Behavior, physiology, and performance of bulls mixed at the onset of finishing to form uniform body weight groups

L. Mounier* †2, I. Veissier*, and A. Boissy*

*INRA, URH-ACS, INRA de Theix, 63122 Saint Genes Champanelles, France; and †ENVL, Unité de Zootechnie, 69280 Marcy l’Étoile, France

ABSTRACT: In some production settings, beef bulls are mixed at the beginning of the finishing period to obtain homogeneous groups to facilitate finishing and to produce more homogeneous carcasses. Given the stress induced by mixing, we questioned whether this practice was profitable. Sixty-four young bulls were finished in groups of four over 8 mo. They were either mixed or unmixed and had either homogeneous vs. heterogeneous BW at the beginning of finishing. Behavioral traits (social behavior following mixing and general activities during the finishing period), stress measurements (cortisol concentration, adrenal weight, catecholamine-synthesizing enzyme activity, and pain sensitivity), and production traits (growth rate, homogeneity, feed efficiency, and carcass measurements) were evaluated. Bulls reacted to the mixing with increased frequency of social interactions (agonistic interactions: 17.9 in mixed vs. 1.2 in unmixed bulls, \( P < 0.001 \); sexual interactions: 9.6 vs. 1.7, \( P < 0.001 \); nonagonistic interactions: 5.25 vs. 3.1, \( P < 0.05 \)). Agonistic and sexual interactions were more frequent between bulls of homogeneous BW (\( P < 0.05 \)). Throughout the finishing period, the synchronization of activity tended to be lower between mixed bulls (\( P = 0.09 \)). At slaughter, the activity of tyrosine hydroxylase, a catecholamine-synthesizing enzyme, was increased in mixed bulls (1.52 vs. 1.16 nmol-h\(^{-1}\)-mg\(^{-1}\), \( P < 0.05 \)), which may indicate chronic stress. No differences were noted in ADG or carcass traits among treatments. The variability of BW within groups increased in groups that were initially homogeneous, whereas it decreased in groups that were initially heterogeneous. In conclusion, mixing young beef bulls at the beginning of the finishing period did not have detrimental effects on health and production, but mixing induced stress, perhaps due to decreased cohesion within groups. Thus, we failed to confirm the proposed benefit of mixing bulls to homogenize their BW at the onset of finishing because BW homogeneity within groups was not maintained throughout the finishing period.

Key Words: Behavior, Body Weight, Finishing Bulls, Mixing, Production, Stress


Introduction

In some production settings, beef bulls are mixed at the beginning of the finishing period to establish homogeneous BW groups, with the aim of facilitating finishing and producing homogeneous carcasses.

In cattle, mixing can promote disease and increase aggression (for reviews, see Bouissou et al., 2001; Boe and Faerevik, 2003), induce stress (Hasegawa et al., 1997; Mench et al., 1990), and affect production traits such as growth (Nakanishi et al., 1993) or milk yield (Hasegawa et al., 1997; Sowerby and Polan, 1978). However, these negative effects seem to be limited to a short period following mixing (Brakel and Leis, 1976; Kondo and Hurnik, 1990).

When mixed animals have similar BW, they usually fight more and for longer periods, especially if feed is not distributed evenly in the environment (pigs; Rushen, 1987). Nevertheless, this does not seem to increase the negative effects of mixing on production (Francis et al., 1996; Hindhede et al., 1999).

The aim of this experiment was to evaluate the effect of mixing on the welfare of finishing bulls and to evaluate if mixing helps to produce homogeneous carcasses. Using a \( 2 \times 2 \) factorial design, we compared mixed vs. unmixed animals finished in groups of homogeneous or heterogeneous BW. We evaluated behavioral traits
Mixing finishing bulls according to body weight
1697

(social behavior after mixing, and general activities during the finishing period), stress measurements (cortisol concentration, adrenal weight, catecholamine-synthesizing enzyme activity, and pain sensitivity), and production traits (growth rate, homogeneity of BW, feed efficiency, and carcass measurements). To avoid diseases that could have masked the effects of mixing on stress and behavior, all of the bulls came from the same farm.

Materials and Methods

In accordance with French regulations for experiments on animals, the scientists responsible for the experiment (L. Mounier, I. Veissier, and A. Boissy) were licensed to perform animal experiments, and all people in charge of caring for the animals or taking samples from them received a special course approved by the French Ministry of Agriculture.

