Can estrus expression be used in Holstein cows breeding to improve fertility traits ?

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Introduction — Fertility traits are important in Holstein cows breeding

Fertility traits were negatively correlated with milk traits, with a particular decline in high-yield population (Kadarmideen *et al*., 2013)

Economic benefits of the farm are closely related:： high conception rates → accelerated milk production cycle (Inchaisri *et al*.. 2011)

Total Performance Index (TPI, 2025)

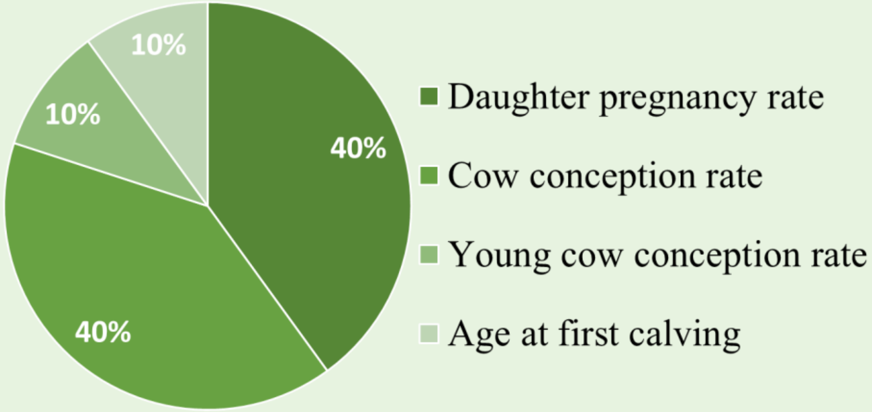
April 2025

TPI®

$$\left[19\frac{(PTAP)}{17} + 19\frac{(PTAF)}{22} + 8\frac{(FE)}{52} + 8\frac{(PTAT)}{0.8} + 11\frac{(UDC)}{0.8} + 6\frac{(FLC)}{0.8} + 5\frac{(PL)}{1.6} + 2\frac{(HT)}{2.0} + 3\frac{(LIV)}{1.4} - 4\frac{(SCS)}{0.13} + 13\frac{(FI)}{1.3} - .5\frac{(DCE)}{0.5} - 1.5\frac{(DSB)}{0.8}\right] 3.8 + 2845$$

引自： https://www.holsteinusa.com/

13% FI: Fertility Index



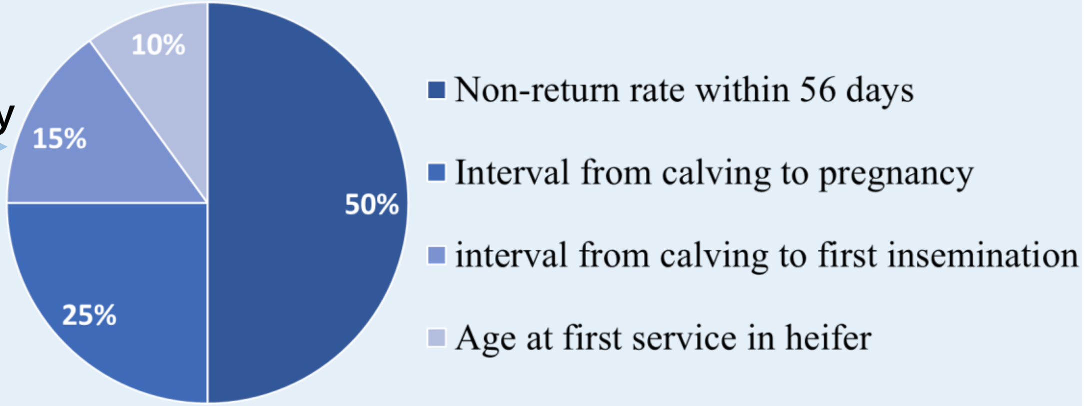
Logistics Performance Intex (LPI, 2023)

