

However, Single-Step EBVs cannot be used as input data for MACE, given that they include a genomic component whereas MACE is a strictly polygenic evaluation. This is why a solution had to be devised to send genomic-free EBVs based on the national Single-Step evaluations.

An Interbull working group proposed several methods to remove the genomic part from Single-Step EBVs and, hence, to obtain genomic-free Single-Step (GFSS) EBVs (Sullivan, 2021). One of the recommended methods is to run a pedigree based BLUP evaluation using pre-adjusted performances obtained from Single-Step evaluations as phenotypes. In our case, it was the most promising scientifically speaking and it represented a reasonable amount of work to implement and to maintain routinely.

The use of GFSS EBVs instead of polygenic EBVs for MACE was the last challenge of the transition from polygenic and genomic evaluations to Single-Step evaluations for the French national evaluations.

The objective of this paper is to present the first French experience of participation in MACE with GFSS EBVs. This was achieved by 1) developing a new pipeline to compute the GFSS EBVs, 2) validating estimated genetic trends using Interbull Trend Tests that are necessary to any participation to MACE, and 3) analyzing the impact of the transition from French national polygenic to GFSS EBVs in MACE on the MACE results.

Materials and Methods

Computation of genomic-free Single-Step EBVs

We performed the same work for all breed-trait combinations involved in MACE, *i.e.* milk production, female fertility, longevity, calving, conformation, workability and udder health traits in 4 breeds (Brown Swiss, Simmental, Montbéliarde and Holstein).

Among the methods proposed by the Interbull working group to compute GFSS EBVs, the most appropriate was to run a BLUP evaluation using Single-Step YDs as phenotypes.

The current Single-Step French evaluation incorporates foreign Holstein and Brown-Swiss bulls using MACE EBVs, with the objective of enhancing the reference evaluation. For this purpose, pseudo phenotypes of foreign bulls' progeny are assessed thanks to deregressed MACE EBVs after the removal of French information. Only pseudo phenotypes of bulls with few domestic daughters with performances (in Holstein: less than 600 daughters) are used in the French routine Single Step.

The Single-Step evaluation produces YDs for all animals. In order to avoid double-counting, Single-Step YDs for foreign daughters are excluded from the BLUP evaluation. We consider YDs for domestic daughters to include only a negligible amount of foreign information.

Computation of genomic-free Single-Step EBVs for milk production, longevity, udder health, female fertility, conformation and workability traits

For all traits except calving traits, Single-Step YDs were calculated as such:

- performances were adjusted for all non-genetic effects estimated from the routine Single-Step evaluation. If a given animal has only one phenotype, its adjusted performance is its YD
- In the genetic evaluations with repeatable traits, the YD of each animal was the weighted average of adjusted performances

More details about the non-genetic effects included in Single-Step models for each trait can be found on GenEval's website (2024).

An adjusted weight for each Single-Step YD was also calculated, as the sum of the adjusted weights of the performances considered in the YD.

The pedigree based BLUP model used to estimate GFSS EBVs was:

$$y_i = by_i + a_i + \varepsilon_i \quad (1)$$

where y_i is the Single-Step YD for animal i , by_i is the fixed effect of the birth year of animal i , a_i is the additive genetic effect for animal and $\varepsilon_i \sim N(0, \text{var}/w_i)$ is the residual, with w_i the weight of y_i .

Computation of genomic-free Single-Step EBVs for calving traits

Single-Step YDs for calving traits were computed correcting performances for all effects included in the Single-Step model, except for the genetic animal and maternal effects.

For these traits, the BLUP model used to estimate GFSS EBVs was as (1), with the addition of the maternal genetic effect of the dam j of animal i (m_j).

$$y_i = by_i + a_i + m_j + \varepsilon_i \quad (2)$$

Models (1) and (2) are univariate models, as are all Single-Step evaluation models in France for these traits.

Validation of genetic trends

Interbull trend validation procedures were used in order to validate GFSS EBVs so that they could then be provided as proofs for routine MACE runs.