Animals and Treatments

Sixty-four Limousin bulls were used in two groups of 32 animals (one batch from 2000 to 2001, and one batch from 2002 to 2003). They were born in mid-winter (January) on the INRA experimental farm at Marcenat (Cantal, France). During winter, until the calves were 4 mo old, they were housed in loose housing and were led to their dams for suckling twice daily. For the next 5 mo, they were left with the cows at pasture, in five groups of 16 male calves (either experimental or not) and their dams. When they were 9 mo old (333 ± 33 kg), experimental calves were weaned (October) and transported for 80 km to the finishing unit of the INRA experimental farm at Theix (Puy de Dôme, France). They did not receive any specific preparation for transport, and no BW loss was observed after transport.

Upon arrival at the finishing unit, the 32 bulls were housed in groups of four animals from the same pasture group. The experimental treatments began 3 wk later using a 2×2 factorial design, with two mixing conditions (mixed vs. unmixed bulls) and two BW conditions (groups made of animals of homogeneous vs. heterogeneous BW based on the BW at weaning). Mixing consisted of making new groups of four bulls, with each bull originating from a different pasture group. Homogeneity of BW corresponded to a CV of BW between bulls of the same group smaller than 2.8%, and heterogeneity to a CV larger than 10%. Over the two replicates, 16 groups of four animals were observed: four groups of unmixed bulls with homogeneous BW, four groups of unmixed bulls with heterogeneous BW, four groups of mixed bulls with homogeneous BW, and four groups of mixed bulls with heterogeneous BW.

During the finishing period, groups were kept in pens separated from each other by solid wooden walls. Each pen was 26 m², including a 16-m² lying area with straw bedding and a 10-m exercise area with a concrete floor, a water bowl, and a trough with four eating places. The animals were fed daily at 0800 with a complete diet that contained corn silage and a concentrate mixture of corn, soybean meal, and urea. Bulls were maintained at the feeding trough for 1 h each morning after the feed distribution. Finishing was conducted in three periods: 1) 3 mo of ad libitum feeding; 2) 2.5 mo of feed restriction to place bulls in a context of feed competition; and 3) ad libitum feeding until slaughter. During the first period of ad libitum feeding, a diet comprising 80% corn silage and 20% concentrate mixture (as-fed basis) was distributed that corresponded to 620 g of PDI (protein truly digestible in the small intestine and 48,800 kJ of NE; INRA, 1989). The quantity of feed provided each day was adjusted so that refusals corresponded to 10% of the amount distributed (expressed in DM). The same diet was distributed during the period of restriction, but the amount was calculated to allow a daily BW gain of 1 kg, which corresponded to 87% of the amount ingested by the bulls in the first period (540 g of PDI and 39,893 kJ of NE). During the last period of ad libitum feeding, a diet of 65% corn silage and 35% concentrate mixture (as-fed basis) was distributed (again adjusted to 10% refusal), allowing a daily BW gain of 1.5 kg (1,040 g of PDI and 69,397 kJ of NE). When the mean BW of bulls within a group reached 650 kg, the four bulls in that group were slaughtered at the INRA slaughterhouse in Theix on the same day. The bulls were 16.5 (±1) mo old at slaughter. This BW corresponds to the normal BW for the late-maturing continental breeds (Charolais and Limousin) slaughtered at 16 to 18 mo of age in mainland Europe (Canali et al., 2001). To stimulate handling during transport and the lairage that occurs under commercial conditions, the bulls were transported for 15 min to a new pen 24 h before slaughter. On the following day, they were led to the slaughterhouse. Inside the slaughterhouse, the animals were separated from each other by solid partitions and were stunned and bled less than 2 h later.

Measurements

Behavior. Four video cameras (SPT-M128CE, Sony Corp., Tokyo, Japan) were fixed above the finishing pens, (i.e., one camera per two pens). Depending on the type of observation, each camera was connected to its own videotape recorder (Sony SVT-1000P), or they were all connected to only one videotape recorder via a quad-ravision system (Computar Quad Processors QS-CX II, Chugai Boyeki [U.K.] Ltd., London, U.K.), which allowed pictures from four cameras to be viewed on one screen. Based on the recorded videos, the behavior of each bull was encoded on a computer with Observer Video Pro software (Noldus, The Netherlands).