Lactanet

13.4% DF: Daughter Fertility

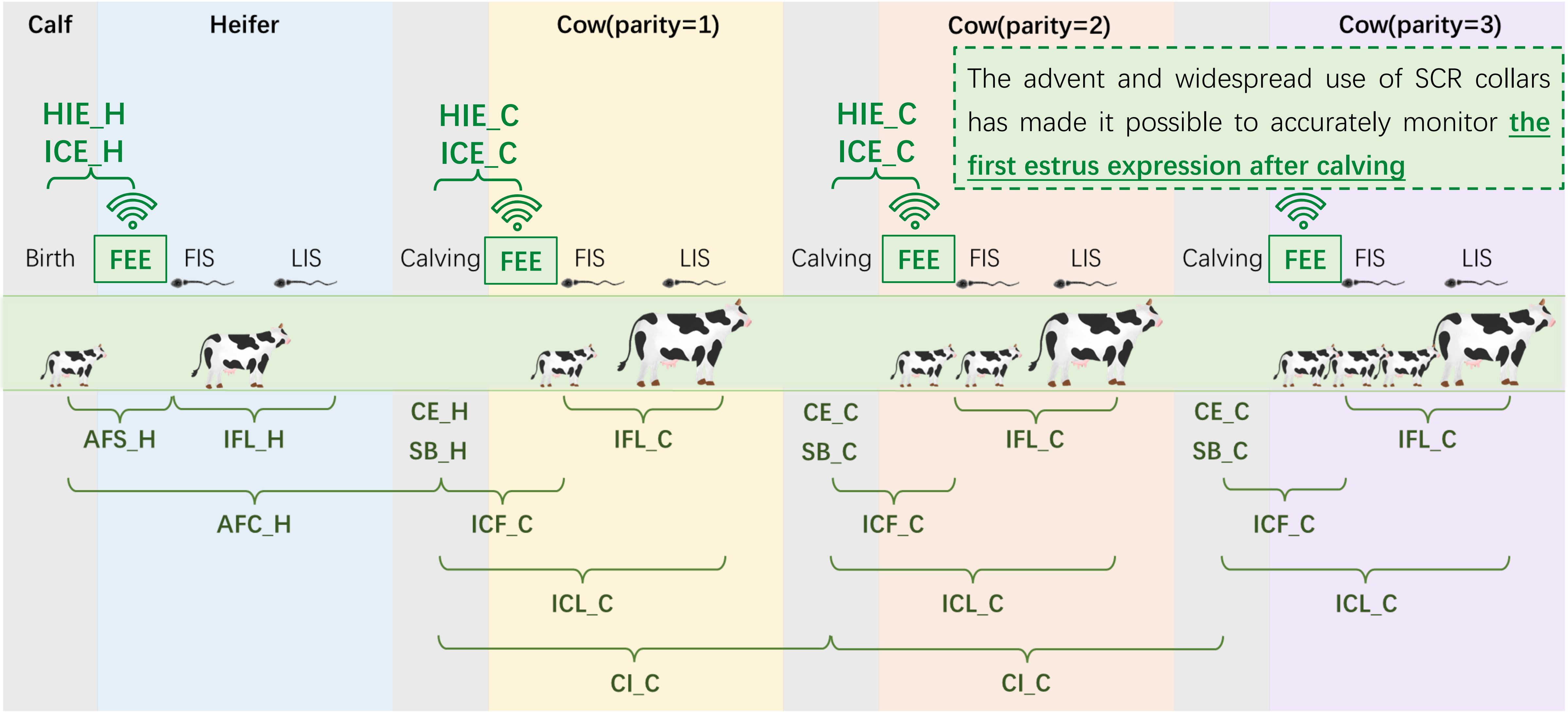
LPI = [Production Component x Emphasis x Factor + Durability Component x Emphasis x Factor + Health & Fertility Component x Emphasis x Factor] + Constant

https://lactanet.ca/



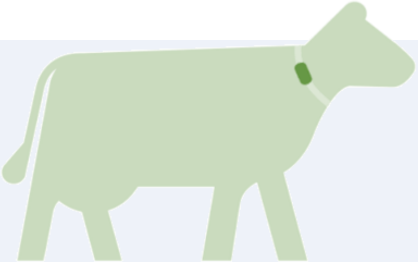
Introduction — Neck collars can catch the first estrus expression after calving

incompletе fertility event records → **Fertility traits unable to accurately reflect differences in the first estrus after calving**



The advent and widespread use of SCR collars has made it possible to accurately monitor **the first estrus expression after calving**

Introduction — Estrus expression and other traits



Heat Indicator (HI): ranging from **0 to 100 au** was calculated for each cow by the DataFlow™ II system and the HIs were recorded every two hours → **quantify the estrus expression ability**

HI and conception rate:

As the HI increases, the conception rate also increases (Macmillan *et al.*, 2020)

Increased HI can improve the conception rate of embryo transfer recipient (Madureira *et al.*, 2021)

The pregnancy rate was significantly higher in the collar group (**52.65%** VS 40.52%, $P < 0.01$)

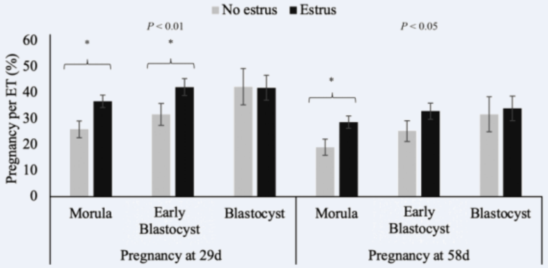


Figure 2. Pregnancy per embryo transfer (ET; mean % ± SEM) according to the stage of embryo development and the occurrence of estrus (estrus) or not (no estrus), in experiment 1. Pregnancy examination was performed at 29 and 58 d after the end of the timed ET. An interaction was observed between the occurrence of estrus (estrus and no estrus) and embryo developmental stage of the transferred embryo on pregnancy per ET. Asterisks indicate differences between bars ($P < 0.05$).

