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INTERPRETIVE SUMMARY

25 **Fertility genetic merit and reproductive efficiency:** By Cummins et al., Page 000.

26 To elucidate the underlying physiological basis of declining reproductive
27 performance, the current study compared the phenotypic performance of cows with
28 divergent genetic merit for fertility traits, but with similar genetic merit for milk
29 production traits. Superior genetic merit for fertility traits was associated with
30 improved fertility performance, increased circulating concentrations of insulin-like
31 growth factor-I, and greater BCS. These results highlight the important contribution of
32 genetic merit for fertility to phenotypic reproductive performance, which may not
33 necessarily be to the detriment of milk production.

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35

36 **FERTILITY GENETIC MERIT AND REPRODUCTIVE EFFICIENCY**

37

38 **The effect of genetic merit for fertility traits on production characteristics and**
39 **reproductive efficiency of Holstein cows in a pasture-based system**

40

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ABSTRACT

55 The objective of the present study was to characterize the phenotypic performance
56 of cows with similar proportions of Holstein genetics, similar genetic merit for milk
57 production traits, but with good (Fert+) or poor (Fert-) genetic merit for fertility traits.
58 Specifically, we tested the hypothesis that cows with a negative EBV for calving
59 interval would have superior fertility performance and would have detectable
60 differences in body reserve mobilisation and circulating concentrations of metabolic
61 hormones and metabolites compared with cows that had a positive EBV for calving
62 interval. For the duration of the study, cows were managed identically as a single herd
63 in a typical grass-based spring-calving production system. A total of 80 lactation
64 records were available from 26 Fert+ and 26 Fert- cows over 2 consecutive years
65 (2008 and 2009). During yr 1, cows were monitored during a 20 wk breeding season
66 to evaluate reproductive performance. Milk production, body condition score (BCS,
67 scale 1 to 5), body weight, grass dry matter intake, energy balance, and metabolic
68 hormone and metabolite data were collected during both years. Fert+ cows had
69 greater daily milk yield (19.5 kg/d vs. 18.7 kg/d), shorter interval from calving to
70 conception (85.6 d vs. 113.8 d) and fewer services per cow (1.78 vs. 2.83). No
71 difference between groups in grass dry matter intake, energy balance, or body weight
72 was observed. Fert+ cows maintained greater BCS during mid (2.84 vs. 2.74 units)
73 and late lactation (2.82 vs. 2.73 units). Circulating concentrations of insulin-like
74 growth factor I were greater throughout the gestation/lactation cycle in Fert+ cows

75 (148.3 ng/mL vs. 128.2 ng/mL). Fert+ cows also had greater circulating
76 concentrations of insulin during the first 4 wk of lactation (1.71 μ IU/ml vs. 1.24
77 μ IU/ml). Analysis of records from national herd data verified the association between
78 genetic merit for fertility traits and phenotypic reproductive performance; Fert+ cows
79 (n = 2436) required 11.1 d less to recalve than Fert- cows (n = 1388), and the
80 percentage of cows that successfully calved for the second time within 365 d and 400
81 d of the first calving was 8% and 13% greater for Fert+ compared with Fert- cows,
82 respectively. These results demonstrate that genetic merit for fertility traits had a
83 pronounced effect on reproductive efficiency, BCS profiles and circulating
84 concentrations of insulin-like growth factor I.

85 **Key words:** genetic selection, fertility traits, reproduction, insulin-like growth factor-I

86

87

INTRODUCTION

88

89 The efficiency of pasture-based milk production systems is based on maximising
90 milk production from low-cost grazed grass (McCarthy et al., 2007). The success of
91 these systems is largely dependent on achieving a compact calving pattern to coincide
92 with the start of the grass growing season. The maintenance of this compact calving
93 pattern is reliant on the cow's ability to successfully calve, complete uterine
94 involution, return to cyclicity, exhibit estrus, conceive, and maintain a pregnancy in a
95 window of 85 d, thus maintaining a 365-d calving interval. The relative importance of
96 female fertility is markedly greater in seasonal calving systems than in year-round
97 calving systems (Veerkamp et al., 2002). Failure to maintain the target calving
98 interval of 365 d in a seasonal calving system results in financial losses at farm level
99 (Evans et al., 2006b). This arises through reduced volumes of saleable milk and

100 increased voluntary and involuntary culling rates and breeding costs (Plaizier et al.,
101 1997).

102 Despite the crucial role fertility plays in maximising economic output, reproductive
103 efficiency in dairy cows has declined during the past half century both in Ireland
104 (Evans et al., 2006a) and internationally (Royal et al., 2000, Washburn et al., 2002).
105 In Ireland, some of this decline has been attributed to the intense selection for milk
106 production traits and the associated introgression of North American Holstein genes
107 (Horan et al., 2004). An extensive study of performance indicators on 14 spring-
108 calving dairy farms between 1990 and 2001 in Ireland confirmed these trends (Evans
109 et al., 2006a). During this 11-yr period, the pedigree index for milk volume increased
110 by 25 kg per yr, the proportion of Holstein genes increased from 8% to 63% and the
111 calving rate to first service declined from 55% in 1990 to 44% in 2001.

112 To address the problem of declining fertility, the Irish national breeding programme
113 shifted from being predominantly focused on milk production traits to a more holistic
114 multi-trait index called the Economic Breeding Index (EBI) (Veerkamp et al., 2002).
115 Introduced in 2001, the EBI included production and non-production traits, thus
116 identifying animals of superior genetic merit for delivering on-farm profit (Berry,
117 2007). Since its introduction, the EBI has evolved to include 6 subindexes (relative
118 emphasis in parenthesis); milk production (38.1%), fertility/survival (34.8%), calving
119 performance (10.3%), beef carcass (7.2%), maintenance (6.1%) and health (3.6%)
120 (<http://www.icbf.com>). The fertility subindex is comprised of 2 traits; calving interval
121 (23.2%) and survival (11.5%). Good genetic merit for fertility traits requires negative
122 EBV's for calving interval and positive EBV's for survival.

123 Previous studies evaluating the effects of genetic influences on reproductive
124 efficiency have compared animals of varying genetic merit for milk production

125 (Buckley et al., 2000) or varying Holstein ancestry (Horan et al., 2004). In both cases,
126 large differences in reproductive performance were reported. The precise mechanisms
127 responsible for fertility differences remain poorly understood. In previous studies
128 comparing models of good and poor fertility, observed differences in phenotypic
129 fertility performance were generally confounded with genetic merit for milk yield and
130 phenotypic milk production. Therefore, the aim of this study was to characterize the
131 phenotypic performance of cows with similar genetic merit for milk production traits
132 and similar proportions of Holstein genes, but divergent genetic merit for fertility
133 traits (genotypes Fert+ and Fert-). Specifically, we tested the hypothesis that cows
134 with negative EBV's for calving interval would have superior fertility performance,
135 would be less reliant on body reserve mobilisation, and would have detectable
136 differences in circulating concentrations of metabolic hormones and metabolites
137 compared with cows that had positive EBV's for calving interval.

138

139 **MATERIALS AND METHODS**

140

141 ***Herd Establishment***

142 Using the autumn 2007 official dairy evaluation published by the Irish Cattle
143 Breeding Federation (ICBF), the national dairy cattle database was screened for
144 heifers due to calve for the first time in Spring 2008. Restrictions were placed on the
145 estimated breeding value (EBV) for milk production (between +200 kg and +900 kg)
146 and proportion of Holstein genetics (> 75%). Within this population, heifers with
147 extreme positive (i.e., poor fertility) and negative (i.e., good fertility) EBVs for
148 calving interval were identified. Poor fertility (Fert-) heifers were restricted to animals
149 where both the sire and maternal grand-sire had positive EBVs for calving interval.

150 Conversely, good fertility (Fert+) heifers were restricted to animals where both the
151 sire and maternal grand sire had negative EBVs for calving interval. Heifers identified
152 as being available for purchase were screened for infectious diseases. A total of 18
153 nulliparous Fert- and 18 nulliparous Fert+ cows passed the Moorepark Biosecurity
154 Protocol and were purchased and moved in January 2008 to the Moorepark Animal &
155 Grassland Research and Innovation Centre in Fermoy, Co. Cork, Ireland (55°10'N
156 8°16'W) .

157 The process of selection described above was repeated with the same criteria (as
158 explained above) during autumn 2008 and an additional 8 nulliparous Fert- and 8
159 nulliparous Fert+ heifers were purchased. The EBVs of the two genotypes are
160 summarized in Table 1. Within the Irish national herd, these animals were
161 representative of the top quartile in genetic merit for milk production, while the Fert+
162 and Fert- groups represented the top 20% and bottom 5% for calving interval,
163 respectively.