For the second replicate of bulls, social behavior was observed by focal sampling for 3 h immediately after the mixing. Social interactions were classified as agonistic interactions, nonagonistic interactions, and sexual interactions (Schein and Fohrman, 1955; Bouissou, 1974;
Bouissou et al., 2001). Agonistic interactions included the following: fighting (animals head against head and pushing each other); butting (violent contact of the head on another animal’s body); threatening (same movement as butting but without contact); and flight (animal turning the head or moving away when another approaches without threat or butt). Nonagonistic interactions included nonsexual sniffing (sniffing another animal’s body but not the anogenital area); nonsexual licking (licking on another animal’s body apart from the anogenital area); head against head (same as a fighting but without pushing); head play (animals rubbing their heads together); head against body (one animal’s head on another animal’s body, without pushing); pushing (same as head against body but with pushing); and small butts (same as butting but not violent and generally repeated). Sexual interactions included sexual sniffing (sniffing the anogenital area of another animal); sexual licking (licking the anogenital area of another animal); head on the back of another animal; mounting; and flehmen response (upper lip reversed). Frequency of any interaction and duration of fights and licks were calculated.

The general activities of the bulls were observed by scan sampling for 24 h at six specific times during the finishing period: from 3 h after mixing, and then after 1, 3, 4, 5, and 6 mo of finishing. Scan duration was 10 s, with 10-min intervals between scans. The general activity states were lying head down (lying down with chin on the floor or on a bull); lying head up (lying down with head up); standing (standing immobile); stepping (taking at least one step); feeding (head in the trough); drinking (nose in the water bowl); and social encounter. These states were mutually exclusive. For each animal, the proportion of scans for which each activity state was observed, and its synchronization (i.e., the proportion of the proportion of scans for which each activity state was observed) were determined by radioimmunoassay with an anti-

Veissier et al., 2000). The bulls were weighed every 2 wk. Once every 4 wk, the bulls were given an ACTH challenge using the method described in Veissier and Le Neindre (1988). Dexamethasone (Dectaneryl, Roussel, Paris, France, 20 μg/kg of BW) was injected i.m. at 1700. At 0800 the following day, ACTH (Synacthène immédiat, Ciba-Geigy, Rueil-Malmaison, France, 1 IU/kg BW) was injected into the jugular vein. Blood samples were collected before the injection of dexamethasone, before the injection of ACTH, and 30, 120, and 180 min after the injection of ACTH. The response to dexamethasone was calculated as the difference between the cortisol concentrations after dexamethasone injection minus that before injection. The integrated response to ACTH was calculated as the area under the curve of cortisol using the following equation:

\[ \frac{n - 1}{2} \sum \left( C_t + C_{t+1} \right) \Delta t \]

where \( n \) is the total number of observations (\( t = 1, \ldots, n \)), \( C \) is the cortisol concentration, and \( \Delta t \) is the interval between successive samples.

Five months after the beginning of the finishing period, the pain sensitivity of bulls was assessed through their reactions to a CO₂ laser beam (MPB Lamsor, Dorval, Quebec, Canada) applied to the skin on the caudal aspect of the metatarsal (Veissier et al., 2000). The bulls were kept at the feeding trough during testing. Six measures were taken between 1400 and 1600, alternating between the left and right legs, with a 30-s pause between consecutive measures. The laser beam was turned off automatically if the animal did not respond within 20 s. In this case, we continued to observe the animal for a further 5 s. Response latency was recorded (maximum = 25 s).

At slaughter, the adrenals were removed from the carcasses. The tissues surrounding the adrenals were thoroughly removed using a scalpel. Then the adrenals were weighed with accuracy to 0.01 g. The adrenals were cut into halves and frozen in liquid N₂ less than 10 min after death. They were stored at −80°C until determination of catecholamine-synthesizing enzymes. Tyrosine hydroxylase (TH) and phenylethanolamine N-methyl transferase (PNMT) activities were determined in the medulla using methods adapted from Waymire et al. (1971) and Axelrod (1962). Briefly, homogenates of the two adrenals of each bull were mixed with a reactional mixture containing [¹⁴C] tyrosine (for TH assay) or S-adenosyl-[¹⁴C] methionine (for PNMT assay). The results are expressed in quantity of product (¹⁴CO₂ for TH and ¹⁴C-methylthanolamine for PNMT) per unit of time per milligram of protein in the tissue. Intra- and interassay CV were 9.26 and 16.13% respectively for TH assay and 9.42 and 16.23% respectively for PNMT assay, against a control having a TH activity of 172.8 nmol h⁻¹mg⁻¹ and a PNMT activity of 8.64 nmol h⁻¹mg⁻¹.