It was difficult to **measure** quantifiable estrus-related indicators, making it difficult to collect population phenotypes and conduct **genetic selection** and **breeding work** in the past

activity **(0.63^{**})**
Silper B. F., JDS, 2015

conception rate **(38.9 vs. 25.2%)**
Cerri R.L.A., JDS, 2017

pregnant rate by AI **(46.2 vs. 32.7%)**
Cerri R.L.A., JDS, 2017

Intensity of estrus

milk yeild **(-0.31^{*})**
Valenza A., JDS, 2012

follicular diameter**(0.20^{*})**
Madureira A.M.L., JDS, 2015

plasma estradiol concentration**(9.1^{a}vs8.2^{b})**
Silper B. F., JDS, 2015

Objects

Ningxia
China

- 🐄 **Select traits to effectively judge the first estrus expression ability of dairy cows**

- 🐄 **Estimate genetic parameters for the first estrus expression traits after calving**

- 🐄 **Finding genomic windows for the first estrus expression traits after calving**

Materials and Methods

Genomic windows of 10-SNPs that explained **0.15% or more of the total additive genetic variance** were associated with traits

Phenotype

The first estrus expression after calving:

- ✓ **DFE:** duration
- ✓ **ICE:** Interval
- ✓ **HIE:** heat indicators

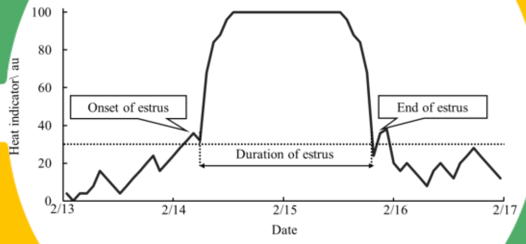
21583 dairy cows with **34836** FEEs

Pedigree

2318 sire and **41541** dam

Genotype

2064 cows with **116364** SNPs (ARS-UCD1.2)



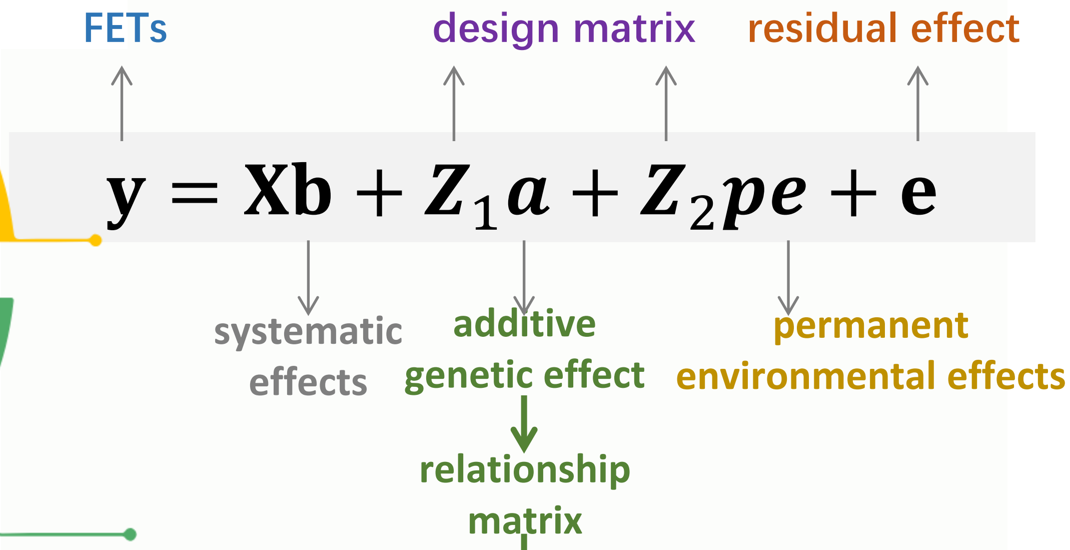
Higher than 35 au for 4h

$$H^{-1} = A^{-1} + \begin{bmatrix} 0 & 0 \\ 0 & G_{\alpha,w}^{-1} - A_{22}^{-1} \end{bmatrix}$$

(Aguilar *et al.*, 2017)

