

# Assessment of milk yield in hair sheep ewes: a comparative study of two methods

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Maritza Zaragoza-Vera<sup>1</sup>

[orcid.org/0000-0003-0453-8660](https://orcid.org/0000-0003-0453-8660)

Erica Preciado-Lopez<sup>1</sup>

Roberto Carlos Barrientos-Medina<sup>2</sup>

[orcid.org/0000-0003-2144-034X](https://orcid.org/0000-0003-2144-034X)

Claudia Virginia Zaragoza-Vera<sup>1</sup>

[orcid.org/0000-0002-8629-7401](https://orcid.org/0000-0002-8629-7401)

Guadalupe Arjona-Jimenez<sup>1</sup>

[orcid.org/0000-0003-4625-5650](https://orcid.org/0000-0003-4625-5650)

Oswaldo Torres-Chable<sup>1</sup>

[orcid.org/0000-0001-7482-6663](https://orcid.org/0000-0001-7482-6663)

Ricardo Alfonso Garcia-Herrera<sup>1\*</sup>

[orcid.org/0000-0003-2456-4727](https://orcid.org/0000-0003-2456-4727)

Alfonso Juventino Chay-Canul<sup>1\*</sup>

[orcid.org/0000-0003-4412-4972](https://orcid.org/0000-0003-4412-4972)

<sup>1</sup> Universidad Juárez Autónoma de Tabasco. División Académica de Ciencias Agropecuarias. Villahermosa, Tabasco, México.

<sup>2</sup> Universidad Autónoma de Yucatán. Facultad de Medicina Veterinaria y Zootecnia. Mérida, Yucatán. México

**\*Corresponding authors:**

alfonso.chay@ujat.mx,  
ricardo.garcia@ujat.mx

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## **Assessment of milk yield in hair sheep ewes: a comparative study of two methods**

### **Abstract**

This study compared two methods for estimating milk yield (MY) in multiparous Pelibuey and Katahdin ewes accounting for litter size and week of lactation under tropical conditions. MY was assessed using the weight-suckle-weight (WSW) technique and hand milking (HM). Exploratory analysis was performed using box plots, followed by a repeated-measures ANOVA based on a mixed linear model including method, breed, litter size, and week as fixed effects, and ewe as a random effect. Method agreement was evaluated using Bland–Altman analysis. MY increased during the first three weeks of lactation, then stabilized or slightly declined. HM consistently produced higher average values than WSW. Katahdin ewes and those rearing twin litters yielded more milk than Pelibuey and single-litter counterparts. The mixed model showed that estimation method and litter size were the main sources of variation, followed by breed and week. The intraclass correlation coefficient (ICC = 28.11 %) indicated that slightly more than one-quarter of total variability was attributable to individual differences. The means  $\pm$  SE plot suggested an interaction, with the largest HM–WSW discrepancy observed in Katahdin twin-litter ewes, the highest-producing group. The Bland–Altman plot revealed a systematic bias (-0.78 kg; HM > WSW) and poor agreement (limits of agreement -2.26 to 0.66 kg), indicating potential individual-level discrepancies. HM tends to overestimate MY relative to WSW, particularly in highly productive ewes. The two methods are not interchangeable without adjustment; their selection should consider ewe characteristics and study objectives, and correction for systematic bias is recommended when comparing results.

**Keywords:** Milk yield estimation; Hair sheep; Tropical production systems; Lactation performance.

### **Study contribution**

This study provides a comprehensive evaluation of two widely used methods for estimating milk yield in multiparous Pelibuey and Katahdin ewes under tropical conditions, accounting for litter size, breed, and lactation stage. By combining exploratory data analysis, mixed-model ANOVA, and Bland–Altman agreement assessment, the research highlights systematic differences between the weight-suckle-weight (WSW) technique and hand milking (HM), demonstrating that HM consistently overestimates milk yield, particularly in highly productive ewes. The findings offer practical guidance for selecting appropriate milk yield estimation methods, emphasize the importance of adjusting for systematic bias, and provide a quantitative basis for improving accuracy in nutritional, genetic, and management studies in tropical small ruminant systems.