Health and Production. The bulls’ health status was checked once daily during feed distribution. Any clinical symptoms and medical treatments were recorded. The bulls were weighed every 2 wk. Once every 4 wk, the
Table 1. Frequency of social interactions within groups of bulls during the first 3 h following mixing at the beginning of the finishing period; bulls were either mixed or unmixed before the observation, and groups were made of animals of homogeneous or heterogeneous body weight

<table>
<thead>
<tr>
<th>Item</th>
<th>Unmixed</th>
<th>Mixed</th>
<th>SE</th>
<th>P-value (mixing)α</th>
<th>P-value (weight)β</th>
<th>P-value (mixing × weight)γ</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Homogeneous BW</td>
<td>Heterogeneous BW</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>4</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. of groups</td>
<td>4</td>
<td>4</td>
<td>4</td>
<td>4</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Agonistic interactionsd</td>
<td>0.9x 1.5x</td>
<td>22.0x 13.7y</td>
<td>1.4</td>
<td>0.003</td>
<td>0.05</td>
<td>0.03</td>
</tr>
<tr>
<td>Nonagonistic interactionse</td>
<td>2.2 4.0</td>
<td>4.3 6.2</td>
<td>0.7</td>
<td>0.04</td>
<td>0.07</td>
<td>0.98</td>
</tr>
<tr>
<td>Sexual interactionsf</td>
<td>1.9x 3.5x</td>
<td>11.3x 7.9y</td>
<td>0.4</td>
<td>&lt;0.001</td>
<td>0.01</td>
<td>0.02</td>
</tr>
<tr>
<td>Totalg</td>
<td>5.0x 7.0x</td>
<td>37.6x 27.8y</td>
<td>1.6</td>
<td>&lt;0.001</td>
<td>0.07</td>
<td>0.02</td>
</tr>
</tbody>
</table>

α Mixing conditions (unmixed vs. mixed at the beginning of finishing).
β Weight conditions (groups made of bulls of homogeneous vs. heterogeneous weight at the beginning of fattening).
γ Interactions between mixing and weight conditions.
δ Fighting, butting, threatening, and flight.
ε Nonsexual sniffing and licking, head against head, pushing, and small butts.
ζSexual sniffing and licking, head on the bottom or on back of another animal, mounting, and flehmen.
|

Statistical Analyses

Statistical analyses were carried out using the SAS statistical package (Version 8.1, SAS Inst., Inc., Cary, NC). To conform to conditions for ANOVA, frequencies of behaviors (observed during the 3 h following mixing) were log-transformed and proportions of scans on which an activity was observed (24 h observations) were transformed into arcsines. In addition, the homogeneity and normality of residuals were checked.

Each group of bulls was considered as one experimental unit. For all variables, we calculated the mean per groups and the CV within groups. The ANOVA were performed on these means and CV using the following model:

\[ y_{ijkl} = \mu + a_i + b_j + c_k + d_{ij} + e_{ijk} + f_{ijk} + g_{ijk} + e_{ijk} \]

where \( a_i \), \( b_j \), \( c_k \) represent the fixed effects of the mixing conditions (unmixed vs. mixed), the weight conditions (homogeneous vs. heterogeneous) and the batch (1 vs. 2), and \( d_{ij} \), \( e_{ijk} \), \( f_{ijk} \), and \( g_{ijk} \) represent their interactions. Blood cortisol (basal cortisol and ACTH challenge), pain sensitivity, and finishing-related data (daily BW gain, feed intake, feed efficiency) were analyzed as repeated measures.

In the results section, we focus on the effects of mixing and BW conditions, and their interactions. Batch effects are only discussed in cases of interactions with the mixing or the BW conditions. The results are expressed as means ± SE. Statistical significance was set at \( P \leq 0.05 \), with a tendency at \( P \leq 0.10 \).

Results

Behavior

Social Behavior Following Mixing. During the 3 h following mixing, mixed bulls exchanged more interactions (agonistic, nonagonistic, and sexual) than unmixed bulls (Table 1). This was true either for agonistic, non-agonistic, and sexual interactions. More specifically, fights were more frequent and longer in groups of mixed bulls (frequency of fights = 1.8 vs. 0, \( SE = 0.2, P < 0.005 \); duration of fights = 25.8 vs. 0 s, \( SE = 4.6, P < 0.001 \)). The proportion of agonistic interactions compared with interactions as a whole was higher in groups of mixed bulls (53.8 vs. 18.8%, \( SE = 4.8, P < 0.01 \)). The proportion of nonagonistic interactions compared with interactions as a whole was lower in groups of mixed bulls (49.5 vs. 16.9%, \( SE = 2.4, P < 0.001 \)). The frequency of agonistic interactions received by bulls varied more within groups of unmixed bulls (within CV of agonistic interactions = 133.9 vs. 41%, \( SE = 29, P = 0.086 \)). Unmixed bulls spent more time licking each other, both nonsexually and sexually (duration of nonsexual licking = 17.9 vs. 0.6 s, \( SE = 5.9, P < 0.05 \); duration of sexual licking = 16 vs. 0.2 s, \( SE = 5.2, P < 0.005 \)).