BLUPF90

repeatability animal models



FETs

design matrix

residual effect

$$\mathbf{y} = \mathbf{Xb} + Z_1 a + Z_2 pe + \mathbf{e}$$

systematic effects

additive genetic effect

permanent environmental effects

relationship matrix

PBLUP, $\boldsymbol{a} \sim N(0, A\sigma_a^2)$ ➡ **pedigree**

ssGBLUP, $\boldsymbol{a} \sim N(0, H\sigma_a^2)$ ➡ **pedigree-genotype**

Result ① — The first estrus expression traits after calving

Table 1. Effects of various factors on estrus expression traits after calving in Holstein cows

Effect	Level	N	DFE/h	ICE/d	HIE/au
Parity	1	12 343	**45.95±0.08^{a}**	**49.42±0.18^{a}**	80.07±0.14^{b}
	2	9 964	45.64±0.09^{b}	**43.79±0.20^{d}**	**80.77±0.16^{a}**
	3	6 828	45.32±0.11^{c}	44.87±0.23^{c}	79.59±0.18^{b}
	≥4	5 701	45.12±0.12^{cd}	45.74±0.24^{b}	78.15±0.20^{c}
AFS	275~405	7 691	45.65±0.09	46.20±0.18^{a}	**79.82±0.15^{a}**
	406~415	4 406	45.55±0.11	**45.54±0.22^{b}**	**79.97±0.18^{a}**
	416~425	4 060	45.46±0.12	**45.50±0.25^{b}**	79.56±0.20^{ab}
	426~786	5 426	45.36±0.10	46.57±0.21^{a}	79.24±0.17^{bc}
Day and night	day	15 828	**46.32±0.08^{a}**	46.72±0.17^{a}	**79.45±0.14^{b}**
	night	19 008	44.70±0.08^{b}	45.19±0.16^{b}	79.84±0.13^{a}

Note: Different letters in the same row means significant difference between the treatments （P <0.05）

1 DFE and ICE of primiparous cows are longest

2 2^{ed} parity cows inhibit lowest ICE & highest HIE

3 Better fertility traits of heifers inhibit higher HIE

4 Cows with earlier or later insemination inhibit longer ICE

5 Cows inhibit longer DFE and lower HIE during the day

✓ **The FEEs of dairy cows based on the data of collars show differences between primiparous cows and multiparous cows**

(陈紫薇等, 2021. Lovendahi *et al.*, 2019. Macmillan *et al.*, 2020.)

Result ② — The FETs' genetic parameters based on PBLUP and ssGBLUP

Table 2. Genetic components and parameters for the first estrus expression traits after calving based on pedigree relationship matrix (PBLUP) and single-step GBLUP (ssGBLUP) in Holstein cows

Traits	PBLUP					ssGBLUP				
	σ_a^2	σ_{pe}^2	σ_e^2	h^2(se)	r^2	σ_a^2	σ_{pe}^2	σ_e^2	h^2(se)	r^2
DFE	**25.505**	14.234	232.978	**(0.011)**	0.146	**27.263**	14.233	232.896	**(0.011)**	0.151
ICE	**0.650**	1.215	58.723	**(0.005)**	0.031	**1.221**	0.720	58.705	**(0.005)**	0.032
HIE	**2.830**	4.735	167.700	**(0.016)**	0.043	**2.925**	4.864	167.629	**(0.016)**	0.044