164

165 ***Feed and Management System***

166 Animals were managed identically as one herd in a typical grass-based spring-
167 calving production system. Following parturition, cows were turned out to grass in
168 early February until mid-November and grazed under a rotational grazing system, as
169 described by Dillon et al. (1995), in a predominantly perennial ryegrass (*Lolium*
170 *perenne* L.) sward. Fresh pasture was allocated daily to the herd following morning
171 milking using temporary fencing. During yr 1, the mean daily herbage allowance of
172 13 ± 1.45 kg DM/cow day⁻¹ was supplemented with 3.9 ± 1.21 kg DM/cow day⁻¹ of
173 concentrate. Similarly during yr 2, the mean daily herbage allowance of 14.3 ± 1.29
174 kg DM/cow day⁻¹ was supplemented with 4.0 ± 1.69 kg DM/cow day⁻¹. The mean

175 pregrazing herbage yield was 1337 ± 494 kg DM/ha in yr 1 and 1146 ± 396 kg DM/ha
176 in yr 2. Cows were dried off 80 d before expected calving date, housed in a free stall
177 barn and given full time access to a total mixed ration (TMR) of grass silage (90%)
178 and concentrate (10%).

179

180 *Animal Measurements*

181 Cows were milked twice daily at 0730 and 1630 h. Milk yield was recorded at each
182 milking using electronic milk meters (Dairymaster, Causeway, Co. Kerry, Ireland).
183 Milk composition (fat, protein and lactose) was determined weekly from successive
184 evening and morning samples by mid-infrared reflectance spectroscopy using a
185 FT6000 Milkoscan instrument (DK-3400, Foss Electric, Hillerød, Denmark). Cow
186 liveweight was measured weekly and BCS (Edmonson et al., 1989) was assessed
187 every 2-3 wk throughout the study. During lactation, live weight and BCS
188 measurements were taken immediately after morning milking by a single operator.
189 Individual animal grass dry matter intake (GDMI) measurements were carried out on
190 three occasions during yr 1 at wk 13, 28 and 35 postpartum ($SD \pm 3.2$ wk for all time
191 points) and on one occasion in yr 2 at wk 13 postpartum ($SD \pm 3.9$ wk) using the n-
192 alkane technique (Mayes et al., 1986) as modified by Dillon and Stakelum et al.
193 (1989). Supplemental concentrate fed at the time of GDMI measurements was added
194 to the GDMI figure to calculate total dry matter intake (TDMI). Energy Balance (EB)
195 was estimated as the difference between energy intake and the sum of the energy
196 required for maintenance and milk production. The French net energy (NE) system
197 was used, where 1 Unité Fourragère Lait (UFL) is the NE content of 1 kg of air-dried
198 barley for milk production (Jarrige, 1989).

199

200 ***Blood Sampling and Laboratory Analysis***

201 In yr 1, blood samples were collected once every 2 wk throughout lactation. In yr 2,
202 blood samples were collected weekly for 3 wk prior to parturition, twice weekly
203 during the first 4 weeks of lactation, once weekly from wk 5 to 9 and once every 2 wk
204 thereafter until the end of lactation. Sampling took place after morning milking and
205 before returning to fresh pasture. Blood samples were collected via coccygeal
206 venipuncture into vacutainers containing lithium heparin as an anticoagulant (Becton
207 Dickinson, Plymouth, UK). Samples were placed in a centrifuge for 15 min at 2,000 ×
208 g, plasma was decanted, and stored at -20 °C until further analysis. Blood plasma was
209 analysed for the metabolites non-esterified fatty acid (NEFA), β-hydroxybutyrate
210 (BHB) and glucose concentrations by enzymatic colorimetry (NEFA kit supplied by
211 Wako Chemicals, GmbH, Niddnstraße, Germany; BHB and glucose kits supplied by
212 ABX Mira, Montpellier, France). Plasma insulin concentrations were determined using
213 a solid-phase fluoro-immunoassay (AutoDELFIA, PerkinElmer Life and Analytical
214 Sciences, Turku, Finland), with appropriate kits (Unitech BD Ltd., Dublin, Ireland).
215 Inter and intra-assay coefficients of variation were 17.5% and 8.5%, respectively.
216 Circulating IGF-I concentrations were quantified using a validated double antibody
217 radioimmunoassay, following ethanol:acetone:acetic acid extraction as described by
218 Enright et al. (1989). Inter and intra-assay coefficients of variation were 15.6% and
219 15.8% respectively. For all hormone assays, each genotype was equally represented in
220 each assay and all samples for a cow of a given genotype were completed in a single
221 assay.

222

223 ***Reproductive Management***

224 The reproductive performance of the herd was monitored in yr 1 during a 20-wk
225 breeding season with a mating start date (MSD) of April 14. Before MSD, all cows
226 greater than 30 d in milk (DIM) were examined using transrectal ultrasonography
227 (7.5-MHz transrectal transducer, Aloka SSD-900, Aloka Ltd., Tokyo, Japan) to
228 determine utero-ovarian status. One cow from each genotype was diagnosed with
229 endometritis, and were treated with an i.m. PGF_{2α} injection containing 25 mg
230 dinoprost tromethamine (Lutalyse; Pfizer Ireland, Dublin, Ireland) followed by
231 intrauterine cephalixin (Metricure; Intervet, Boxmeer, the Netherlands) 2 – 4 days
232 later after observation of estrus. During the breeding season, heat detection was
233 carried out a minimum of 3 times daily with the aid of tail paint. Cows detected in
234 heat were inseminated with frozen-thawed semen from sires of their own genetic
235 group to generate replacement heifers. For each sire used, semen was from a single
236 ejaculate, and sperm viability and quality was verified before use on the experiment.
237 All inseminations were carried out by a single AI technician during the breeding
238 season. Pregnancy diagnosis was carried out by transrectal ultrasound at 30-36 d and
239 again at 60-66 d post-insemination to measure the rate of late embryo loss. Final
240 pregnancy status was determined by transrectal ultrasound 80 d after the completion
241 of the breeding season. In yr 2, breeding was delayed to allow collection of biological
242 samples of interest and hence no fertility performance indicators were available.

243

244 ***Data Handling***

245 All data handling was carried out using SAS (SAS Institute, 2006). Data were
246 checked for normality. A Box-Cox transformation was used to normalise the
247 distribution of IGF-I, insulin, NEFA and BHB data. Linear interpolation was used to
248 calculate values for every day of the study for blood metabolites, hormones, and BCS,

249 and the estimated daily values were collapsed into weekly means. Circulating
250 concentrations of blood metabolites, metabolic hormones, and BCS data for each cow
251 were analysed over the full gestation/lactation cycle (from wk -3 (-6 for BCS) to wk
252 42). These data were also divided into 4 time periods (dry period: from wk -3 (-6 for
253 BCS) to 0 pre partum; early lactation: from wk 1 to 12 post partum; mid lactation:
254 from wk 13 to 28 post partum; and late lactation: from wk 29 to 42 post partum).
255 Additionally, separate analysis was carried out on blood metabolite and hormone
256 concentration data during wk 1 to 4 of lactation. The week of BCS nadir was
257 determined by identifying the earliest postpartum occurrence of the lowest BCS value
258 recorded during the first 15 wk of lactation.

259

260 *Study replicated using National Herd Data*

261 Data from the Irish national herd were also used to determine whether results from
262 the Fert+/Fert- herd were consistent with on-farm performance records from cows of
263 similar genetic merit. Data on first parity Holstein dairy cows calving between the
264 years 2006 and 2010 inclusive (n = 501,922) were extracted from the Irish Cattle
265 Breeding Federation database. Herds that were predominantly spring calving, where
266 80% of cows calved between the 1st January and 31st July, were retained. Herds with
267 less than 5 cows in any year were removed for that year. Only cows from a known AI
268 sire and AI maternal grand sire (n = 185,354) were retained. The pedigree index for
269 each cow was calculated as $0.5 \times \text{sire EBV} + 0.25 \times \text{maternal grand sire EBV}$. To
270 avoid any environmental covariance between cow phenotypic performance and cow
271 pedigree index, the preceding year's official dairy evaluations published by the Irish
272 Cattle Breeding Federation were used to estimate cow pedigree index. Restrictions
273 were placed on cow EBV for milk production (between +256 kg and +514 kg),

274 calving interval (Fert+: sire < -3.5, maternal grand sire <-3.5, Fert-: sire > 3.5,
275 maternal grand sire > 2) and proportion of Holstein genes (> 71%) in line with the
276 selection criteria set out for the Fert+ and Fert- cows used in the controlled study. A
277 total of 2371 Fert- and 4859 Fert+ first lactation cows in 2560 herds were available
278 for inclusion in the analysis. The number of cow records available for each of the five
279 yr (2006-2010) were 460, 978, 1198, 1801, and 2793, respectively. An additional
280 79,408 first parity herd-mates of these animals were included in the analysis for
281 improved estimation of fixed effects, especially contemporary group effects.