### **Introduction**

Pelibuey and Katahdin are two of the main hair sheep breeds currently used across different agroecological regions of Mexico, either as purebreds or crossbreds.<sup>(1, 2)</sup> Among them, Pelibuey is the most prevalent due to its adaptability to diverse environmental conditions. Desirable traits of this breed include rusticity, year-round reproduction, prolificacy, and parasite tolerance.<sup>(1, 3)</sup> In contrast, Katahdin was introduced from the United States in the early 2000 with the aim of improving the productivity of local hair sheep breeds such as Pelibuey and Blackbelly.<sup>(1)</sup> The most widely used methods for

estimating milk production (MP) in sheep include manual milking, mechanical milking, the weight-suckle-weight (WSW) technique, and the measurement of udder dimensions.<sup>(4, 5)</sup>

However, direct measurement of MP is challenging in non-dairy breeds such as hair sheep, as their small teats often result in milk retention during milking.<sup>(6)</sup> For this reason, indirect methods are commonly employed to estimate MP.<sup>(7)</sup> The WSW technique estimates milk intake in lambs. In practice, lambs are separated from their dams for approximately 3–4 hours, weighed, and then allowed to suckle for a short period (10–15 minutes). The WSW method requires repeated separation and weighing of lambs every few hours, which is labour-intensive and may be impractical for large flocks. Frequent separation can also induce stress in both lambs and ewes, potentially affecting lamb behaviour and milk letdown, and introducing bias. After suckling, lambs are weighed again, and the difference between pre- and post-suckling weights represents milk consumption during that interval.<sup>(1, 4, 8)</sup>

Daily milk yield is then estimated by summing the values from successive measurements and extrapolating to 24 hours. This approach has been applied in cows, sheep, and goats.<sup>(4, 8)</sup> Given the limited information available and the variability in reported results, it is necessary to evaluate simple and reliable methods for estimating milk production in hair sheep that minimize animal handling. Therefore, the objective of this study was to compare two methods —the WSW technique and hand milking (HM)— for estimating milk yield in multiparous Pelibuey and Katahdin ewes.

## **Materials and methods**

### *Ethical statement and study site*

The trial was conducted in compliance with the animal welfare guidelines of the División Académica de Ciencias Agropecuarias, Universidad Juárez Autónoma de Tabasco (ID project PFI: UJAT-DACA-2015-IA-02). The study was performed at *El Rodeo* ranch (17°84' N, 92°81' W; Tabasco, Mexico), under humid tropical conditions, with a mean temperature of 28 °C, average rainfall of 63 mm, and relative humidity ranging from 54 % to 95 percent.<sup>(9)</sup>

### *Animals*

Forty-nine multiparous ewes (Pelibuey, n = 28; Katahdin, n = 21), clinically healthy and 3–5 days postpartum, were included. Average body weight and body condition score (BCS; scale 1–5)<sup>(10)</sup> were 47.5 ±1.3 kg and 2.6 ±0.1 for Pelibuey, and 43.9 ±1.5 kg and 1.7 ±0.1 for Katahdin, respectively. The lamb population comprised 39 Pelibuey and 28 Katahdin, with data collected on litter size, sex, and weekly body weights until 56 days of age. No mortality was recorded during the experimental period.

### *Management*

Ewes and their lambs were housed in individual pens for 56 days under a semi-intensive feedlot system. Ewes had free access to water and were dewormed with Cydectin NF® (Pfizer, Brazil; 0.2 mg/kg BW). The diet consisted of star grass hay (*Cynodon nlemfuensis*), ground maize, soybean meal, molasses, and minerals, formulated to supply 12 MJ/kg of metabolizable energy per kg of dry matter and 15 % crude protein.<sup>(11)</sup> Feed

allowance was adjusted weekly according to ewe body weight to meet lactation requirements.

### *Milk production estimation*

MP was estimated using the WSW technique.<sup>(1, 4)</sup> Lambs were separated from their dams at 03:00 a.m. for 3 h and then allowed to suckle for 10 min at 06:00 a.m. Following a second 3 h separation (until 09:00 a.m.), lambs were weighed immediately before and after suckling, and the weight difference was considered milk intake, representing 3 h of milk production. Lamb weights were recorded using a digital scale (Torrey MFQ-20, EQB, Torrey®, Mexico) with a precision of  $\pm 2$  g. This procedure was repeated at 06:00, 09:00, 12:00, 15:00, and 18:00 h, covering a 12 h-period. Daily MP was calculated as the sum of all measurements, extrapolated to a 24-h period.