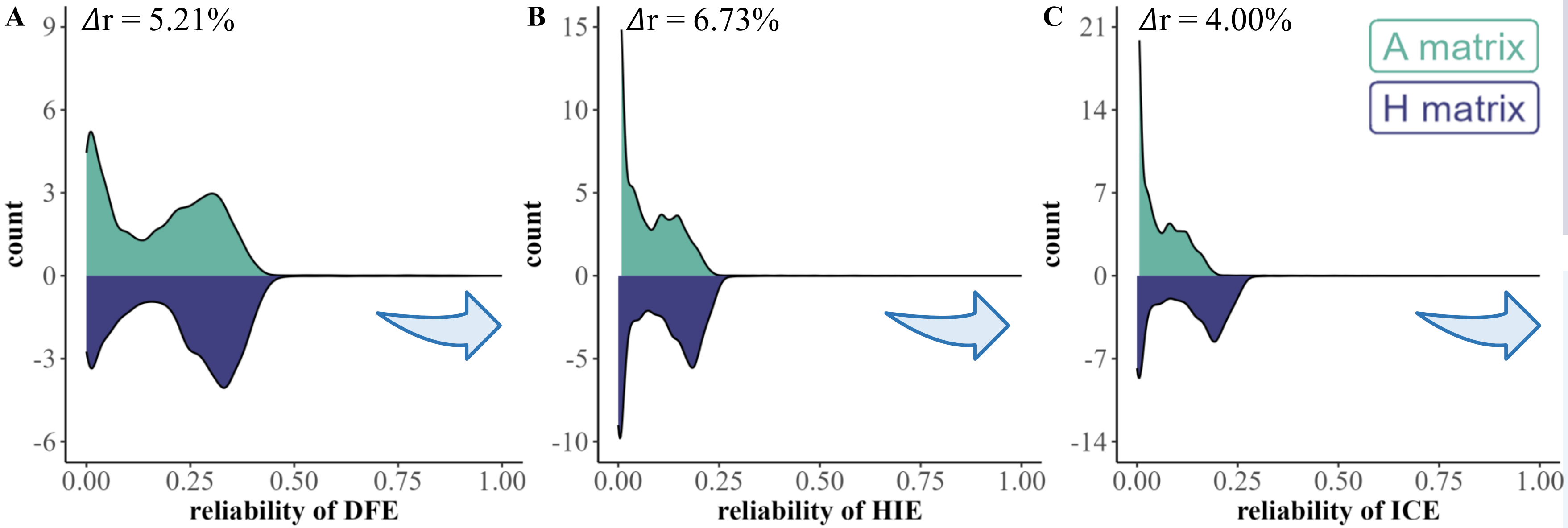


Figure 1. Density distribution of the estimated breeding value reliability for first estrus expression traits after calving

✓ Compared with PBLUP, both and σ_a^2 and h^2 improved in ssGBLUP

✓ Compared with PBLUP, the standard error was not reduced

- **Low proportion of chips**
- **High quality of pedigree**

✓ The reliability of EBVs for all FEEs showed improvement

Result ② — Genetic parameters for FETs and fertility traits

Table 3. Heritabilities, genetic correlations and phenotypic correlations for first estrus expression traits after calving in Holstein cows

Traits	**DFE**	**ICE**	**HIE**
DFE	**0.094±0.011**	**-0.067±0.107**	**0.722±0.095***
ICE	0.153±0.005*	**0.011±0.005**	**-0.335±0.094***
HIE	0.545±0.004*	0.179±0.005*	**0.016±0.005**

Note: Heritabilities, genetic correlations and phenotypic correlations located in the diagonal, upper, and lower triangles, respectively. Significance of genetic and phenotypic correlations was tested based on *t*-tests, and * represents (*P* <0.05).

Table 4. Genetic correlations between the first estrus expression traits after calving and routine fertility traits in Holstein cows

Traits	**DFE**	**ICE**	**HIE**
CR	**0.766±0.262***	**-0.327±0.075***	**0.580±0.193***
IFL	0.013±0.583	0.118±0.168	**-0.642±0.298***
CI	-0.124±0.065	**0.188±0.042***	**-0.146±0.068***

Note: Significance of genetic and phenotypic correlations was calculated based on t-tests, and * represents (*P* <0.05).

1 **DFE, ICE, and HIE all showed low heritability ($h^2 < 0.1$)**

2 **When ICE shortens, DFE be longer but HIE be higher**

3 **When DFE lengthens, CR be higher**

4 **When ICE lengthens, CR be lower and CI be longer**

5 **When HIE increases, CR be higher, IFL&CI be shorter**

✓ **FEEs are heritable and genetic correlation were observed with them, which can be used as selecting dairy cows with high fertility traits**

(董刚辉等, 2021. Ioannidis *et al*., 2018. Ismael *et al*., 2015.)

Result ③ — Twenty bulls with the high and low FTE's EBV

Table 5. The summary of bulls with highest and lowest FETs' EBV ($R^2 > 0.4$)

Bull	EBV_DFE	R^2_DFE	Daughter	EBV_ICE	R^2_ICE	Daughter	EBV_HIE	R^2_HIE	Daughter
Top	1.74	0.55	173	-0.11	0.62	34	2.66	0.43	112
	1.39	0.54	145	-0.08	0.44	25	2.44	0.47	156
	1.33	0.60	139	-0.08	0.48	5	2.14	0.49	99
	1.21	0.53	100	-0.08	0.44	31	2.11	0.51	172
	1.16	0.46	22	0.03	0.45	20	2.08	0.48	71
	1.12	0.60	191	0.12	0.60	54	2.02	0.64	284
	1.06	0.72	304	0.14	0.56	33	1.96	0.55	191
	1.02	0.65	145	0.15	0.43	17	1.93	0.58	202
	0.95	0.40	45	0.15	0.47	19	1.93	0.54	153
	0.83	0.53	152	0.16	0.88	215	1.86	0.78	638
Bottom	-0.98	0.68	121	10.50	0.81	122	-1.72	0.71	360
	-0.96	0.49	121	8.79	0.73	61	-1.70	0.65	264
	-0.95	0.74	542	7.88	0.74	98	-1.65	0.44	92
	-0.95	0.51	34	7.21	0.61	45	-1.43	0.41	28
	-0.94	0.41	50	5.65	0.91	299	-1.40	0.42	79
	-0.92	0.74	293	5.63	0.65	50	-1.16	0.44	33
	-0.88	0.43	128	4.99	0.77	92	-1.13	0.63	249
	-0.84	0.48	98	4.71	0.71	73	-1.11	0.68	149
	-0.84	0.59	110	4.56	0.80	155	-0.96	0.67	532
	-0.82	0.45	127	4.52	0.76	167	-0.95	0.45	97