In the present study, Trend Test method II was chosen as reference method for most of traits (Boichard et al., 1995), as it is the most stringent of the Trend Test methods. Few traits could not be validated using Trend Test method II. In this case, they were validated with Trend Test method III.

Investigation of the impact of transition from polygenic EBVs to genomic-free Single-Step EBVs on MACE EBVs of Holstein bulls

The aim of this study was to analyze the impact of the transition from French polygenic to GFSS EBVs on the MACE EBVs of bulls, on the Holstein Breed only, so that we could

have an idea of the consequences of this change on the national evaluation.

For this, we compared the output of MACE evaluations, depending on whether polygenic information or GFSS was used. We also looked at them in comparison to the routine Single-Step evaluation and the GFSS in order to compare genetic gain between these four evaluations.

Overall, this work was based on 4 categories of EBVs of Holstein bulls:

- French Single-Step EBV (SSEBV) published by the French breed societies in August 2023;
- French genomic-free Single-Step EBVs (GFSSEBV) sent to Interbull for the September 2023 test-run;
- MACE EBVs (MACE EBV) based on polygenic French proofs and expressed in the French scale. These EBVs were performed by Interbull during the routine evaluation of August 2023;
- MACE EBVs expressed in the French scale (SSMACE EBV) and based on French genomic-free Single-Step EBVs. These EBVs were performed by Interbull during the test run of September 2023.

We considered six traits, in order to focus on a large panel of heritabilities: production traits (milk, protein and fat yield), somatic cell score, mastitis and heifer conception rate.

In some parts of this study, two sub-populations were considered: bulls with only domestic daughter information taken into account in the routine evaluation (FR) and bulls with foreign information included in the evaluation (FOR). As mentioned before, the FOR bulls have less than 600 domestic daughters (as only French performances are used for bulls with more domestic progeny).

Several statistical analyses were conducted: genetic trends estimated with the four categories of EBVs, correlations and regressions of SSMACE on MACE EBVs for FOR animals, and reranking between MACE

and SSMACE, comparing the top100 bulls between the two rankings.

The changes in genetic correlations between France and the other countries estimated by Interbull were also analyzed, but they are not reported in this article.

Table 1: number of bulls considered in the impact study.

Trait	FR	FOR
Milk yield	16495	2532
Protein yield	16883	2144
Fat yield	16883	2144
Somatic cell score	16140	2861
Mastitis	8677	2714
Heifer conception rate	13483	2127

Results & Discussion

Validation of genetic trends

All traits, except cow conception, cow interval and locomotion in Holstein and longevity in Brown Swiss, successfully passed the genetic trend validations using Trend Test method II. For the four aforementioned traits, genetic trend was successfully validated by means of Trend Test method III.

The validation of the estimated genetic trend in all populations and traits involved in the international evaluations represented the last step of an extensive validation process conducted in France for more than 5 years and involving research partners, GenEval and the users (AI industry and breed societies). This enabled the use of French GFSS EBVs in MACE since Decembre 2023. As a result, all French domestic proofs sent to Interbull for routine international evaluations are now derived from Single-Step evaluations, as domestic GEBVs calculated with the Single-Step methodology have been included in GMACE since 2022.

Impact of the transition from polygenic EBVs to genomic-free Single-Step EBVs on MACE results

Genetic gain estimated with bulls in FOR for milk yield was lower with MACE EBV than with the SSEBV. Genomic-free evaluations (SSMACE and GFSS) had an intermediate genetic trend. These results indicate that the correction for genomic preselection in the genomic-free Single-Step evaluation is only