For manual milking, lambs were separated at 06:00 a.m. and allowed to suckle at 09:00 a.m. for 10 min. At 12:00 m., each ewe received an intramuscular injection of 3 IU of synthetic oxytocin (Pisa®, Mexico), and milking commenced approximately a minute after injection to ensure adequate milk letdown. Manual milking was performed by the same trained operator using a standardized technique, applying gentle and continuous pressure until milk flow ceased. Milking was considered complete when no further milk could be obtained after three consecutive stripping attempts, combined with visual confirmation of udder emptying. MY recorded over the 3-h period was extrapolated to a 24-h basis to estimate daily MY.<sup>(4-7)</sup>

### *Data analysis*

First, variability in milk production across the experimental period was explored by estimation method, considering breed and litter size of the experimental subjects, and visualized using box-and-whisker plots. Prior to inferential analyses, model assumptions were evaluated by inspecting the normality of residuals using Q–Q plots and the Shapiro–Wilk test, as well as homoscedasticity through residuals versus fitted values plots. Based on this preliminary analysis, a repeated-measures analysis of variance was performed using a mixed linear model. In this model, breed, litter size, week, and estimation method were included as fixed effects, while individual ewes were considered as a random effect. The magnitude of between-subject variability was assessed using the intraclass correlation coefficient (ICC), and results were presented as weekly means  $\pm$  standard errors by litter size and breed using error plots.

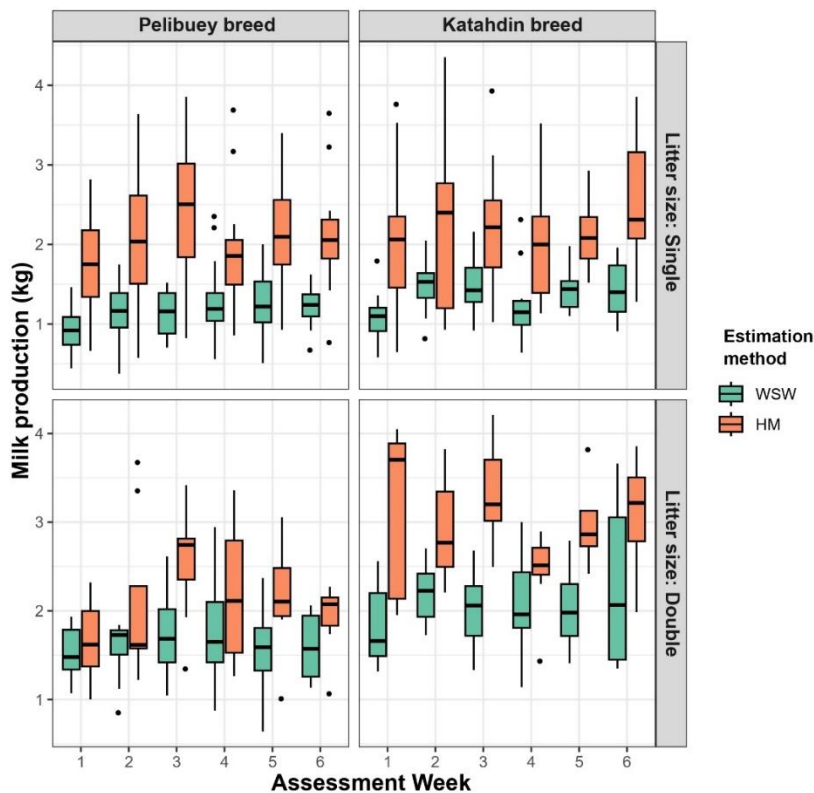
To quantify the uncertainty associated with the ICC, we obtained a 95 % confidence interval using a parametric bootstrap procedure implemented via the `bootMer` function in the *lme4* package. The bootstrap was performed with 5 000 resamples using the fitted mixed-effects model as the data-generating mechanism, which provides a distribution-free interval estimate consistent with the repeated-measures structure of the design. To evaluate the agreement between estimation methods, Bland–Altman analysis was applied. This approach involved plotting the mean of paired measurements on the X-axis against the difference between methods on the Y-axis, thereby assessing both bias and limits of agreement. All analyses and graphs were conducted in the *R* programming environment,<sup>(12)</sup> using the following packages: *BlandAltmanLeh*,<sup>(13)</sup> *dplyr*,<sup>(14)</sup> *ggplot2*,<sup>(15)</sup> *ggpubr*,<sup>(16)</sup> *lme4*,<sup>(17)</sup> *lmerTest*,<sup>(18)</sup> and *tidyr*.<sup>(19)</sup>