282 Standardized 305-d milk, fat and protein yield were estimated by area under the
283 curve from d 0 to d 305 (Olori, 1999). Milk fat and protein percent was calculated by
284 dividing fat and protein yields by milk yield. The fat to protein ratio was calculated by
285 dividing fat yield by protein yield. Calving interval was defined as the number of days
286 from the date of first calving to the date of second calving. Animals that failed to
287 recalve or had calving intervals outside the range of 300 to 800 d were removed from
288 the analysis of fertility variables. Two separate traits were used to describe the
289 survival of an animal to its second lactation: recalving within 365 d of first calving
290 (the target for a seasonal calving system) and recalving within 400 d of first calving (a
291 minimum target for a seasonal calving system).

292 An algorithm was used to generate contemporary groups for herd-year-season at
293 first calving for milk production and calving interval data separately. The algorithm
294 initially grouped first parity animals within herd that had a calving date within 10 d of
295 each other. If the number of records within any contemporary group was less than 7
296 they were combined with the next contemporary group, if the start date of one
297 contemporary group and the end date of the other contemporary group was < 182 d.
298 Only contemporary groups with a minimum of 5 animals were retained. A total of

299 86,638 animals in 4290 contemporary groups remained for inclusion in analysis of
300 milk production data while 49,661 animals in 3572 contemporary groups remained for
301 inclusion in the analysis of calving interval and survival.

302

303 *Statistical Analyses*

304 ***Experimental Herd Data.*** All statistical analyses were carried out using SAS (SAS
305 Institute, 2006). The effect of genotype on variables with repeated measures such as,
306 weekly production variables (milk yield, milk fat, protein and lactose concentrations,
307 BCS and BW), GDMI, TDMI and energy balance variables (yr 1), and blood
308 metabolite and hormone concentration were determined using mixed models with cow
309 nested within genotype as a random effect. A first-order autoregressive covariance
310 structure with homogeneous variances among weekly records within cow-parity
311 provided the best fit to the data. Transformed data (IGF-I, insulin, NEFA and BHB)
312 were used to calculate *P*-values, and the estimated group means and 95% confidence
313 intervals were derived from back-transformed values. The effect of genotype, parity,
314 calving date, yr, lactation wk, and their interactions were included in the final model
315 where significant ($P < 0.1$). Where an interaction existed between genotype and
316 parity, orthogonal contrasts were used to compare differences between genotypes
317 within parity.

318 The effect of genotype on continuous variables without repeated measures such as
319 number of services, calving to first service interval (all cows were served at least
320 once), calving to conception interval, GDMI, TDMI and energy balance variables (yr
321 2), and selected BCS variables (i.e., BCS at calving, postpartum BCS nadir, wk of
322 BCS nadir, changes in BCS from parturition to nadir and from nadir to end of
323 lactation) were determined using mixed model with cow nested within genotype as a

324 random effect. The effect of genotype, parity, and the interaction between genotype
325 and parity were tested, calving date and yr were included as adjustment variables, and
326 significant effects ($P < 0.1$) were maintained in the final model.

327 Differences between genotypes for variables with a binomial distribution
328 (conception rate to first service, pregnancy rate to first and second service, submission
329 rate, 42-d pregnancy rate, embryo mortality and overall pregnancy rates) were tested
330 using the Fishers exact test. The interval from mating start date to conception was
331 used to calculate a Kaplan-Meier estimate of survival function. Log-rank tests were
332 used to compare the resulting survival analysis curves. For illustrative purposes the
333 proportion of cows pregnant was extrapolated from the survival distribution function
334 for each genotype.

335 A Wilmink (Wilmink, 1987) exponential model curve was fitted to daily milk yield
336 to generate lactation profiles for each lactation. The Wilmink function is described by
337 the equation: $y_t = a + b e^{-0.05 \times t} + ct$.

338 In this model y_t represents daily milk yield at day t of lactation. The a parameter
339 represents the height of the curve, a negative b value and c value represent the rate of
340 increase of daily milk yield during the initial lactation stage, and the rate of decline
341 from peak, respectively. Regression parameters were estimated separately for each
342 cow-parity. Residuals of predicted daily milk yield were then calculated as the
343 difference between the predicted and the actual at each day of lactation. The mean
344 square prediction error was calculated as the standard deviation of the calculated
345 residuals.

346 ***National Herd Data.*** Fixed effects linear models were used to quantify the
347 association between genotype and milk production and fertility variables. Herd-year-
348 season contemporary group, proportion of Holstein genes and age at first calving were

349 included as fixed effects in the model where significant ($P < 0.1$). The association
350 between genetic merit and recalving within 365 d of first calving and recalving within
351 400 d of first calving was determined using generalized linear mixed models; herd-
352 year-season, age at first calving and proportion Holstein genes were included as
353 adjustment variables, where significant ($P < 0.1$).

354

355

RESULTS

356

Milk Production

358 Mean daily milk production and the parameters for the Wilmink exponential function
359 are summarized in Table 2. During the full lactation, genotype had a significant effect
360 ($P < 0.05$) on milk yield, with Fert+ cows having greater milk yield (19.5 ± 0.02 kg/d)
361 than Fert- cows (18.7 ± 0.02 kg/d). Milk solids yield tended to be greater ($P = 0.09$) in
362 Fert+ cows (Figure 1). Mean milk fat and lactose concentrations did not differ (both P
363 > 0.05) between the Fert+ and Fert- cows, but Fert- cows tended to have greater milk
364 protein concentration ($P = 0.07$).

365 The Wilmink function fit the daily milk yield data well, with a median R^2 value of
366 0.72. Fert+ cows tended to have a higher ($P = 0.08$) lactation curve (a parameter)
367 compared to Fert- cows (Figure 1). There was no effect of genotype or genotype by
368 parity interaction for either the rate of increase to peak milk yield (b parameter), or the
369 rate of decline from peak milk yield (c parameter). Days post calving when peak daily
370 milk yield occurred did not differ between genotypes (31.9 d and 29.0 d for the Fert+
371 and Fert- cows, respectively).

372

Reproduction and Fertility

374 The reproductive performance of the herd was monitored during yr 1 of the study on
375 36 first-parity cows. The calving period ranged from January 8 to April 14 with the
376 median calving date being February 2 and 9, for the Fert- and Fert+ groups,
377 respectively. The effect of genotype on reproductive performance is summarized in
378 Table 3. There was no effect of genotype on interval from calving to first service or
379 21 d submission rate, but the interval from calving to conception was significantly
380 shorter ($P < 0.05$) for Fert+ (85.6 ± 7.0 d) compared to Fert- (113.8 ± 7.8 d) cows.
381 Pregnancy rate to first and second service and the six wk in-calf rate tended to be
382 greater ($P = 0.08$ and $P = 0.09$, respectively) for the Fert+ cows. The number of
383 services per cow was fewer ($P < 0.05$) for the Fert+ compared to the Fert- cows over
384 the course of the breeding season. The proportion of cows establishing pregnancy
385 during the course of the breeding season is illustrated in Figure 2, and indicates the
386 difference between genotypes in the interval from mating start date to successful
387 pregnancy establishment (effect of genotype, $P < 0.05$).

388

389 ***Energy balance, DMI, BCS and BW,***

390 No difference between Fert+ and Fert- cows was observed for GDMI (12.4 and 12.1
391 kg/day, respectively) or TDMI (15.4 and 15.1 kg/d, respectively) across the 3
392 measurement timepoints in yr 1. In addition, no effect of genotype was observed on
393 mean daily energy balance at the DMI measurement timepoints (2.87 ± 0.23 vs. 3.09
394 ± 0.23 UFL/d, Fert+ and Fert-, respectively). Parity had a significant effect ($P <$
395 0.001) on GDMI in yr 2, being greater in parity 2 (17.5 ± 0.39 kg/day) than parity 1
396 (12.5 ± 0.24 kg/d) cows. No effect of genotype or genotype by parity interaction was
397 observed for energy balance or GDMI at the single measurement timepoint in yr 2.
398 The BCS and BW profiles of both genotypes are illustrated in Figure 3. No

399 differences between genotypes were observed for BW, but Fert+ cows had greater
400 BCS during early (+0.13 units, $P = 0.06$), mid (+0.1 units, $P < 0.05$) and late (+0.09
401 units, $P < 0.05$) lactation compared with Fert- cows (Table 4). There was a significant
402 genotype by wk interaction for BCS during the gestation/lactation cycle and early
403 lactation time periods. The difference between genotypes was greatest in first parity
404 cows at calving and during early lactation (both $P < 0.05$). The Fert+ cows tended to
405 have greater ($P = 0.07$) BCS at nadir, but no differences were observed for wk of
406 lactation at nadir, or changes in BCS from parturition to nadir and from nadir to end
407 of lactation.