Bulls of homogeneous BW exchanged more agonistic and sexual interactions and less nonagonistic interactions than bulls of heterogeneous BW. The proportion of nonagonistic interactions compared with interactions as a whole was lower between bulls of homogeneous BW (39.3 vs. 27.1%, \( SE = 2.4, P < 0.05 \)). Bulls of heterogeneous BW spent more time licking each other, and this concerned only nonsexual licking (17 vs. 1.6 s, \( SE = 5.9, P < 0.05 \)).
We also observed an interaction between mixing and homogeneity of BW, with the effects of mixing on agonistic (P < 0.05) and sexual interactions (P < 0.05) being more marked in homogeneous groups.

**General Activities.** When animals were observed for 24 h at regular intervals during the finishing period, we found no difference between treatments in the time spent in each activity. There was a correlation between the combined effects of mixing condition, BW condition, and batch on the synchronization of activities. For the second replicate of bulls, the synchronization of activities was lower within groups of mixed homogeneous bulls than within groups of either mixed heterogeneous groups or unmixed homogeneous groups (percentage of synchronization = 50.7 vs. 54.7 and 54.3%, P = 0.09).

**Physiology**

One animal did not respond to dexamethasone and another did not respond to ACTH. These animals were excluded from the respective analyses. Data on physiological measures are shown in Table 2.

Basal concentrations of cortisol in plasma, and the responses to dexamethasone and to ACTH did not vary between treatments. The adrenals were heavier in unmixed bulls (P < 0.05) and in bulls of homogeneous BW from the second replicate (14.37 vs. 13.34 g, SE = 0.37, P = 0.085). The TH activity was greater in mixed bulls than in unmixed bulls (P = 0.062), but the PNMT activity and pain sensitivity did not differ between treatments.

**Health and Production**

Only one bull became sick, and it received antibiotics for 2 d. Mean daily BW gains and feed efficiencies for the entire period and for each of the three finishing periods did not differ among treatments (Table 3). Bulls were 497 ± 27 d of age and weighed 621 ± 25 kg at slaughter. Their carcasses weighed 390 ± 17 kg and were most often classified as U2 according to the EUROP scale, with no differences among treatments.

Over the whole finishing period, the CV for BW increased within groups of homogeneous BW (+0.01%/d), whereas it decreased within groups of heterogeneous BW (−0.02%/d, P < 0.001, Figure 1).

**Discussion**

Mixing unfamiliar bulls resulted in clear modifications of behavior immediately following the mixing; groups of mixed bulls exhibited more interactions, particularly agonistic and sexual interactions. This was more evident when bulls were of homogeneous BW. Mixing did not modify the general activities of bulls along the finishing period but the synchronization of activity was higher within groups of unmixed bulls. At slaughter, we found a decrease in the weight of the adrenals and an increase in TH activity in mixed bulls. The heterogeneity of BW within a group decreased during the finishing period in groups that originally were heterogeneous and increased in groups that originally were homogeneous.

Immediately after mixing, the mixed bulls exchanged more interactions than unmixed bulls. They exchanged more agonistic and sexual interactions, and their fights lasted longer, which agrees with previous findings (Kondo and Hurnik, 1990; Mohan Raj et al., 1991). The proportion of agonistic interactions (fights, butts, thrusts, and flights) compared with interactions as a whole was higher in groups of mixed animals. This result is in accordance with the literature on cattle (Kondo et al., 1984) and pigs (Arey and Franklin, 1995; Arey and Edwards, 1998; Jensen and Yngvesson, 1998).

### Table 2. Physiological data in groups of mixed vs. unmixed bulls and of homogeneous vs. heterogeneous body weight at the beginning of the finishing period

<table>
<thead>
<tr>
<th>Item</th>
<th>Unmixed</th>
<th>Mixed</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Homogeneous BW</td>
<td>Heterogeneous BW</td>
</tr>
<tr>
<td>No. of groups</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Blood basal cortisol, ng/mL</td>
<td>2.8</td>
<td>3.5</td>
</tr>
<tr>
<td>Response to dexamethasone, ng/mL</td>
<td>−2.3</td>
<td>−2.8</td>
</tr>
<tr>
<td>Response to ACTH, ng·min⁻¹·mL⁻¹</td>
<td>4,330</td>
<td>4,410</td>
</tr>
<tr>
<td>Adrenal weight, g</td>
<td>15.40</td>
<td>14.57</td>
</tr>
<tr>
<td>TH activity, nmol·h⁻¹·mg⁻¹</td>
<td>1.16</td>
<td>1.17</td>
</tr>
<tr>
<td>PNMT activity, nmol·h⁻¹·mg⁻¹</td>
<td>0.11</td>
<td>0.11</td>
</tr>
<tr>
<td>Latency after laser, s</td>
<td>5.64</td>
<td>5.92</td>
</tr>
</tbody>
</table>

*Values are means ± SE.

