MASTER INTERUNIVERSITARIO EN MEJORA GENÉTICA ANIMAL Y BIOTECNOLOGÍA DE LA REPRODUCCIÓN

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MITIGATION OF GREENHOUSE GASES IN LIVESTOCK VIA GENETIC SELECTION:

INCORPORATION OF METHANE EMISSIONS INTO THE BREEDING GOAL IN DAIRY CATTLE UNDER DIFFERENT SCENARIOS

Master Thesis

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Albert Einstein

"We shall require a substantially new manner of thinking if mankind is to survive."

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The results obtained in this thesis have led to **two oral communications**:

- In the National Meeting of Animal Breeding at the University of Leon , Spain:

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Abstract

The objective of this study was to analyze the impact of the incorporation of enteric methane (CH_4) into the breeding objective of dairy cattle in Spain, and to evaluate both genetic and economic response of traits in the selection index under foreseen scenario, that aims to reduce the carbon footprint of enteric CH_4 in dairy cattle in terms of lower CH_4 emissions: i. Current situation as benchmark (without putting an economic value on CH_4 emissions); ii. Penalization of CH_4 emissions through a carbon tax; iii. CH_4 emissions in a carbon quota; and iv. Including CH_4 as a net energy loss cost.

In order to include CH_4 emission as a breeding goal, one of the first tasks is to define the economic importance of each trait included in the aggregate genotype and his economic weighting in the current and planned situation. Thus, economic value (EV) of CH_4 emissions, which represents a loss of dietary energy in ruminants and is an important contributor to global warming, needed to be estimated. To achieve this, first we developed a bio economic model to derive the EV for production and CH_4 traits. Then we estimated variance components for CH_4 as well as its genetic correlations with other traits in the Total Merit Index (ICO). Finally, we calculated the genetic and economic responses to selection for each scenario that has been tested.

The estimated EVs are 0.01, 1.94, and 4.48 (€/kg) for milk volume, milk fat and milk protein yields in scenario 1. For CH_4, the economic values are calculated as -1.21, -9.32 and -0.67 (€/kg) for scenarios 2, 3 and 4, respectively.

The estimated heritability for CH_4 is 0.38 ± 0.16, showing that CH_4 is a heritable trait in dairy cattle. The genetic correlations between CH_4 and the traits in the ICO, and between CH_4 and the ICO are generally low and negatives, suggesting that has been selecting for better efficiency that leads to lower CH_4 emissions from lactating cows, and that more profitable cows produce less CH_4.

All of the studied indices resulted in a favorable response in overall CH_4 emissions ranging from a reduction of -0.51 kg/cow/year with the current index, to -0.70, -0.86, and -2.41 kg/cow/annum respectively for the three environmental indices : CO_2 tax, CH_4 quota and net energy loss cost. On the other hand, the incorporation of CH_4 to the baseline index, generates an increase in benefit of 38.35 €/cow/yr. this benefit was negligibly changed in the case of CO_2 tax and net energy loss scenarios, whilst in the situation of CH_4 quota, the overall economic response of the index falls to 33.87 €/cow/yr (-21%).

The sensitivity analysis considering the variation if different key parameters when CH_4 was included into the breeding goal under each scenario, showed that both the total benefit and genetic responses are insensitive to the changes. Except for the case of CH_4 quota, the genetic responses are highly sensitive when the economic weights and genetic correlations of CH_4 with other traits were varied.

This study showed that there is a potential in mitigating CH_4 emissions by genetic selection through the inclusion of CH_4 in selection objectives of dairy cattle while remaining profitable. Its inclusion in selection indices, will allow to select for low emitting and more efficient cows.

Keywords: Holstein, methane, bio economic model, selection index.

Resumen

El objetivo de este estudio fue analizar el impacto de la incorporación del metano entérico (CH_4) en el objetivo de selección del ganado lechero en España, y evaluar la respuesta genética y económica de los caracteres en el índice de selección en diferentes escenario, que apunta a reducir la huella de carbono del CH_4 entérico en el ganado lechero en términos de menor emisiones de CH_4: i. Situación actual como referencia (sin poner un valor económico a las emisiones de CH_4); ii. Penalización de las emisiones de CH_4 a través de un impuesto al carbon (CO_2); iii. Emisiones de CH_4 en una situacion de cuota de carbono; y iv. Incluyendo CH_4 como una pérdida de energía neta para la vaca.