408

409 *Circulating concentrations of blood metabolites and metabolic hormones*

410 No differences between genotypes were observed during the dry period (all $P >$
411 0.05) for plasma concentrations of metabolites (Table 5). There was a significant
412 genotype by parity interaction for plasma NEFA concentration ($P < 0.05$) during early
413 lactation; first parity Fert+ cows had greater circulating concentrations compared to
414 first parity Fert- cows, but no differences were observed in second parity cows.
415 Genotype had no effect ($P > 0.05$) on circulating concentrations of NEFA or glucose
416 during the gestation/lactation cycle. During early lactation, Fert+ cows had a greater
417 ($P < 0.01$) mean circulating concentrations of BHB compared with Fert- cows (0.340
418 vs. 0.276 mmol/L, respectively), but no differences were detected during mid- or late-
419 lactation. The effect of genotype on mean plasma concentrations of IGF-I and insulin
420 during the gestation/lactation cycle are illustrated in Figure 4. Fert+ cows had greater
421 ($P < 0.01$) mean circulating concentrations of IGF-I throughout the study period
422 compared to Fert- cows. There was no overall effect of genotype on mean circulating
423 concentrations of plasma insulin during the gestation/lactation cycle ($P > 0.05$);

424 however, mean plasma insulin concentrations were greater ($P < 0.05$) in Fert+ (1.71
425 $\mu\text{IU/ml}$) than Fert- cows (1.24 $\mu\text{IU/ml}$) during the first 4 wk of lactation.

426

427 *Analysis of National herd data*

428 Performance of first parity cows from the Irish national herd ($n = 7230$) that fit the
429 selection criteria of the Fert+/Fert- herd are summarized in Table 6. A significant ($P <$
430 0.01) association was observed between genotype and 305-day milk yield, with Fert+
431 cows having greater milk yield (5556 ± 11.0 kg) than Fert- cows (5503 ± 15.9 kg). No
432 differences ($P > 0.05$) were observed between genotypes for 305-day protein or fat
433 yield, but Fert- cows had greater mean milk fat ($P < 0.05$) and protein ($P < 0.001$)
434 concentration than Fert+ cows. Fert+ cows took 11.1 d less ($P < 0.001$) to recalve than
435 Fert- cows. The proportion of cows that re-calved within 365 d (+8%) and within 400
436 days (+13%) were greater for Fert+ compared with Fert- cows ($P < 0.001$).

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DISCUSSION

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440 This study clearly indicates that genetic merit for fertility traits has a profound effect
441 on phenotypic fertility performance. Other factors that are known to impact fertility
442 performance were similar for both groups (herd management, plane of nutrition,
443 proportion of Holstein genes and genetic merit for milk production traits).
444 Furthermore, production and fertility data for Fert+ and Fert- cows obtained from the
445 Irish national herd database substantiated the findings from the experimental herd.
446 Hence, this unique herd represents a powerful animal model for future fertility
447 research to elucidate the biological mechanisms underpinning differences in fertility
448 without being confounded by milk production.

449 The superior reproductive performance observed in the Fert+ cows confirms the
450 strong association between genetic merit for fertility traits and the subsequent
451 reproductive performance. In agreement with our findings, Horan et al. (2004)
452 suggested that cows of Holstein or New Zealand Friesian origin that had been selected
453 for superior fertility and survival exhibit superior reproductive performance.
454 Similarly, Coleman et al. (2009) reported differences in pregnancy rate to first service
455 and six wk in-calf rate between Holstein cows with a modest 2.46 d difference in
456 EBV for calving interval (12.24 d difference in the current study) but with similar
457 genetic merit for milk yield. These results confirm the contribution of poor genetic
458 merit for fertility traits to suboptimal fertility performance. Incorporation of fertility
459 traits in national breeding programmes can reverse these trends (Norman et al., 2009).

460 An antagonistic relationship between genetic merit for milk production and fertility
461 has been reported in lactating dairy cows (Evans et al., 2006a, Pryce et al., 2004).
462 Cows with high genetic merit for milk yield prioritise the partitioning of nutrients
463 towards milk production, rendering them susceptible to prolonged periods of NEB
464 and BCS loss, resulting in impaired reproductive function (Butler and Smith, 1989).
465 In the current study, genetic merit for milk yield was similar for both groups, but
466 Fert+ cows maintained higher daily milk yield throughout lactation compared with
467 Fert- cow. When daily milk output was expressed as milk solids yield, however, the
468 difference between genotypes was modest. These results suggest that on a grass-based
469 system, Fert+ cows were more capable of expressing their genetic potential for milk
470 yield. This concurs with Buckley et al. (2003), who reported a positive association
471 between phenotypic milk yield and reproductive performance when genetic merit for
472 milk yield and proportion of Holstein genes were included as adjustment variables in
473 the statistical model. There is, however, a lack of consensus within the literature on

474 the association between phenotypic milk yield and dairy cow fertility. No association
475 (Leblanc, 2010, Patton et al., 2007), a negative association (Nebel and McGilliard,
476 1993, Sakaguchi, 2011), and a positive association (Buckley et al., 2003, Mackey et
477 al., 2007) between phenotypic milk production and measures of reproductive
478 performance have been reported. Despite these seemingly conflicting results, early
479 lactation energy balance and associated BCS changes have been consistently
480 demonstrated to affect fertility performance (Roche et al., 2009).

481 Mobilisation of body reserves during early lactation is part of an orchestrated series
482 of adaptations in high yielding dairy cows to support mammary milk synthesis
483 (Bauman and Currie, 1980). The severity and duration of NEB during early lactation
484 has been reported to influence fertility (Butler and Smith, 1989). As a subjective
485 visual assessment of adipose tissue reserves, BCS has a strong association with NEB
486 and fertility (Butler, 2003). In the present study, early lactation trends in BCS
487 suggested that Fert+ cows maintained a higher threshold BCS level. This occurred
488 despite maintaining a higher milk yield throughout lactation. The difference in BCS
489 levels seen in early lactation were more pronounced during mid and late lactation,
490 when the Fert+ cows maintained a higher BCS level indicating superior energy status.
491 These findings concur with Berry et al. (2003), who reported that BCS during mid to
492 late lactation had the strongest genetic relationship with fertility. Roche et al. (2006)
493 concluded that cows with a strong genetic drive to produce milk maintained lower
494 BCS throughout lactation. The ability of the Fert+ cows to produce more milk while
495 also maintaining BCS at a higher threshold would suggest that differences in DMI
496 existed. Neither DMI nor calculated EB were different, however, during the 5
497 measurement periods throughout lactation. The earliest DMI measurement was
498 recorded on average 13 wk postpartum (for yr 1 and 2), and perhaps differences in

499 DMI existed before this measurement. Further work to measure daily intakes is
500 required to more closely examine DMI and EB relationships in this animal model.

501 The link between metabolic status and reproductive efficiency involves the complex
502 integration and regulation of endocrine and metabolic signals (Chagas et al., 2007).
503 Alterations in circulating concentrations of blood metabolites and metabolic hormones
504 such as IGF-I, insulin, glucose, NEFA and BHB have been demonstrated to play
505 important roles in the dialogue between nutritional status and the reproductive axis
506 (Butler, 2000, 2003, Leroy et al., 2008). In the current study, genetic merit for fertility
507 traits had no effect on circulating concentrations of glucose or NEFA, but plasma
508 concentrations of BHB were higher during early lactation in Fert+ cows. In support of
509 these findings, Patton et al. (2007) found concentrations of plasma NEFA and glucose
510 measured during the first 4 wk of lactation to have no association with calving to
511 ovulation interval or conception rate. A possible reason for the lack of association
512 between blood metabolite concentrations and measures of reproductive performance
513 in the current study is that levels of NEFA, BHB and glucose were within critical
514 thresholds for healthy cows. Outside these thresholds, the likely development of
515 postpartum metabolic disorders has been shown to have detrimental effects on fertility
516 (Nydam, 2009).