**Note:**
- BW: Body weight.
- TH: Tyrosine hydroxylase.
- PNMT: Phenylethanolamine N-methyltransferase.
- Latency: Time to remove the leg from a CO₂ laser beam.
- Differences between cortisol concentrations (after minus before i.m. injection of dexamethasone).
- Area under the curve of cortisol concentrations after the ACTH challenge.
- Sum of right and left glands after removing surrounding tissues.
- Activities of tyrosine hydroxylase (TH) and phenylethanolamine N-methyl transferase (PNMT; catecholamine-synthesizing enzymes) expressed as quantity of substrate metabolized per unit of time and per milligram of protein in the tissue.
- Latency to remove the leg from a CO₂ laser beam.
It corresponds to the fact that right after mixing, cattle generally exchanged numerous aggressive interactions before the dominance hierarchy was established. Unmixed groups did not exhibit agonistic interactions at all, which confirms that the dominance relationships between unmixed bulls were well-established before the start of observations. The variability of agonistic interactions was greater between unmixed bulls than mixed bulls, meaning that in these groups, some bulls had more agonistic interactions than others, whereas in mixed groups, all of the animals had agonistic interactions; this confirms that the hierarchy was better established between unmixed bulls at the beginning of the finishing period. The total of the sexual interactions (sniffing and licking the anogenital area of another animal, head on the back, mounting, and flehmen) and the duration of licking of anogenital parts between animals were all increased with mixing. Mounting is often observed after the mixing of bulls (Tenessen et al., 1985; Kenny and Tarrant, 1987; Mohan Raj et al., 1991). Hinch et al. (1982) suggested that immediately after mixing, mounting plays a role in the establishment of dominance. Aggressiveness would be expressed by agonistic interactions as well as by mountings. In our study, however, the frequency of mounting did not differ between mixed and unmixed bulls. According to Mohan Raj et al. (1991), dominance is established more through agonistic interactions and less through sexual interactions when the production of testosterone is low. Our bulls were probably not already pubescent at the time of mixing, and so aggressiveness might have been expressed essentially by agonistic interactions. One alternative explanation would be that their sexual behavior was not fully developed and that other sexual interactions (sniffing, licking, etc.) were used to establish dominance. The mixed bulls also exchanged more nonagonistic interactions. Such an effect was not reported in earlier studies; however, in our study, the proportion of nonagonistic interactions compared with interactions as a whole was lower between mixed bulls than between unmixed bulls. Hence, the absolute increase of nonagonistic interactions in groups of mixed bulls was probably due to a general increase of all activities following mixing. Moreover, the duration of nonsexual licking between animals was shorter when bulls were mixed. In stable groups of cattle, animals develop affinity relationships, which increase the cohesion of the group and results in a decrease in agonistic interactions and an increase in nonagonistic interactions (Boissy et al., 2001). Thus, affinity between unmixed bulls was probably stronger than between mixed bulls. These results are in agreement with the observations of Veissier et al. (1998), who reported in their review that domestic herbivores establish social bonds early in their life. Preferential associations are established between calves when they are reared together with the dams, especially from 6 mo of age, when calves spend less time with their dam and more time with other calves (Bouissou and Hovels, 1976; Sato and Wood-Gush, 1988). The bonds between calves are strengthened when they are separated from their dam for weaning (Veissier and Le Neindre, 1989; Boissy et al., 2001). In our experiment, the bulls that were not mixed had probably developed strong affinities, especially at weaning. In contrast, mixing bulls 3 wk after weaning (i.e., when the establishment of affinities between animals is no longer facilitated by weaning; Veissier et al., 1989a) seems to result in weak affinities.