Para incluir la emisión de CH_4 como objetivo de selección, una de las primeras tareas es definir la importancia económica de cada carácter incluido en el genotipo agregado y su ponderación económica en la situación actual y la situación planificada. Por lo tanto, es necesario estimar el valor económico (VE) de las emisiones de CH_4, que representa una pérdida de energía alimentaria en los rumiantes y es un importante contribuyente al calentamiento global. Para lograr esto, primero desarrollamos un modelo bio-económico para derivar el VE para los caracteres de producción y de CH_4. Luego estimamos los componentes de varianza para CH_4 así como sus correlaciones genéticas con otros caracteres en el Índice de mérito genético (ICO). Finalmente, calculamos las respuestas genéticas y económicas a la selección para cada escenario que ha sido estudiado.

Los VE estimados son 0,01, 1,94 y 4,48 (€ / kg) para el volumen de leche, el rendimiento de la grasa y el rendimiento de la proteína en el escenario 1. Para CH_4, los valores económicos se calcularon como -1,21, -9,32 y -0,67 (€ / kg) para los escenarios 2, 3 y 4, respectivamente.

La heredabilidad estimada para CH_4 es 0.38 ± 0.16, mostrando que CH_4 es un caraceter hereditario en el ganado lechero. Las correlaciones genéticas entre CH_4 y

los caracteres en el ICO, y entre CH_4 y el ICO son generalmente bajos y negativos, lo que significa que se ha seleccionado por una mejor eficiencia que conduce a menores emisiones de CH_4 de vacas lactantes, y que las vacas más rentables producen menos CH_4 .

Todos los índices estudiados dieron como resultado una respuesta favorable en las emisiones totales de CH_4 que van desde una reducción de -0.51 kg / vaca / año con el índice actual, a -0.70, -0.86 y -2.41 kg / vaca / año respectivamente para los tres Índices ambientales: impuesto sobre el CO_2, cuota de CH_4 y pérdida de energía neta. Por otro lado, la incorporación de CH_4 al índice base, genera un incremento en el beneficio de 38,35 € / vaca / año. Este beneficio se modificó de manera insignificante en el caso de los escenarios del impuesto sobre el CO_2 y el escenario de pérdida de energía neta, mientras que en la situación de la cuota de CH_4, la respuesta económica general del índice baja a 33,87 € / vaca / año (-21%).

El análisis de sensibilidad teniendo en cuenta la variación en diferentes parámetros claves cuando se incluyó CH_4 en el objetivo de seleccion en cada escenario, mostró que tanto el beneficio total como las respuestas genéticas son insensibles a los cambios. Excepto en el caso de la cuota de CH_4, en el que las respuestas genéticas son muy sensibles cuando se variaron los pesos económicos y las correlaciones genéticas de CH_4 con otros caracteres.

Este estudio mostró que existe un potencial para mitigar las emisiones de CH_4 mediante la selección genética a través de la inclusión de CH_4 en los objetivos de selección del ganado lechero mientras se mantiene rentable. Su inclusión en los índices de selección, permitirá seleccionar vacas de bajas emisiones y más eficientes.

Palabras clave: Holstein, metano, modelo bioeconómico, índice de selección.

Résumé

L'objectif de cette étude était d'analyser l'impact de l'incorporation du méthane entérique (CH_4) dans l'objectif de sélection des vaches laitières en Espagne et d'évaluer la réponse génétique et économique des caractères de l'indice de sélection dans quatres differents scenarios, visant à réduire l'empreinte du dioxide de carbone (CO_2) du CH_4 entérique chez les bovins laitiers en termes de réduction des émissions de CH_4: i. Situation actuelle comme référence (sans mettre une valeur économique sur les émissions de CH_4); ii. Pénalisation des émissions de CH_4 à travers d'une taxe sur le CO_2; iii. Émissions de CH_4 dans une situation de quota de CO_2; et iv. Incluir le CH_4 comme coût de perte d'énergie nette pour la vache.

Afin d'inclure les émissions de CH_4 en tant qu'objectif de selection, l'une des premières tâches consiste à définir l'importance économique de chaque caractère inclus dans le génotype agrégé et sa pondération économique dans la situation actuelle et prévue. Il fallait donc estimer la valeur économique (VE) des émissions de CH_4, qui représente une perte d'énergie alimentaire chez les ruminants et qui contribue de manière importante au réchauffement de la planète. Pour ce faire, nous avons d'abord mis au point un modèle bioéconomique permettant de dériver les VE pour les caractères de production et de CH_4. Ensuite, nous avons estimé les composantes de la variance pour le CH_4 ainsi que ses corrélations génétiques avec d'autres caracteres de l'indice de mérite total (ICO). Enfin, nous avons calculé les réponses génétiques et économiques à la sélection pour chaque scénario testé.