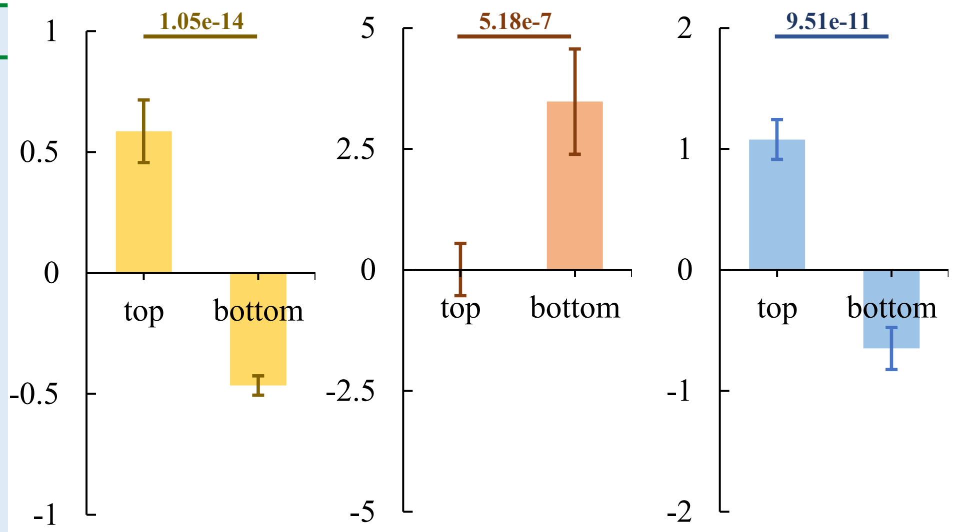


Figure 2. Twenty bulls' daughter's EBV of DFE, ICE, and HIE

- ✓ **The FEEs of the daughters of the top 10 bulls are significantly superior than those of the daughters of the bottom 10 bulls**

Result ④ — The 10-SNPs genomic windows for FETs based on WssGWAS

1 A total of 78 protein-coding genes located in 47 genomic windows

2 3 genomic windows were identified by 2 traits at the same time

3 Genomic windows overlap with 35 known QTLs for fertility traits

Table 6. The genomic windows explaining more than 0.15% of the total additive genetic variance for FETs

Traits	genomic windows	genetic variance	candidate genes
DFE	17	**3.78%**	37
ICE	18	**3.24%**	17
HIE	12	**2.28%**	26

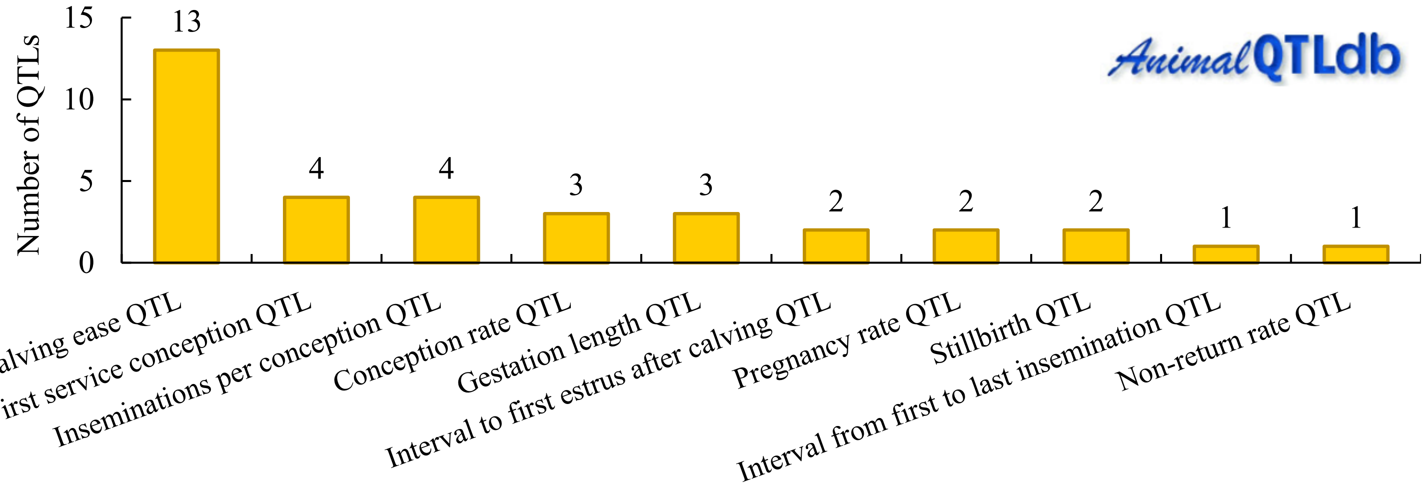


Figure 3. The relative QTLs of the 10-SNP windows explaining more than 0.15% of the total additive genetic variance for the first estrus expression traits after calving in Holstein cattle