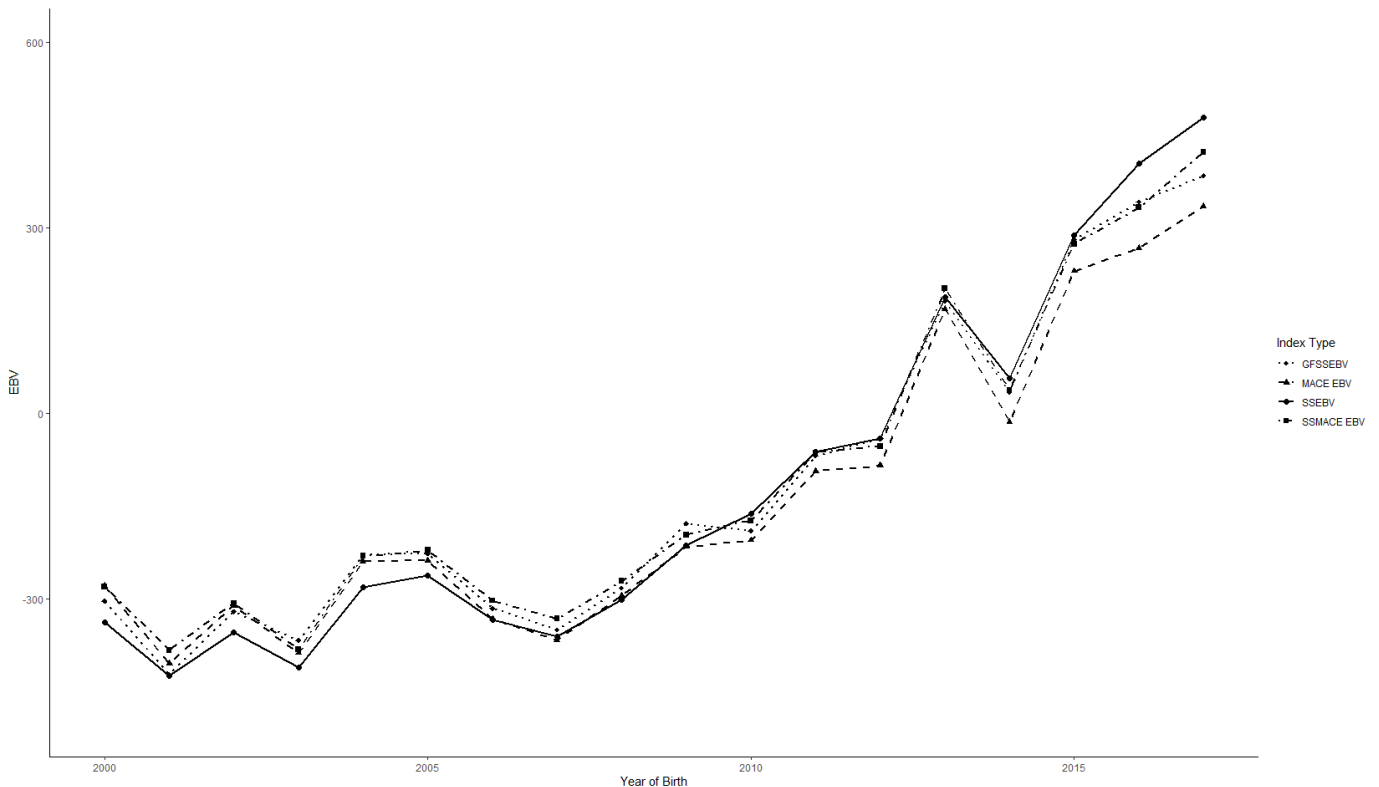


Figure 1. Genetic trend for milk production in the FOR population

Table 2. Correlation between SSMACE EBVs and MACE EBVs and slope of the regression of SSMACE EBVs on MACE EBVs, in the FOR population.

Trait	Correlation	Slope
Milk yield	0.994	0.963
Protein yield	0.996	0.951
Fat yield	0.996	0.979
Somatic cell score	0.994	1.025
Mastitis	0.996	0.997
Heifer conception rate	0.971	0.913

partial, which is consistent with the conclusions of the Interbull working group dedicated to this topic (Sullivan, 2021). The same pattern was observed for protein and fat yield. For heifer conception rate and mastitis, the genetic trends for all evaluations are much lower, and more stable between evaluations, with no type of evaluation showing a clearly higher or lower genetic trend.