Les VE estimés sont 0,01, 1,94 et 4,48 (€ / kg) pour le volume de lait, lerendement de la matière grasse du lait et les protéines du lait dans le scénario 1. Pour le CH_4, les valeurs économiques sont : -1,21, -9,32 et -0,67 (€ / kg) pour les scénarios 2, 3 et 4, respectivement.

L'héritabilité estimée pour le CH_4 est de 0,38 ± 0,16, ce qui montre que le CH_4 est un trait héréditaire chez les vaches laitières. Les corrélations génétiques entre le

CH_4 et les caracteres de l'ICO et entre le CH_4 et l'ICO sont généralement faibles et négatives, ce qui montre qu'il a ete seleccionne pour une meilleure efficacité conduisant à une réduction des émissions de CH_4 chez les vaches laitieres .

Tous les indices étudiés ont genere une réponse favorable aux émissions totales de CH_4 allant d'une réduction de -0,51 kg / vache / année avec l'indice actuel à -0,70, -0,86 et -2,41 kg /vache/an respectivement pour les trois indices environnementaux: taxe sur le CO_2, quota de CH_4 et perte d'énergie nette. D'autre part, l'incorporation du CH_4 à l'indice de référence génère une augmentation du profit de 38,35 €/vache /an. Ce profit a été modifié de manière négligeable dans le cas des scénarios de taxe sur le CO_2 et de perte d'énergie nette, tandis que dans le cas du quota de CH_4, la réponse économique globale de l'indice est reduite à 33,87 € /vache/an (-21%).

L'analyse de sensibilité en considérant la variation de différents paramètres clés lorsque le CH_4 était inclus dans l'objectif de sélection pour chaque scénario, a montré que le bénéfice total et les réponses génétiques sont insensibles aux changements. À l'exception du cas de quota de CH_4, ou les réponses génétiques sont très sensibles lorsque les poids économiques et les corrélations génétiques du CH_4 avec d'autres caractères ont été variés.

Cette étude a montré qu'il était possible d'atténuer les émissions de CH_4 par la sélection génétique en incluant le CH_4 dans les objectifs de sélection des vaches laitières tout en restant rentable. Son inclusion dans les indices de sélection permettra de sélectionner des vaches à faible émission et plus efficaces.

Mots-clés: Holstein, méthane, modèle bioéconomique, indice de sélection.

Abbreviations

AI	Artificial Insemination
CH_4	Enteric Methane
CO_2	Carbone Dioxide
CONAFE	Spanish Holstein Association
DMI	Dry matter Intake
ECM	Energy Corrected Milk
FLI	Feet and Leg Index
FPCM	Fat and Protein Corrected Milk Production
GEBV	Genomic Breeding Value
GHG	Greenhouse Gas
GS	Genomic Selection
Gt eqCO_2	Gigatonnes Of Equivalent Carbon Dioxide
H	Hydrogen
ICO	Total official genetic index of CONAFE
LW	Live Weight
N_2O	Nitrous Oxide
RMP	Residual Methane production
SCC	Somatic Cell Count
SNP	Single Nucleotide Polymorphism
UCI	Udder Composite Index
UFL	Unité Fourragère Lait
YR	Year

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GENERAL INTRODUCTION

1. Introduction

Greenhouse gas (GHG) production from animals and their impact on smog forming emissions and climate change is a growing public concern worldwide. In the European Union, livestock farming is responsible for 13% of total greenhouse gas emissions (Leip et al., 2010), in particular for methane (CH_4) which has a global warming potential 28 times greater than CO_2 (Myhre et al., 2013). Cattle is considered as an important contributor to global CH_4 emissions. Methane from enteric fermentation is the main GHG coming from ruminants (approximately two-thirds), whereas manure handling contributes around one-third (Moss et al., 2000; Olesen et al., 2006). Furthermore, eructated methane production typically generates a net energy loss between 2% and 12% of net energy intake (Johnson and Johnson, 1995; Lassey et al., 1997; de Haas et al., 2011). Therefore, CH_4 is not only an important contributor to global warming, but also represents a loss of dietary energy in ruminants (Negussie et al., 2017). Consequently, reduction of enteric CH_4 emission in ruminants, and specifically in dairy cattle has become an important area of research, aligning with the commitment in the European Union to reduce its GHG emissions by 20% relative to 1990 by 2020 (de Haas et al., 2017).