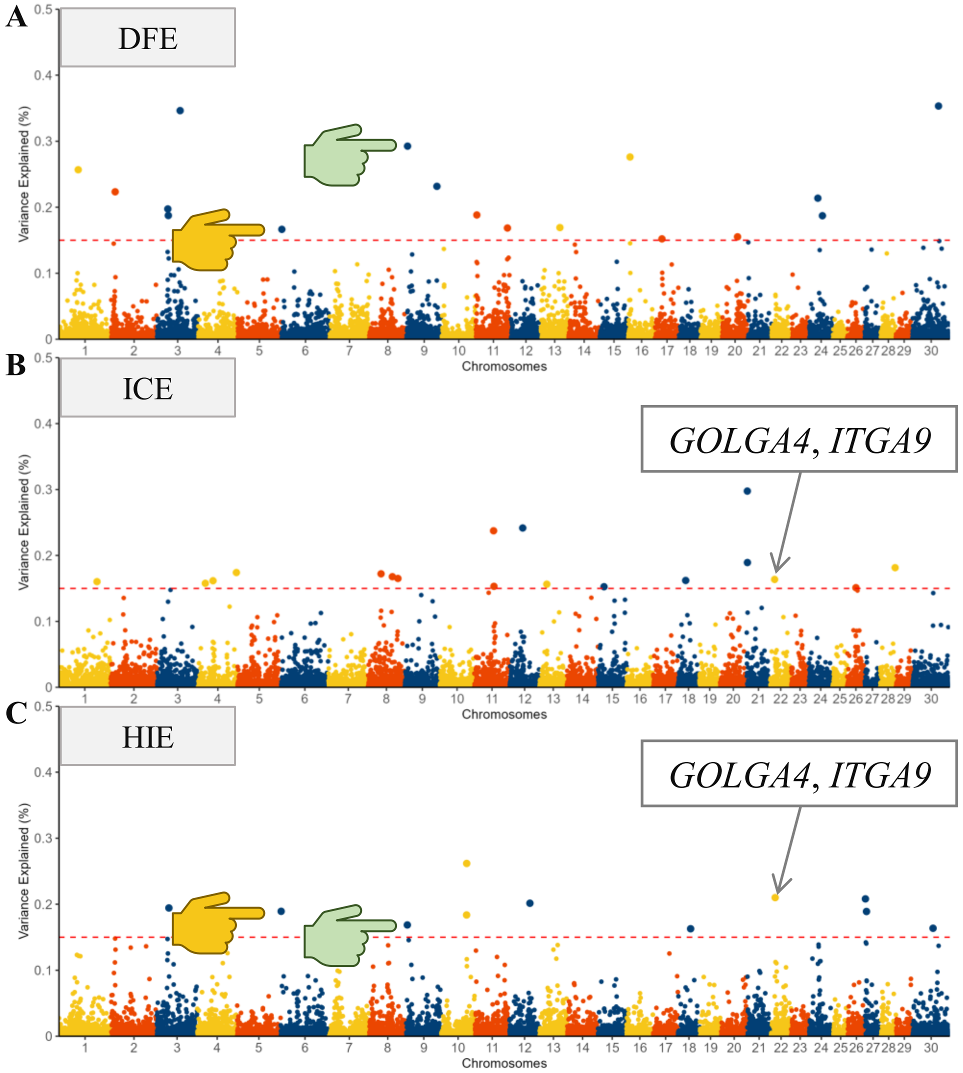


Figure 4. Proportion of the total additive genetic variance of 10-SNP genomic windows based on the weighted single-step genome association studies for the first estrus expression traits after calving.

Result ④ — Candidate genes for FETs

1 Candidate genes enriched in **meiosis** and potassium **ion transmembrane**

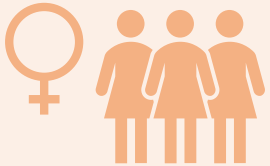
2 Candidate genes participate in four important **KEGG pathways**

✓ovarian steroidogenesis ✓oocyte meiosis

✓arachidonic acid metabolic ✓progesterone-mediated oocyte maturation

- ***CYM*** is associated with **low fertility traits** in goats (Lai *et al.*, 2016)
- ***ZNF52*** is associated with fertility index, number of inseminations, IFL, and non-return in Nordic red cattle (Sigdel *et al.*, 2020. Höglund *et al.*, 2015.)