## Results

Exploratory analysis using box plots revealed clear patterns in milk production (**Figure 1**). Production increased during the first three weeks of evaluation, after which it stabilized or showed a slight decline. The milk production estimate using the HM method consistently yielded higher average values than the WSW method. In addition, Katahdin ewes and those rearing twin litters had greater average production than Pelibuey ewes and those with single litters, respectively. The results of the repeated-measures ANOVA showed that estimation method and litter size were the main sources of variation in milk production, followed by breed and week (**Table 1**). The ICC for the random intercept of ewe was 0.281, indicating that slightly more than one-quarter of the total variability was attributable to between-ewe differences. The 95 % confidence interval obtained via parametric bootstrap ( $B = 5\ 000$ ) ranged from 0.162 to 0.293, reflecting moderate between-subject variability and supporting the stability of the ICC estimate within the mixed-model framework.

**Table 1.** Mixed linear model results from the repeated-measures ANOVA of milk production in hair ewes

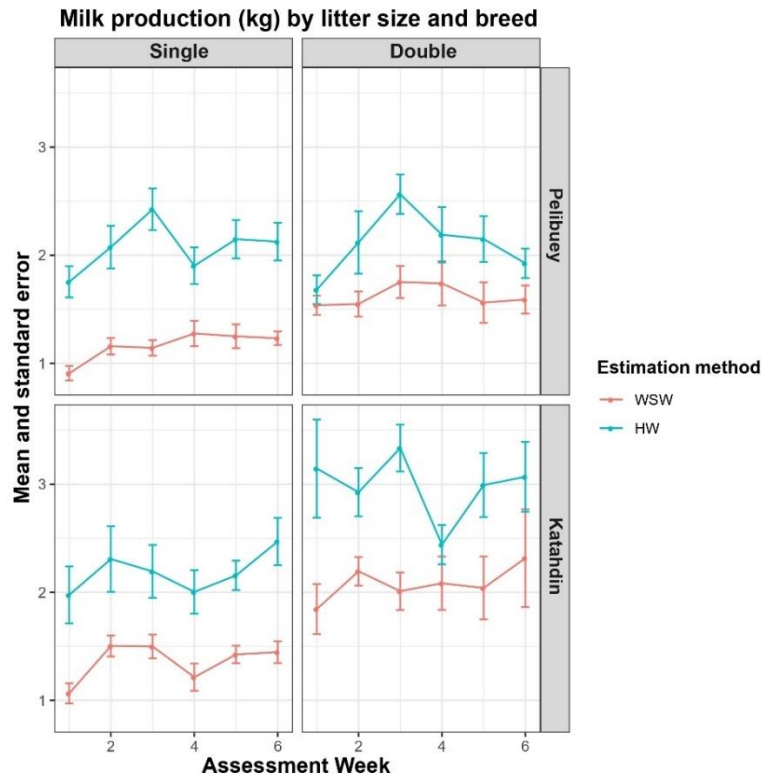
| Fixed effects     | Mean square | Degrees of freedom | F       | Significance    |
|-------------------|-------------|--------------------|---------|-----------------|
| Weeks             | 1.9109      | 1 480.03           | 6.8487  | P < 0.01        |
| Estimation method | 17.5492     | 1 473.27           | 62.8983 | P < 0.001       |
| Breed             | 2.9889      | 1 45.36            | 10.7126 | P < 0.01        |
| Litter size       | 4.6721      | 1 45.37            | 16.7453 | P < 0.001       |
| Weeks:Method      | 0.0187      | 1 473.27           | 0.0671  | not significant |



