined as forced pouring of a liquid preparation down the throat of treated animals. As such, it can be considered as a stressor with a possible negative effect; misapplication may potentially cause irritation to the respiratory tract of piglets. Nevertheless, recently published research did not associate handling due to drenching with a negative impact on the survival or general health of low-birth-weight piglets (Van Tichelen et al. 2021).

In order to confirm and better understand the possible impact of this particular intervention on performance, data collected at more frequent time points would needed to be assessed and possibly, the size of the treatment groups increased.

In conclusion, the current study showed that administering the treatment to control iron-deficiency anaemia and coccidiosis together in a single parenteral injection decreases the time of administration in comparison with individual treatments for both conditions, making it more convenient. Regarding the general behaviour, this group of piglets spent more time suckling after treatment administration than those whose administration protocol involved both oral and parenteral treatments. Hence, their average daily gain during lactation was higher and therefore, so was their weaning weight.

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References

Blavi L, Solà-Oriol D, Llonch P, López-Vergé S, Martín-Orúe SM, Pérez JF 2021: Management and feeding strategies in early life to increase piglet performance and welfare around weaning: A review. Animals **11**: 302

Boletín Oficial del Estado (BOE) 2013: Real Decreto 53/2013, de 1 de febrero, por el que se establecen las normas básicas aplicables para la protección de los animales utilizados en experimentación y otros fines científicos, incluyendo la docencia. (In Spanish). Boletín Oficial del Estado **34**: 11370-11421

Brown J, Edwards S, Smith W, Thompson E, Duncan J 1996: Welfare and production implications of teeth clipping and iron injection of piglets in outdoor systems in Scotland. Prev Vet Med **27**: 95-105

Carceles S, Garza L, Oliver S, Casanovas C, Espigares D 2021: Time assessment of the administration of two alternatives to prevent anaemia and coccidiosis in piglets. In: Proceedings of the 12th European Symposium of Porcine Health and Management (ESPHM), Bern, Switzerland

Danilova V, Roberts T, Hellekant G 1999: Responses of single taste fibers and whole chorda tympani and glossopharyngeal nerve in the domestic pig, *Sus scrofa*. Chem Senses **24**: 301-316

Fraser A, Broom D 1997: Farm animal behaviour and welfare. CABI Publishing, Oxon, UK, pp. 69-267

Garza L, Casanovas C, Carceles S, Oliver S, Espigares D 2021: Optimizing labor time for prevention of coccidiosis and anaemia in suckling piglets using an injectable combination of toltrazuril and iron. In: Proceedings of the 12th European Symposium of Porcine Health and Management (ESPHM), Bern, Switzerland.

Hiob L, Holzhausen I, Sperling D, Pagny G, Meppiel L, Isaka N, Daugschies A 2019: Efficacy of an injectable toltrazuril–gleptoferron (Forceris®) to control coccidiosis (*Cystoisospora suis*) in comparison with iron supplemented piglets without anticoccidial treatment. Vet Parasitol **276**: 100002

Joachim A, Ruttkowski B, Sperling D 2018: Detection of *Cystoisospora suis* in faeces of suckling piglets–when and how? A comparison of methods. Porc Health Manag **4**: 1-11

Kennedy JM, Baldwin BA 1972: Taste preferences in pigs for nutritive and non-nutritive sweet solutions. Anim Behav **20**: 706-718

Leslie E, Hernandez M, Newman R, Holyoake P 2010: Assessment of acute pain experienced by piglets from ear tagging, ear notching and intraperitoneal injectable transponders. Appl Anim Behav Sci **127**: 86-95

Lipiński P, Starzynski R, Canonne-Hergaux F, Tudek B, Olinski R, Kowalczyk P, Dziaman T, Thibaudeau O, Gralak M, Snauda E, Wolinski J, Usinska A, Zabielski R 2010: Benefits and risks of iron supplementation in anemic neonatal pigs. Am J Pathol **177**: 1233-1243

Marchant-Forde JN, Lay Jr DC, McMunn KA, Cheng HW, Pajor EA, Marchant-Forde RM 2009: Postnatal piglet husbandry practices and well-being: the effects of alternative techniques delivered separately. J Anim Sci **87**: 1479-1492

Marchant-Forde JN, Lay Jr DC, McMunn KA, Cheng HW, Pajor EA, Marchant-Forde RM 2014: Postnatal piglet husbandry practices and well-being: The effects of alternative techniques delivered in combination. J Anim Sci **92**: 1150-1160

Martínez-Miró S, Tecles F, Ramón M, Escribano D, Hernández F, Madrid J, Orengo J, Martínez-Subiela S, Manteca X, Cerón JJ 2016: Causes, consequences and biomarkers of stress in swine: an update. BMC Vet Res **12**: 171

Noonan G, Rand J, Priest J, Ainscow J, Blackshaw J 1994: Behavioural observations of piglets undergoing tail docking, teeth clipping and ear notching. Appl Anim Behav Sci **39**: 203-213

Rincker MJ, Hill GM, Link JE, Rowntree JE 2004: Effects of dietary iron supplementation on growth performance, hematological status, and whole-body mineral concentrations of nursery pigs. J Anim Sci **82**: 3189-3197

Salmon-Legagneur E, Fevrier R 1956: Les préférences alimentaires du porcelet II. – le sucre dans les aliments pour porcelets. Ann Zootech **5**: 73-79

Sperling D, Freudenschuss B, Shrestha A, Hinney B, Karembe H, Joachim A 2018: Comparative efficacy of two parenteral iron-containing preparations, iron gleptoferron and iron dextran, for the prevention of anaemia in suckling piglets. Vet Rec Open **5**: 00031

Szalai A 1966: Trends in comparative time-budget research. Am Behav Sci **9**: 3-8

Tinti JM, Glaser D, Wanner M, Nofre C 2000: Comparison of gustatory responses to amino acids in pig and in humans. Lebensm Wiss Technol **33**: 578-583

Valenzuela C, Lagos G, Figueroa J, Tadich T 2016: Behavior of suckling pigs supplemented with an encapsulated iron oral formula. J Vet Behav **13**: 6-9

Van Tichelen K, Prims S, Ayuso M, Van Kerschaver C, Vandaele M, Degroote J, Van Cruchten S, Michiels J, Van Ginneken C 2021: Handling associated with drenching does not impact survival and general health of low birth weight piglets. Animals **11**: 404