- ***HUWE1*** deficiency causes **oocyte death** and **infertility** (Eisa *et al.*, 2020)
- ***HENMT1*** mutations cause a significant decrease in the number of **follicles** and **litter size** in female mice (Karla *et al.*, 2021)
- ***SEMA4D*** mutations cause a decrease in **GnRH** (Romain *et al.*, 2011)



- ***PROK1*** is an important gene that regulates the proliferation and development of reproductive tissue cells (Eddid *et al.*, 2015) (Peek *et al.*, 2024)
- ***ST6GALNAC5*** upregulated in women with ovarian dysfunction

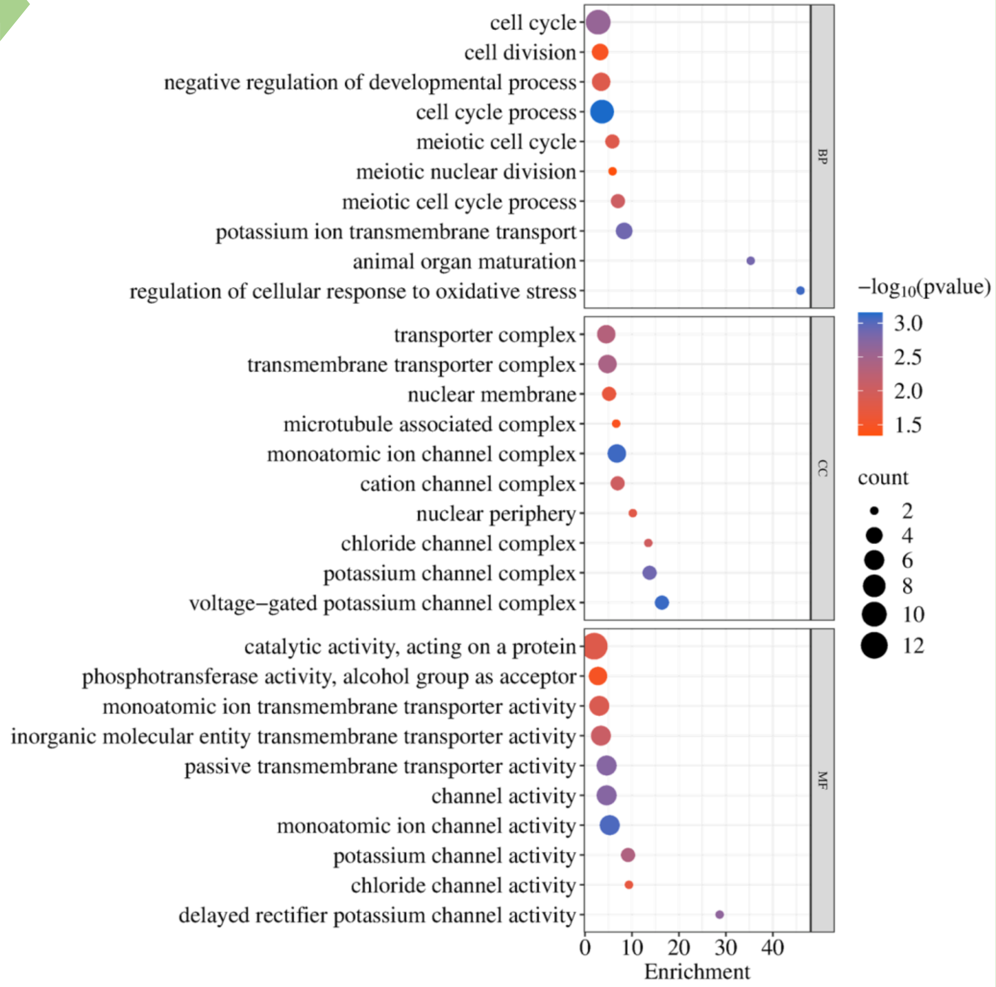
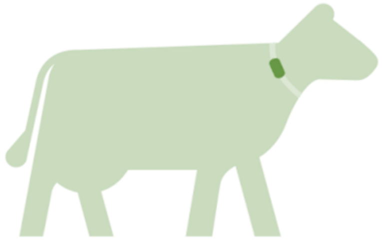


Figure 5. Enriched gene ontology (GO) terms for genes within the 10-SNP windows explaining more than 0.15% of the total additive genetic variance for the first estrus expression traits after calving in Holstein cattle

Take home messages

- **Parity, age at first service in heifer, day-night, herd, and calving season all have a significant impact on the first estrus expression traits of after calving (FEEs)**
- **All FEEs showed low heritability**
- **FEEs can be used for selecting dairy cows with high fertility traits**



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Thanks for your attention!

Q&A

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