517 The uncoupling of the somatotropic axis in early lactation is a characteristic of high
518 producing cows, and occurs when the liver becomes unresponsive to growth hormone
519 (GH) and hepatic IGF-I output is reduced (Lucy, 2001). The observed differences in
520 circulating IGF-I concentrations in the current study indicate a genetic effect in the
521 regulation of the somatotropic axis. The ability of the Fert+ cows to maintain greater
522 circulating concentrations of IGF-I is consistent with a large body of evidence linking
523 increased IGF-I with improved reproductive outcomes (Taylor, 2004). Contrary to

524 previous findings, the observed differences in circulating plasma IGF-I concentrations
525 were independent of alterations in nutrition, Holstein ancestry, milk production or
526 genetic merit for milk production (Lucy et al., 2009), highlighting for the first time the
527 effect of genetic merit for fertility traits on key components of the somatotropic axis.
528 The precise mechanisms by which Fert+ cows maintained greater concentrations of
529 IGF-I warrants further examination.

530 Insulin plays a central role in the metabolism of body tissues, acting as an indicator
531 of energy status. In the present study plasma insulin concentrations were greater for
532 the Fert+ cows during the first 4 wk of lactation. In studies where early postpartum
533 dairy cows were subjected to a hyperinsulinemic-euglycemic clamp infusion, it was
534 shown that elevated circulating insulin concentrations directly effected dominant
535 follicle oestradiol output (Butler et al., 2004), and the recoupling of the somatotropic
536 axis through increasing hepatic GHR 1A and IGF-I mRNA expression (Butler et al.,
537 2003). Elevated plasma insulin concentrations as a result of dietary manipulation
538 during the first 50 to 100 d postpartum resulted in shorter intervals from calving to
539 ovulation and from calving to conception, and increased conception rate (Gong et al.,
540 2002, Gong, 2001). In vitro studies have demonstrated that increased insulin
541 concentrations stimulate granulosa cell proliferation and progesterone steroidogenesis
542 (Spicer and Echtenkamp, 1995). The early rise in plasma insulin concentrations
543 observed in Fert+ cows during the immediate postpartum period could be important in
544 the regulation of the somatotropic axis and reproductive events in the postpartum
545 period.

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CONCLUSIONS

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549 Genetic merit for fertility traits had a significant effect on reproductive efficiency in
550 the current study. The Fert+ group had improved calving to conception interval, fewer
551 services and conceived more quickly after breeding began than the Fert- group. The
552 observed differences in reproductive performance were independent of management,
553 plane of nutrition, proportion Holstein ancestry, genotypic and phenotypic milk yield,
554 and blood metabolite concentrations of NEFA and glucose. Fert+ cows maintained
555 greater BCS and circulating concentrations of IGF-I throughout lactation and had
556 greater circulating concentrations of insulin during the first 4 wk of lactation. This
557 supports the premise that animals with superior fertility are less reliant on body
558 reserves for milk synthesis, thus reducing the degree to which the somatotropic axis is
559 uncoupled. These results highlight the effect of genetic merit for fertility on
560 phenotypic reproductive performance, which may not necessarily be to the detriment
561 of milk production.

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REFERENCES

564

- 565 Bauman, D. E. and W. B. Currie. 1980. Partitioning of Nutrients During Pregnancy
566 and Lactation: A Review of Mechanisms Involving Homeostasis and Homeorhesis. *J*
567 *Dairy Sci* 63(9):1514-1529.
- 568 Berry, D. P., F. Buckley, P. Dillon, R. D. Evans, M. Rath, and R. F. Veerkamp. 2003.
569 Genetic parameters for body condition score, body weight, milk yield, and fertility
570 estimated using random regression models. *J Dairy Sci* 86(11):3704-3717.
- 571 Berry, D. P., L. Shalloo, A. R. Cromie, R. F. Veerkamp, P. Dillon, P. R. Amer, J. F.
572 Kearney, R. D. Evans and B. Wickham. 2007. The economic breeding index: a
573 generation on. Technical report to the Irish Cattle Breeding Federation, Co. Cork,
574 Ireland.
- 575 Buckley, F., P. Dillon, M. Rath, and R. F. Veerkamp. 2000. The relationship between
576 genetic merit for yield and live weight, condition score, and energy balance of spring
577 calving Holstein Friesian dairy cows on grass based systems of milk production. *J*
578 *Dairy Sci* 83(8):1878-1886.
- 579 Buckley, F., K. O'Sullivan, J. F. Mee, R. D. Evans, and P. Dillon. 2003. Relationships
580 among milk yield, body condition, cow weight, and reproduction in spring-calved
581 Holstein-Friesians. *J Dairy Sci* 86(7):2308-2319.

582 Butler, S. T., A. L. Marr, S. H. Pelton, R. P. Radcliff, M. C. Lucy, and W. R. Butler.
583 2003. Insulin restores GH responsiveness during lactation-induced negative energy
584 balance in dairy cattle: effects on expression of IGF-I and GH receptor 1A. *J*
585 *Endocrinol* 176(2):205-217.

586 Butler, S. T., S. H. Pelton, and W. R. Butler. 2004. Insulin increases 17 β -estradiol
587 production by the dominant follicle of the first postpartum follicle wave in dairy
588 cows. *Reproduction* 127(5):537-545.

589 Butler, W. R. 2000. Nutritional interactions with reproductive performance in dairy
590 cattle. *Animal Reproduction Science* 60-61:449-457.

591 Butler, W. R. 2003. Energy balance relationships with follicular development,
592 ovulation and fertility in postpartum dairy cows. *Livestock Production Science* 83(2-
593 3):211-218.

594 Butler, W. R. and R. D. Smith. 1989. Interrelationships between energy balance and
595 postpartum reproductive function in dairy cattle. *J Dairy Sci* 72(3):767-783.

596 Chagas, L. M., J. J. Bass, D. Blache, C. R. Burke, J. K. Kay, D. R. Lindsay, M. C.
597 Lucy, G. B. Martin, S. Meier, F. M. Rhodes, J. R. Roche, W. W. Thatcher, and R.
598 Webb. 2007. Invited Review: New Perspectives on the Roles of Nutrition and
599 Metabolic Priorities in the Subfertility of High-Producing Dairy Cows. *Journal of*
600 *Dairy Science* 90(9):4022-4032.

601 Coleman, J., K. M. Pierce, D. P. Berry, A. Brennan, and B. Horan. 2009. The
602 influence of genetic selection and feed system on the reproductive performance of
603 spring-calving dairy cows within future pasture-based production systems. *J. Dairy*
604 *Sci.* 92(10):5258-5269.

605 Dillon, P., S. Crosse, G. Stakelum and F. Flynn. 1995. The effect of calving date and
606 stocking rate on the performance of spring-calving dairy cows. *Grass & Forage*
607 *Science* 50(3):286-299.

608 Dillon, P. and G. Stakelum. 1989. Herbage and dosed alkanes as a grass management
609 technique for dairy cows. *Ir. J. Agric. Res.* 28:104 (Abstr.).

610 Edmonson, A. J., I. J. Lean, L. D. Weaver, T. Farver, and G. Webster. 1989. A Body
611 Condition Scoring Chart for Holstein Dairy Cows. *Journal of Dairy Science* 72(1):68-
612 78.

613 Enright, W. J., L. T. Chapin, W. M. Moseley, S. A. Zinn, M. B. Kamdar, L. F.
614 Krabill, and H. A. Tucker. 1989. Effects of infusions of various doses of bovine
615 growth hormone-releasing factor on blood hormones and metabolites in lactating
616 Holstein cows. *J Endocrinol* 122(3):671-679.

617 Evans, R. D., P. Dillon, F. Buckle, D. P. Berry, M. Wallace, V. Ducrocq, and D. J.
618 Garrick. 2006a. Trends in milk production, calving rate and survival of cows in 14
619 Irish dairy herds as a result of the introgression of Holstein-Friesian genes *Animal*
620 *Science* 82(1):423-433.

621 Evans, R. D., M. Wallace, L. Shalloo, D. J. Garrick, and P. Dillon. 2006b. Financial
622 implications of recent declines in reproduction and survival of Holstein-Friesian cows
623 in spring-calving Irish dairy herds. *Agricultural Systems* 89(1):165-183.

624 Gong, J. G., W. J. Lee, P. C. Garnsworthy, and R. Webb. 2002. Effect of dietary-
625 induced increases in circulating insulin concentrations during the early postpartum
626 period on reproductive function in dairy cows. *Reproduction* 123(3):419-427.