The modification of behavior immediately following mixing also depended on the variability of BW within groups. When they were mixed, the bulls of homogeneous BW exchanged more agonistic and sexual interactions but less nonagonistic interactions than bulls of heterogeneous BW. This finding agrees with results

### Table 3. Data of production pooled over all groups of bulls during the finishing perioda

<table>
<thead>
<tr>
<th>Item</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>Overall finishing</th>
</tr>
</thead>
<tbody>
<tr>
<td>ADG, g</td>
<td>1,430 ± 41</td>
<td>929 ± 27</td>
<td>1,640 ± 34</td>
<td>1,280 ± 30</td>
</tr>
<tr>
<td>Feed intake, UFVb</td>
<td>6.4 ± 0.02</td>
<td>5.1 ± 0.02</td>
<td>9.2 ± 0.02</td>
<td>6.6 ± 0.02</td>
</tr>
<tr>
<td>Feed efficiency, g/UFV</td>
<td>224 ± 4</td>
<td>181 ± 4</td>
<td>178 ± 4</td>
<td>193 ± 4</td>
</tr>
</tbody>
</table>

aThe finishing period was conducted in three periods: 1) ad libitum; 2) feed restriction; and 3) ad libitum periods. The bulls had been mixed or unmixed to make groups of homogeneous or heterogeneous BW at the beginning of fattening. There were four groups of four animals each per treatment.  

bUFV = French unit corresponding to 7,626 kJ of NE.

Figure 1. Changes over time in the variability of bull body weight within groups of initial homogeneous (●) vs. heterogeneous (○) BW (means ± SE).
obtained on pigs, where a higher weight difference led to shorter fights and fewer bites (Andersen et al., 2000; Rushen, 1987). For cows, Hindehe et al. (1999) observed more aggressive interactions in heterogeneous groups of heifers than in homogeneous groups; however, they observed groups where the hierarchy was well established, and thus gave no information about the behavior of animals at mixing. The increase in aggressive interactions between animals of homogeneous weight may reflect their greater difficulty in establishing dominance relationships. Sexual interactions also were more frequent in mixed groups of homogeneous weight than in mixed groups of heterogeneous weight. As mentioned before, sexual interactions may be involved in the establishment of dominance relationships. Again, the increased frequency of sexual interactions in homogeneous groups may reflect difficulties in the establishment of the hierarchy, which may in turn explain why homogeneous mixed groups exhibited fewer nonagonistic interactions than heterogeneous mixed groups. In fact, several animal attributes are involved in the establishment of a dominance relationship, including the presence of horns and the weight differences between two animals (Schein and Fohrman, 1955; Bouissou, 1972). If there is not a sufficient interanimal difference between these attributes, then animals may have difficulties in establishing a relationship, as observed in our groups of mixed homogeneous bulls. The practice of mixing animals at the beginning of the finishing period to compose groups of homogeneous weight may therefore be particularly disturbing for animal welfare. In addition, the frequency of agonistic interactions increases when space allowance is limited and when group size increases (Kondo et al., 1989; Boe et al., 2000). Hence, in commercial farms, where groups generally contain more animals than our experimental groups and where density is often higher, the effects of mixing on behavior are probably greater than we observed in this study.

The effects of grouping on behavior seem to be restricted to a short period (1 to 2 wk) in beef bulls (Mench et al., 1990). Indeed, mixing at the beginning of the finishing period had little effect on the long-term behavior of our bulls. We did not notice an alteration in general activities, and we only observed a lower synchronization of activity between mixed homogeneous bulls of the second batch. Affinity between animals is expressed in a better synchronization of their activities (Winfield et al., 1981). Hence, homogeneity of BW in mixed bulls is likely to decrease social cohesion. The difficulty of establishing dominance right after mixing in bulls of homogeneous BW may hinder the future development of affinities.