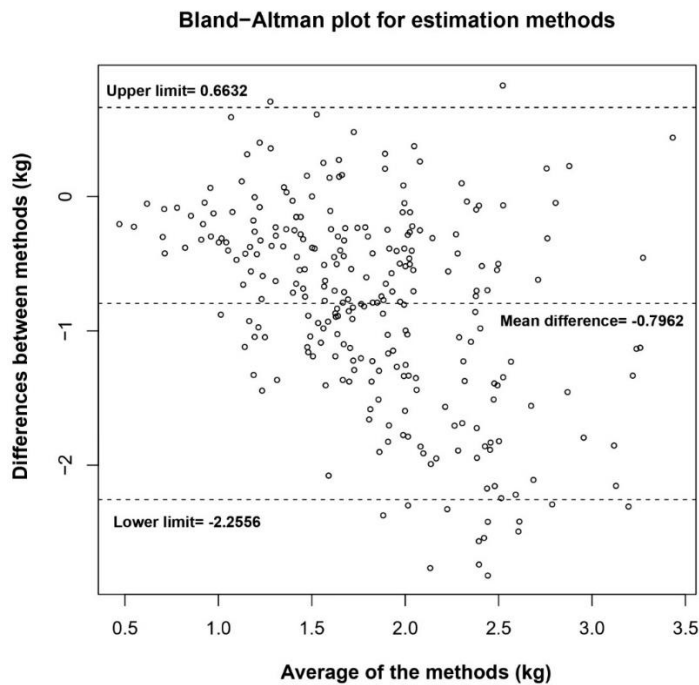
**Figure 1.** Box plots of weekly milk production (kg) by estimation method, breed, and litter size.

The graph of means and standard errors confirmed the lactation curve pattern: an increase during the first weeks, a peak at mid-lactation, and a subsequent decrease or stabilization (**Figure 2**). The plot also suggested interactions among factors. The difference between estimation methods was most evident in Katahdin ewes with twin litters, the group with the highest production, suggesting that the bias between methods may be more pronounced in animals with greater productive performance. The Bland–Altman plot (**Figure 3**) confirmed these patterns. The mean difference of -0.78 kg indicated a systematic bias that should be considered when comparing results.

The limits of agreement (-2.26, 0.66) reflected poor concordance, as estimates from the two methods could differ by up to 2.92 kg for the same ewe. Although no clear fan-shaped distribution was observed, indicating that the magnitude of the differences remained relatively consistent across the range of measurements, extreme values below the lower limit reinforced the presence of systematic bias, highlighting variability at the individual level.



**Figure 2.** Weekly average milk production  $\pm$  standard-error by estimation method, litter size, and breed.



**Figure 3.** Bland-Altman plot showing the differences in milk production between estimation methods.

## Discussion

There are differences in the milk production profiles of wool and hair sheep breeds. In non-dairy breeds such as Katahdin and Saint Croix, which are primarily used for meat production, evaluating the ewes' milk yield is important because it is closely related to lamb growth and survival.<sup>(20–22)</sup> Despite its importance, information on milk production and quality in hair sheep during lactation is limited, with only a few studies available for breeds such as Saint Croix, Blackbelly, Pelibuey, and Katahdin.<sup>(23, 24)</sup> In general, ewes from tropical breeds produce less milk than those from temperate breeds, consistent with the milk yields observed in tropical sheep breeds. Nevertheless, milk and lambs remain an important source of protein for people living in tropical regions.<sup>(21, 22, 25)</sup>

In hair sheep, measuring MY through milking is challenging because of the small size of the teats. Although milk production has been estimated using common non-dairy methods, such as WSW or HM after oxytocin administration,<sup>(24)</sup> it is necessary to evaluate potential differences between these approaches to identify the most accurate and reliable indirect methods. The results demonstrated that the HM after oxytocin administration method consistently overestimates milk yield in multiparous hair sheep compared with the WSW method. These findings contrast with those reported by Peniche-González et al.,<sup>(4)</sup> who observed no differences between the WSW method and its combination with HM, as confirmed by orthogonal regression analysis.