627 Gong, J. G., Troup, K.D, McCullough, E, Garnsworthy, P.C, Webb, R, Armstrong,
628 D.G. 2001. The effect of feeding a diet to increase circulating insulin concentrations
629 on reproductive performance in dairy cows. The Fourth International Conference on
630 Farm Animal Endocrinology T, Parma, Italy, 2001 October 7–10.

631 Horan, B., J. F. Mee, M. Rath, C. P. O, and P. Dillon. 2004. The effect of strain of
632 Holstein-Friesian cow and feeding system on reproductive performance in seasonal-
633 calving milk production systems. *Animal Science* 79(3):453-467.

634 Jarrige, J. 1989. INRAtion, 1989. V2.7. Microsoft computer program of ration
635 formulation for ruminant livestock. Publishers CNERTA, 26 Boulevard du Docteur
636 Petit Jean 21000, Dijon, France.

637 Leblanc, S. 2010. Assessing the Association of the Level of Milk Production with
638 Reproductive Performance in Dairy Cattle. *The Journal of Reproduction and*
639 *Development* 56(S):S1-S7.

640 Leroy, J. L. M. R., A. Van Soom, G. Opsomer, and P. E. J. Bols. 2008. The
641 consequences of metabolic changes in high-yielding dairy cows on oocyte and
642 embryo quality. *animal* 2(08):1120-1127.

643 Lucy, M. C. 2001. Reproductive loss in high-producing dairy cattle: where will it
644 end? *J Dairy Sci* 84(6):1277-1293.

645 Lucy, M. C., G. A. Verkerk, B. E. Whyte, K. A. Macdonald, L. Burton, R. T.
646 Cursons, J. R. Roche, and C. W. Holmes. 2009. Somatotropic axis components and
647 nutrient partitioning in genetically diverse dairy cows managed under different feed
648 allowances in a pasture system. *J. Dairy Sci.* 92(2):526-539.

649 Mackey, D. R., A. W. Gordon, M. A. McCoy, M. Verner, and C. S. Mayne. 2007.
650 Associations between genetic merit for milk production and animal parameters and
651 the fertility performance of dairy cows. *animal* (01):29-43.

652 Mayes, R. W., C. S. Lamb, and P. M. Colgrove. 1986. The use of dosed and herbage
653 n-alkanes as markers for the determination of herbage intake. *The Journal of*
654 *Agricultural Science* 107(01):161-170.

655 McCarthy, S., B. Horan, P. Dillon, P. O'Connor, M. Rath, and L. Shalloo. 2007.
656 Economic Comparison of Divergent Strains of Holstein-Friesian Cows in Various
657 Pasture-Based Production Systems. *J. Dairy Sci.* 90(3):1493-1505.

658 Nebel, R. L. and M. L. McGilliard. 1993. Interactions of High Milk Yield and
659 Reproductive Performance in Dairy Cows. *J. Dairy Sci.* 76(10):3257-3268.

660 Norman, H. D., J. R. Wright, S. M. Hubbard, R. H. Miller, and J. L. Hutchison. 2009.
661 Reproductive status of Holstein and Jersey cows in the United States. *J. Dairy Sci.*
662 92(7):3517-3528.

663 Nydam, D. V., Ospina, P.A. , Stokol, T., Overton, T.R. 2009. Evaluation of the effect
664 of Non-Esterified Fatty Acids (NEFA) and B-Hydroxybutyrate (BHB) concentrations
665 on health, reproduction and production in transition dairy cattle from the northeast
666 USA. *Proceedings of the Cornell Nutrition Conference for Feed Manufacturers 71st*
667 *Meeting, DoubleTree Hotel Syracuse, East Syracuse, New York:97 - 103.*

668 Olori, V. E., and P. J. B. Galesloot. . 1999. Projection of partial lactation records and
669 calculation of 305-day yields in the Republic of Ireland. *Interbull Bull* 22::149-154.

670 Patton, J., D. A. Kenny, S. McNamara, J. F. Mee, F. P. O'Mara, M. G. Diskin, and J.
671 J. Murphy. 2007. Relationships Among Milk Production, Energy Balance, Plasma
672 Analytes, and Reproduction in Holstein-Friesian Cows. *J. Dairy Sci.* 90(2):649-658.

673 Plaizier, J. C. B., G. J. King, J. C. M. Dekkers, and K. Lissemore. 1997. Estimation of
674 Economic Values of Indices for Reproductive Performance in Dairy Herds Using
675 Computer Simulation. *Journal of Dairy Science* 80(11):2775-2783.

676 Pryce, J. E., M. D. Royal, P. C. Garnsworthy, and I. L. Mao. 2004. Fertility in the
677 high-producing dairy cow. *Livestock Production Science* 86(1-3):125-135.

678 Roche, J. R., D. P. Berry, and E. S. Kolver. 2006. Holstein-Friesian Strain and Feed
679 Effects on Milk Production, Body Weight, and Body Condition Score Profiles in
680 Grazing Dairy Cows. *Journal of Dairy Science* 89(9):3532-3543.

681 Roche, J. R., N. C. Friggens, J. K. Kay, M. W. Fisher, K. J. Stafford, and D. P. Berry.
682 2009. Invited review: Body condition score and its association with dairy cow
683 productivity, health, and welfare. *Journal of Dairy Science* 92(12):5769-5801.
684 Royal, M. D., A. O. Darwash, Flint, A.P.F., Webb, R., Woolliams,, and G. E. J.A. and
685 Lamming. 2000. Declining fertility in dairy cattle: changes in traditional and
686 endocrine parameters of fertility. *Animal Science* 70.:487-501.
687 Sakaguchi, M. 2011. Practical Aspects of the Fertility of Dairy Cattle. *The Journal of*
688 *Reproduction and Development* 57(1):17-33.
689 Spicer, L. J. and S. E. Echtenkamp. 1995. The ovarian insulin and insulin-like growth
690 factor system with an emphasis on domestic animals. *Domestic Animal*
691 *Endocrinology* 12(3):223-245.
692 Taylor, V. J., Kebreab, E., Beever, D. E., Mills, J., Beever, D. E., Wathes, D. C. 2004.
693 Physiological adaptations to milk production that affect the fertility of high yielding
694 dairy cows. *British Society of Animal Science* 29:37-71.
695 Veerkamp, R. F., P. Dillon, E. Kelly, A. R. Cromie, and A. F. Groen. 2002. Dairy
696 cattle breeding objectives combining yield, survival and calving interval for pasture-
697 based systems in Ireland under different milk quota scenarios. *Livestock Production*
698 *Science* 76(1-2):137-151.
699 Washburn, S. P., W. J. Silvia, C. H. Brown, B. T. McDaniel, and A. J. McAllister.
700 2002. Trends in Reproductive Performance in Southeastern Holstein and Jersey DHI
701 Herds. *Journal of Dairy Science* 85(1):244-251.
702 Wilmink, J. B. M. 1987. Adjustment of lactation yield for age at calving in relation to
703 level of production. *Livestock Production Science* 16(4):321-334.
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TABLES AND FIGURES

Table 1. The mean estimated breeding value¹ (and SD) for the 2 groups of Holstein cows studied based on their milk production, individual calving interval, sire calving interval, and maternal grand sire calving interval.

Yr Group	Yr 1		Yr 2 ²	
	Fert+	Fert-	Fert+	Fert-
Number of animals	18	18	23	21
Holstein (%)	92 (5.6)	93 (6.3)	91(6.0)	93 (6.2)
Milk (kg)	+468 (161.8)	+526 (153.8)	+462 (133.4)	+478 (145.2)
Fat (kg)	+18.8 (7.96)	+19.2 (7.52)	+22 (7.34)	+17 (6.86)
Protein (kg)	+18.4 (6.36)	+20.8 (6.42)	+18.8 (5.52)	+17.8 (6.1)
Fat (g/kg)	+0.032 (0.1112)	-0.014 (0.1464)	+0.088 (0.112)	-0.016 (0.133)
Protein (g/kg)	+0.062 (0.0542)	+0.068 (0.0922)	+0.072 (0.046)	+0.042 (0.0855)
Survival (%)	+3.2 (0.88)	+0.2 (1.08)	+3.2 (0.78)	-0.2 (1.16)
Calving interval (d)	-6.4 (1.584)	+5.86 (3.434)	-6.36 (1.41)	+5.86 (2.88)
Sire calving interval (d)	-8.94 (4.336)	+8.12 (3.368)	-9.06 (3.6)	+7.3 (3.044)
Maternal grandsire calving interval (d)	-5.22 (2.54)	+6.48 (3.414)	-5.44 (2.496)	+7.92 (4.992)

¹All PTA's were obtained from the Autumn 2008 and autumn 2007 official dairy evaluations published by the Irish Cattle Breeding Federation and multiplied by two to convert to EBV's.