Aggressive interactions following mixing of animals may cause acute stress (Deguchi and Akuzawa, 1998). The lack of stable social relationships can result in chronic stress, as shown by the higher sensitivity of the hypothalamo-pituitary-adrenocortical axis to ACTH injection or the higher activity of catecholamine-synthe-
sizing enzymes (TH, PNMT) in calves (Dantzer et al., 1983; Veissier et al., 2001), gilts (Janssens et al., 1994), and rats (Mormède et al., 1990). We found no difference in basal concentrations of cortisol or in the cortisol response to ACTH injection between treatments. Nevertheless, TH activity was greater in groups of mixed animals, suggesting a higher degree of chronic stress. The weight of the adrenal glands also can increase due to higher activity in the hypothalamo-pituitary-adrenocortical axis (Lemaire et al., 1993). For instance, in cattle, dominated animals, which are supposed to be submitted to a chronic activity of the hypothalamo-pituitary-adrenocortical axis, have heavier adrenals than dominant animals (Bouissou, 1985). In our experiment, the adrenal glands were heavier in unmixed animals, and heavier in groups of homogeneous BW of the second batch compared with heterogeneous groups. This finding somewhat contradicts the conclusion of a higher chronic stress in mixed bulls as suggested by the TH activity. Nevertheless, Gonyou et al. (1986), found that pigs receiving aversive contacts and which were considered to have undergone chronic stress, did not show a modification in adrenal weight but did show a modification in adrenal morphology, which casts some doubt on the use of adrenal weights as an index of stress. Chronic stress also can increase pain sensitivity (Amit and Galina, 1986; Rushen and Ladewig, 1991; Rushen et al., 1999). In our study, we found no difference in pain sensitivity between unmixed animals and mixed animals or between groups of bulls of homogeneous and heterogeneous BW. As with ACTH challenge, the test was run 5 mo after the beginning of the finishing period, whereas the adrenals were analyzed 2 mo later. Thus, it may be that chronic stress had not developed at the time of the measurement of pain sensitivity and responses to ACTH, but that stress had become more marked at the end of the finishing period. Alternatively, it could be argued that mixing triggers responses of the sympathetic system, with an upregulation of catecholamine-synthesizing enzymes, rather than responses of the hypothalamo-pituitary-adrenocortical axis. The sympathetic system is preferentially stimulated in situations where animals can react actively, whereas the hypothalamo-pituitary-adrenocortical axis is predominantly stimulated when animals cannot control a situation (Dantzer and Mormède, 1979). Our mixed bulls probably exerted some active control on their social environment, as seen through the increased frequency of social interactions.

Regarding the productions traits, we found no difference in growth rates or feed efficiency between treatments. Hence, the supposed benefit for production of mixing young beef bulls at the beginning of the finishing period was not confirmed by our experiment; however, negative effects on production were not observed throughout the finishing period in our study. Earlier reports seem to suggest that the negative effects on growth rates associated with mixing are either short-lived (Gonyou et al., 1988; Stookey and Gonyou, 1994)
or undetected (Friend et al., 1983). For instance, Stookey and Gonyou (1994) found that mixing pigs decreased ADG only during the 2 wk following mixing. Based on our results, mixing young bulls at the beginning of the finishing period seems to have neither positive nor detrimental effects on production over the finishing period. However, mixing may have negative effects on growth when it is coupled with another stressor factor, such as limited feeding or decreased space (Sherritt et al., 1974), which was not the case in our experiment, where bulls were fed ad libitum with unrestricted access to the trough (Hyun et al., 1998).

We observed no health differences between treatments; however, all of our bulls came from the same farm and therefore presumably did not have different microbial backgrounds. In commercial conditions, mixing involves animals from different farms, and thereby with different microbial backgrounds, and pathologies often develop after mixing. Moreover, stress may increase the development of pathologies by decreasing immunity (Moberg, 1987). To study the effect of mixing on pathologies, we plan to run a survey on commercial farms in reference to animal behavior.

Another objective of mixing bulls according to BW at the beginning of the finishing period is to produce homogeneous carcasses. In our experiment, the evolution of the variability of BW within groups differed between treatments, and it increased in groups that were initially homogeneous and decreased in groups that were initially heterogeneous. Hence, bulls that were heterogeneous became more homogeneous, whereas bulls of homogeneous BW tended to heterogenize. The fact that the hierarchy is more easily established in groups of homogeneous BW may help animals to obtain access to the trough and to eat, as seen through the better synchronization of activities. This may explain why these animals, which had similar genetic potentials for growth, ended finishing with similar BW. In contrast, in homogeneous groups, where aggressions were more frequent, some animals may have had more difficulty accessing the trough, and this may have increased the variability of BW at the end of the finishing period, which is confirmed by the fact that the synchronization of activities was lower in groups made homogeneous at the beginning of finishing compared with heterogeneous ones. Nevertheless, the carcasses obtained from all bulls were within the normal range for Europe (400 kg and U2 on the EUROPE scale).

In conclusion, mixing young bulls resulted in an increase of agonistic interactions immediately following mixing, especially when the bulls were of similar BW, which was related to the establishment of dominance relationships. During the finishing period, whatever the differences in BW of bulls from a group, average production of a group and differences in carcass weights tended to be similar. Although mixing seems to have no detrimental effects on production, at least when animals have the same origin, it can lead to moderate chronic stress. Hence, the present results suggest that mixing young beef bulls according to the BW at the beginning of the finishing period does not seem beneficial, as it does not improve production and may be detrimental for animal welfare. Moreover, homogeneity may not be maintained during the finishing period.

### Literature Cited