This discrepancy may be explained by the relatively short suckling period permitted by the WSW protocol, which may prevent lambs from completely emptying the udder after separation. In contrast, oxytocin administration during HM likely promoted complete milk letdown. When combined with thorough manual extraction, this resulted in higher milk yield estimates. Conversely, Van der Linden et al.<sup>(5)</sup> found that the WSW method often underestimates milk yield, particularly in well-fed ewes with high production potential. In the present study, the ewes were fed diets that fully met their nutritional requirements. The higher milk yields recorded with HM suggest that the ewes' production capacity was elevated, meaning it is unlikely that the lambs could have consumed all the available milk using the WSW method alone.<sup>(26, 27)</sup>

The differences observed between WSW and HM are consistent with previous studies on both dairy and meat-type ewes,<sup>(26, 27)</sup> and are likely due to incomplete udder emptying with WSW, stress-induced inhibition of milk ejection, or potential overestimation by HM due to residual milk letdown induced by oxytocin.<sup>(27)</sup> Despite these differences, the strong correlations and relatively narrow limits of agreement between the two methods suggest that both are suitable for ranking animals by production potential; however, absolute values should not be used interchangeably. Both the boxplot and error graphs revealed a consistent lactation curve: an initial rise in milk production, a peak at mid-lactation, and a gradual decline toward the end of the study.

The error plot, together with the mixed linear model results, confirmed that HM consistently produced higher estimates than WSW, indicating a systematic bias. Moreover, Katahdin ewes exhibited higher average milk yields than Pelibuey ewes, and ewes nursing twins produced significantly more milk than those nursing singletons,

reflecting both genetic and litter-size effects. The Bland–Altman plot further confirmed the presence of systematic bias, and the identification of outliers highlights the need for careful selection of estimation methods and interpretation of results within the context of biological and methodological variability.

From a practical perspective, WSW remains a less invasive, field-friendly technique that minimizes handling stress and equipment requirements, making it well-suited for large-scale or extensive systems. Conversely, HM is more labour-intensive but yields more precise estimates of milk production, making it preferable for research applications that require accurate quantification, such as nutritional trials or genetic selection programs. Our findings are particularly relevant for tropical production systems where Pelibuey and Katahdin ewes are used for dual-purpose meat and milk production. Reliable and feasible estimation methods are crucial for identifying highly productive females, supporting genetic improvement programs, and enhancing the efficiency of production systems.

These results emphasize the importance of evaluating the degree of agreement between methods rather than assuming their equivalence. Future research should aim to integrate complementary techniques, such as mammary ultrasound, digital image analysis, or automated suckling sensors, to improve accuracy and minimize the impact of management on animal welfare.

## **Conclusions**

This study demonstrates that milk yield in hair ewes is influenced by breed, litter size, and stage of lactation, with Katahdin ewes and those rearing twin litters showing the highest production. The hand-milking method consistently produced higher estimates than the weight-suckle-weight technique, revealing a systematic bias that was more evident in high-yielding animals. The Bland-Altman analysis confirmed poor agreement between methods, highlighting that differences of up to 2.92 kg can occur for the same ewe. These findings emphasize the need for caution when selecting and interpreting estimation methods and underscore the importance of using consistent approaches for evaluating milk production in non-dairy sheep breeds under both experimental and production conditions.

### **Data availability**

Data are available with the corresponding author of this publication upon reasonable request.

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### **Conflicts of interest**

The authors have no conflict of interest to declare regarding this publication.

### **Author contributions**

Conceptualization: AJ Chay-Canul, RA Garcia-Herrera, OM Torres-Chable.

Data curation and formal analysis: AJ Chay-Canul, RC Barrientos-Medina.

Investigation: E Preciado-Lopez, M Zaragoza-Vera, CV Zaragoza-Vera.

Methodology: E Preciado-Lopez, M Zaragoza-Vera, AJ Chay-Canul, RA Garcia-Herrera.

Project administration: CV Zaragoza-Vera, G Arjona-Jimenez.

Resources: CV Zaragoza-Vera, G Arjona-Jimenez, RA Garcia-Herrera.

Software: M Zaragoza-Vera, AJ Chay-Canul, RC Barrientos-Medina.

Supervision: M Zaragoza-Vera, CV Zaragoza-Vera.

Writing: M Zaragoza-Vera, AJ Chay-Canul, RC Barrientos-Medina.

Writing-review and editing: M Zaragoza-Vera, CV Zaragoza-Vera, G Arjona-Jimenez, RA Garcia-Herrera, OM Torres-Chable, AJ Chay-Canul, RC Barrientos-Medina.

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