²In yr 2, the Fert+ group comprised of 15 second and 8 first parity cows, the Fert- group comprised of 13 second and 8 first parity cows

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Table 2. The effect of genetic merit for fertility traits on mean daily milk production performance over the complete lactation.

Variable	Genotype		SED ¹	P-Value		
	Fert+	Fert-		Geno	Geno × wk	Geno × parity
Number of animal records	41	39				
Milk Yield (kg/d)	19.54	18.69	0.39	0.02	0.2	0.2
Protein (g/kg milk)	33.2	33.6	0.02	0.07	0.06	0.09
Fat (g/kg milk)	40.7	41.3	0.05	0.3	0.2	< 0.001
Lactose (g/kg milk)	46.7	46.6	0.01	0.4	0.9	0.3
Milk solids ² (kg)	1.43	1.39	0.02	0.09	0.7	0.3
Wilmink parameters						
<i>a</i> (peak milk yield)	28.79	27.34	0.91	0.08	–	0.4
<i>b</i> (rate of increase to peak milk yield)	-7.13	-5.83	1.66	0.4	–	0.9
<i>c</i> (rate of decline from peak milk yield)	-0.061	-0.060	0.0038	0.7	–	0.9
DIM to peak ³ (day)	31.9	29.0	5.14	0.5	–	0.8

¹SED =pooled standard error of the difference
²=combined yield of fat and protein
³=The day when Wilmink parameter *a* was estimated

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Table 3. The effect of genetic merit for fertility traits on reproductive performance during a 20 wk breeding season

Variable	Genotype		SED ¹	P-Value
	Fert+	Fert-		
Number of animal records	18	18		
Mean calving date at 1 st parturition (d)	Feb-15	Feb-09	7.71	0.5
Calving to first service interval (d)	74.2	80.1	7.00	0.4
Calving to conception interval (d)	85.6	113.8	10.69	0.01
Number of services per cow	1.78	2.83	0.51	0.05
Number of services per pregnancy	1.44	2.23	0.41	0.06
21-d submission rate (%)	83.3	72.2	-	0.7
Conception rate to first service (%)	55.6	33.3	-	0.3
Pregnancy rate to first and second service (%)	83.3	50	-	0.08
42-d pregnancy rate (%)	72.2	41.2	-	0.09
Late embryo mortality (%)	0	11.1	-	0.5
Overall pregnancy rate (%)	88.9	72.2	-	0.4

¹SED =pooled standard error of the difference

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Table 4. The effect of genetic merit for fertility traits on mean body condition score (BCS) and bodyweight (BW) variables.

Variable	Genotype			P-Value		
	Fert+	Fert-	SED ¹	Geno	Geno × wk	Geno × parity
Number of animals records	41	39				
Mean BW (kg)	509.7	515.8	8.96	0.5	0.1	0.4
Mean BCS (units)	2.96	2.88	0.065	0.2	0.04	0.07
BCS early lactation ² (units)	3.01	2.88	0.076	0.06	0.03	0.02
BCS mid lactation ² (units)	2.84	2.74	0.049	0.03	0.9	0.2
BCS late lactation ² (units)	2.82	2.73	0.043	0.02	0.7	0.2
BCS at calving ³ (units)	3.65	3.55	0.097	0.3		0.003
BCS at nadir (units)	2.63	2.53	0.06	0.07		0.2
Wk of nadir (wk)	10.7	10.2	0.87	0.6		0.5
BCS change (units)						
Calving to nadir	1.02	0.98	0.09	0.8		0.08
Nadir to drying off	0.25	0.37	0.12	0.4		0.9

¹SED =pooled standard error of the mean

² Early lactation = week 1 to 12 post partum, Mid lactation = week 13 to 28 post partum, Late lactation = week 29 to 42.

³ BCS at calving was the BCS measured on the week of calving or less than 2 wk prior to calving

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Table 5. The effect of genetic merit for fertility traits on mean circulating metabolite concentrations¹.

Variable	Genotype		P-Value		
	Fert+	Fert-	Geno Geno	Geno × wk	Geno × parity
Dry period (wk -3-0)					
NEFA (mmol/L)	0.157 (0.119, 0.206)	0.155 (0.119, 0.204)	0.9	0.8	0.07
BHB (mmol/L)	0.231 (0.188, 0.281)	0.190 (0.154, 0.232)	0.2	0.9	0.06
Glucose (mmol/L)	2.84 (2.51, 3.16)	2.77 (2.44, 3.10)	0.8	0.4	0.3
Early lactation (wk 1-12)					
NEFA (mmol/L)	0.341 (0.290, 0.389)	0.320 (0.269, 0.379)	0.9	0.9	0.06
BHB (mmol/L)	0.340 (0.304, 0.379)	0.276 (0.243, 0.312)	0.009	0.2	0.3
Glucose (mmol/L)	2.68 (2.49, 2.88)	2.65 (2.44, 2.86)	0.8	0.5	0.06
Mid lactation (wk 13-28)					
NEFA (mmol/L)	0.108 (0.094, 0.124)	0.109 (0.094, 0.127)	0.9	0.9	0.08
BHB (mmol/L)	0.275 (0.243, 0.31)	0.247 (0.216, 0.282)	0.2	0.7	0.4
Glucose (mmol/L)	2.57 (2.37, 2.77)	2.59 (2.37, 2.8)	0.9	0.7	0.1
Late lactation (wk 29-42)					
NEFA (mmol/L)	0.077 (0.067, 0.088)	0.068 (0.058, 0.080)	0.2	0.06	0.3
BHB (mmol/L)	0.304 (0.264, 0.349)	0.264 (0.223, 0.312)	0.2	0.4	0.3
Glucose (mmol/L)	2.58 (2.36, 2.80)	2.49 (2.24, 2.74)	0.6	0.5	0.2

¹Geometric mean (95% CI)

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Table 6. The effect of genetic merit for fertility traits on milk production and reproductive performance within the Irish national herd during the years 2006 to 2010

Variable	Genotype		SED ¹	P-Value
	Fert+	Fert-		
Milk production²				
Number of animal records	4859	2371		
305d milk yield (kg)	5556	5503	19.3	0.007
305d protein yield (kg)	188.8	189.2	0.61	0.5
305d fat yield (kg)	219.4	218.3	0.75	0.1
Protein (g/kg milk)	34.1	34.5	0.01	< 0.001
Fat (g/kg milk)	39.9	40.1	0.01	0.04
Fat to protein ratio	1.17	1.16	0.003	0.003
Reproductive performance³				
Number of animal records	2436	1388		
EBV for Calving Interval (d) ⁴	-5.6 (1.21)	5.2 (1.44)		
Calving interval (d)	391.6	402.6	2.65	< 0.001
Recalve within 365 days (%)	41	33	1.6	< 0.001
Recalve within 400 days (%)	77	64	1.6	< 0.001

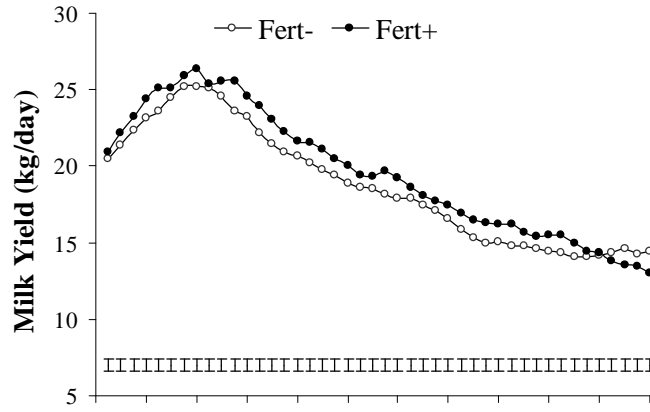
¹SED =pooled standard error of the difference

²Milk production data available for 2006-2010 inclusive

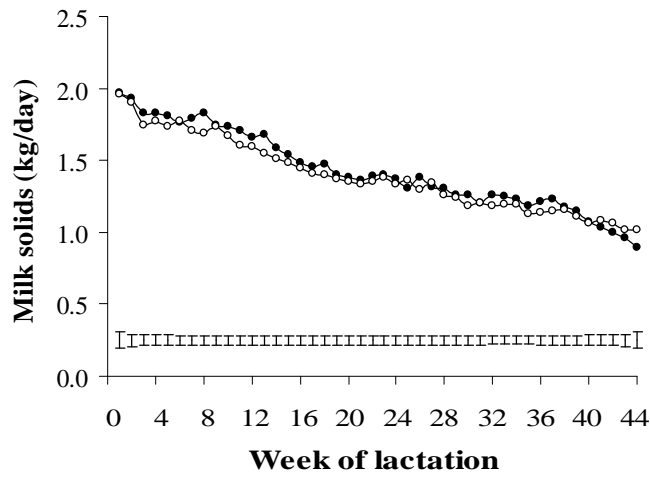
³Reproductive performance data available for 2006-2009 inclusive