The differences between MACE and SSMACE EBVs of bulls were limited (Table 2), with regression slopes between 0.951 and 1.025 and correlations above 0.994 for all traits except for heifer conception rate (above 0.97). This lower correlation might be due to the very low heritability of heifer conception rate (0.019) and the subsequent low reliability of EBVs. EBVs on this trait are more susceptible to vary due to a change in the evaluation approach and are the ones for which the removal of genomic information might be the most detrimental.

Reranking was mostly moderate: for all traits except heifer conception rate, more than 80% of the bulls in the top 100 bulls are common between ranking based on MACE or SSMACE EBVs (Table 3). Once again, we observed higher reranking for lower heritability traits, and especially for heifer conception rate.

All these results indicate that some variations are expected between former MACE EBVs and new SSMACE EBVs, but that the magnitude of these changes is limited.

Table 3. Reranking in the top100 bulls when ranking is based on MACE vs SSMACE EBVs.

Trait	Common ¹	In ²	Out ³
Milk yield	91	/	/
Protein yield	90	0	1
Fat yield	95	/	/
Somatic cell score	80	7	1
Mastitis	87	1	1
Heifer conception rate	77	8	2

¹: Number of bulls common in the two top100. ²: Number of French bulls getting in the top100 in SSMACE compared to MACE. ³: Number of French bulls getting out of the top100 in SSMACE compared to MACE.

Therefore, the impact of the inclusion of SSMACE EBVs of foreign bulls included in the reference population used in the next French Single-Step evaluation is expected to be negligible.

Conclusion

For the past years, the transition from multi-step genomic evaluations to Single-Step genomic evaluations has been one of the main challenges for French research teams, evaluation center, breeding companies and breed societies.

In this paper, we presented a practical approach to estimate genomic-free Single-Step proofs, based on the recommendations of an Interbull working group (Sullivan, 2021). This approach is easy to implement at a national level and represents a reasonable amount of routine work to prepare the information needed for MACE evaluations. For all traits and breeds, the proofs produced by this approach passed the Interbull Trend Tests, most using the stringent method II. Consequently, the transition to Single-Step is now complete for dairy breeds, and, since December 2023, French proofs sent to MACE are based on Single-Step evaluations.

The estimated genetic trends based on genomic-free Single-Step EBVs were intermediate between those estimated with polygenic and Single-Step evaluations. This

confirms that there is a correction for the genomic preselection bias using genomic-free Single-Step EBVs, but that it is only partial. MACE and SSMACE EBVs are highly correlated, with regression slopes close to one and rankings are mostly preserved, hence French breeders can expect only minor differences in the subsequent national SSEBVs.

Acknowledgments

The authors thank M. Barbat (GenEval, France), S. Fritz (Eliance, France) and J. Promp (Idele, France) for the support provided in the different stages of the study. The contribution of Idele to this work received the financial support of the Ministry of Agriculture through the CASDAR.

References

Boichard, D., Bonaiti, B., Barbat, A. and Mattalia, S. 1995. Three methods to validate the estimation of genetic trend for dairy cattle. *J. Dairy Sci.* 78, 431-437.

Croué, I., Barbat, M., Launay, A., Promp, J., Guillern, M., Boulesteix, P., Minéry, S., Fritz, S., Tribout, T. and Boichard B. 2022. In France, Single-Step is going live. Available at:

https://interbull.org/static/web/220530_13h30_MONTREAL4-5_IolaCroule.pdf

GenEval. 2024. Evaluation génétique des bovins laitiers - production laitière – morphologie – caractères fonctionnels. Available at:

<https://www.geneval.fr/indexations-races-bovine>

Legarra, A. Chrisensen, O. F., Aguilar, I. and Misztal, I. 2014. Single Step, a general approach for genomic selection, *Livestock Science*, 166, 54-65.

Patry, C. and Ducrocq, V. 2011. Evidence of biases in genetic evaluations due to genomic preselection in dairy cattle. *J. Dairy Sci.* 94, 1011-1020

Sullivan, P. 2021. Genomic-free EBV for MACE. Available at:

https://interbull.org/ib/2021_webinar_summary