⁴Standard deviation in parenthesis

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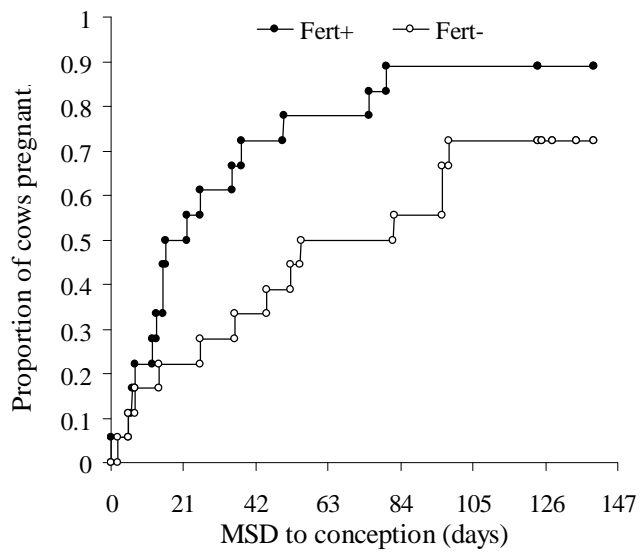
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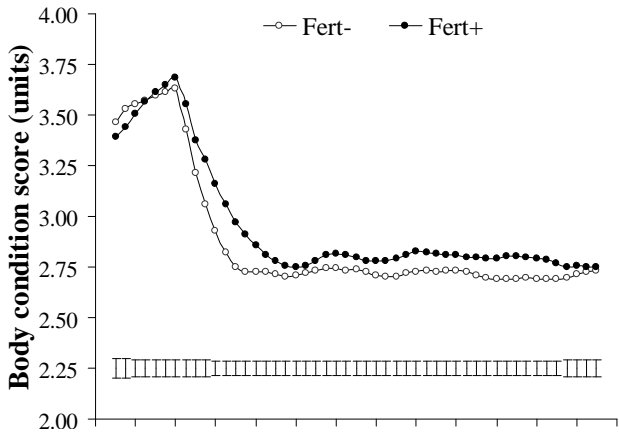
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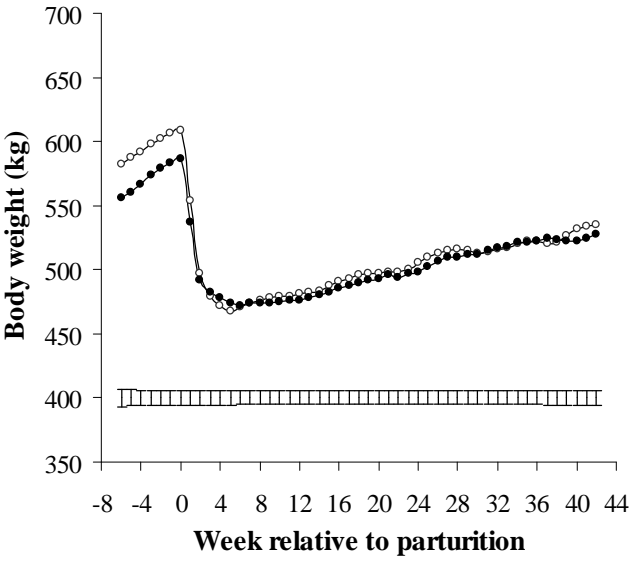
Figure 1. Cummins



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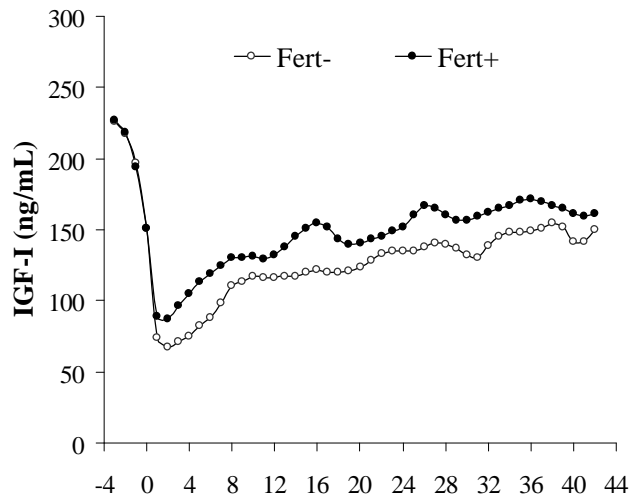


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Figure 3. Cummins



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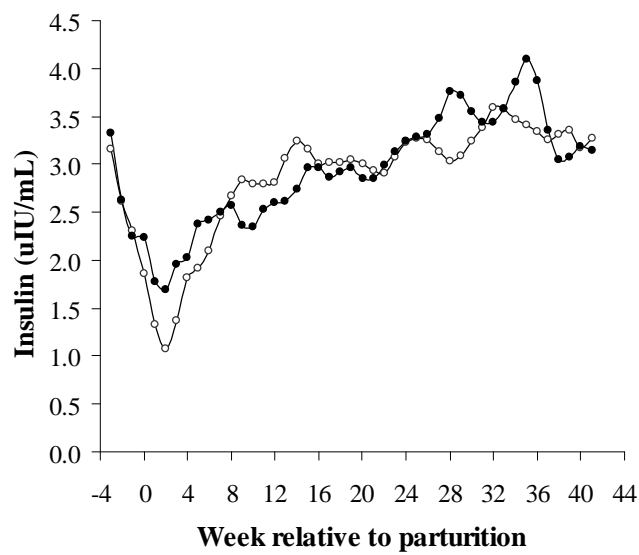


Figure 4. Cummins

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959 **Figure 1.** The mean daily milk yield and milk solid profiles of the Fert+ and Fert- cows during the
960 full lactation. Vertical bars indicate the pooled standard error of the difference
961 Top and middle panels: Mean daily milk yield was greater ($P < 0.05$) in Fert+ cows (SED = 0.39
962 kg/day) than in Fert- cows. No genotype \times wk or genotype \times parity effects were detected. Mean milk
963 solids yield tended to be greater ($P = 0.094$) in Fert+ cows (SED = 0.02), while no effect of genotype \times
964 wk or genotype \times parity interaction was observed.

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966 **Figure 2.** The interval in days from mating start date (MSD) to conception for Fert+ and Fert- cows.
967 The y axis represents the proportion of animals pregnant. Fert+ cows conceived at a significantly
968 greater rate than the Fert- cows ($P < 0.05$). The median number of days (95% confidence intervals in
969 parentheses) for 50% of the Fert+ cows to conceive was 19 days (13, 38 days), compared to 68.5 days
970 (36, 98 days) for 50% of the Fert- cows to conceive.

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972 **Figure 3.** The mean body condition score (BCS) and Body weight profiles of the Fert+ and Fert-
973 cows. Vertical bars indicate the pooled standard error of the difference
974 Top panel: No genotype effect was observed for mean BCS ($P = 0.207$, SED = 0.065 units) during the
975 gestation/lactation cycle. There was a significant genotype \times wk interaction ($P < 0.05$) for BCS, with
976 the interaction of genotype \times parity tending towards significance ($P = 0.072$).
977 Bottom panel: No genotype, genotype \times wk or genotype \times parity effect was detected for BW (SED =
978 8.96 kg).

979
980 **Figure 4.** Circulating concentrations of IGF-I and insulin during the gestation/lactation cycle of
981 Fert+ and Fert- cows.

982 Top panel: Plasma IGF-I concentrations were significantly higher in Fert+ cows ($P < 0.01$) throughout
983 the study period. The mean (95% confidence intervals in parentheses) circulating IGF-I concentration
984 were 148.3 (139.2, 157.6) and 128.2 (119.1, 137.7) for Fert+ and Fert-, respectively. No genotype \times wk
985 or genotype \times parity interactions were observed for IGF-I.

986 Bottom panel: No effect of genotype or genotype \times parity interaction was observed for plasma insulin
987 concentrations ($P > 0.05$). There was a significant genotype \times wk interaction ($P < 0.05$) for insulin
988 during the study period. The mean (95% confidence intervals in parentheses) circulating insulin
989 concentration were 2.89 (2.71, 3.07) and 2.83 (2.64, 3.03) for Fert+ and Fert-, respectively.

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