There is a potential for adopting genetic and genomic selection to tackle CH_4 emissions from ruminants, given that several studies have reported that CH_4 emissions is a heritable and repeatable trait (Pickering et al., 2013a; Haque et al., 2015; Lassen and Løvendahl, 2016). Indeed, decreasing enteric CH_4 emissions from ruminants without altering animal production is desirable both as a strategy to reduce global GHG emissions as well as means of improving feed conversion efficiency, and consequently farm profitability (Martin et al., 2010; de Haas et al., 2017). In this sense, several studies showed that it is possible to decrease CH_4 emission by selecting more efficient cows (Hegarty et al., 2007; de Haas et al., 2011; Basarab et al., 2013).

At present, GHG emissions are not part of dairy cattle breeding goals in any country (de Haas et al., 2017). Despite of the global interest on reducing GHG emissions, worldwide policies do not tax emissions neither compensate its reduction, and hence there is no incentive to include CH_4 emissions in the breeding goal. In Spain, the breeding goals of the Holstein breed combines the quality and quantity of milk with functional characteristics such as longevity, fertility, morphological traits, and somatic cell count in a selection index, called ICO (Charfeddine and Pérez-Cabal, 2014). In order to include CH_4 emission, which represents 10.01% of total GHG emissions in Spain (Butnar and Llop, 2007), as a breeding goal, one of the first tasks is to define the economic importance of each trait included in the aggregate genotype and its economic weight in the future expected situation. Those economic values are derived from a formulated bio economic model (profit function) that translates the production and system conditions into a mathematical equation. The economic value represents the expected change in the benefits at modifying the trait in one unit. Proper weight of the traits considering the genetic and phenotypic (co)variances are expected to maximize the benefits of future generations.

Even if the selection for low CH_4 emission became a reality, there would be limited consensus on the choice of target phenotype. Some of the potential phenotypes include direct breathing methane measurements (production or concentration) or indirect selection through several indicator traits such as feed intake, milk spectral data, and rumen microbiota (de Haas et al., 2017). An individual measurement of CH_4 emission at the farm level is not an easy task (Garcia-Rodriguez and Gonzalez-Recio, 2017), but with the advent of genomic selection, including CH_4 emission as a breeding objective trait is attainable, even with a limited number of records (de Haas et al., 2017).

This thesis aims to highlight the importance of including greenhouse gas emissions in breeding goals of dairy cattle, specifically CH_4 emissions, by defining the economic weight of each trait included in the selection index and its correlative response under four different scenarios:

- Current ICO as benchmark (i.e. economic value of CH_4 emissions = 0), however, genetic correlations between CH_4 and ICO traits were incorporated, so that the correlative genetic response of CH_4 to selection on the index could be recorded.
- Penalization of CH_4 emissions through a carbon tax.
- CH_4 emissions in a carbon quota.
- Including CH_4 as a net energy loss cost.

This thesis encompasses three main parts:

In the first section, a bibliographic review gathers all the knowledge acquired in the literature about GHG emissions in relationship with livestock, specifically CH_4 emissions, methods of measuring CH_4, and its association with other traits in dairy cattle, as well as key concepts in bio-economic models and selection index theory. After describing the main and specific objectives of the study, the second section presents the material and methods used to develop the bio economic model and genetic analysis. Finally, the results obtained are presented and discussed. The conclusion highlights key results obtained and provide recommendations for further work in the future.

2. Importance of CH_4 emissions in ruminants and dairy cattle

Non-CO_2 emissions derive mostly from agriculture, predominated by N_2O emissions from agricultural soils and CH_4 emissions from livestock enteric fermentation, manure management, and emissions from rice paddies, totalling 5.0 – 5.8 gigatonnes of equivalent carbon dioxide (Gt eqCO_2) per year in 2010 (Figure **1**) (Edenhofer et al., 2014). CH_4 is mainly produced in the rumen, which is the site of conversion of hydrogen (H_2) and CO_2 to CH_4 by anaerobic archaeal microorganisms (de Haas et al., 2017). This is a complex process that provides energy to micro-organisms, and is called methanogenesis (Knapp et al., 2014):

$$CO_2 + 8H + 8e- \rightarrow CH_4 + 2H_2O$$

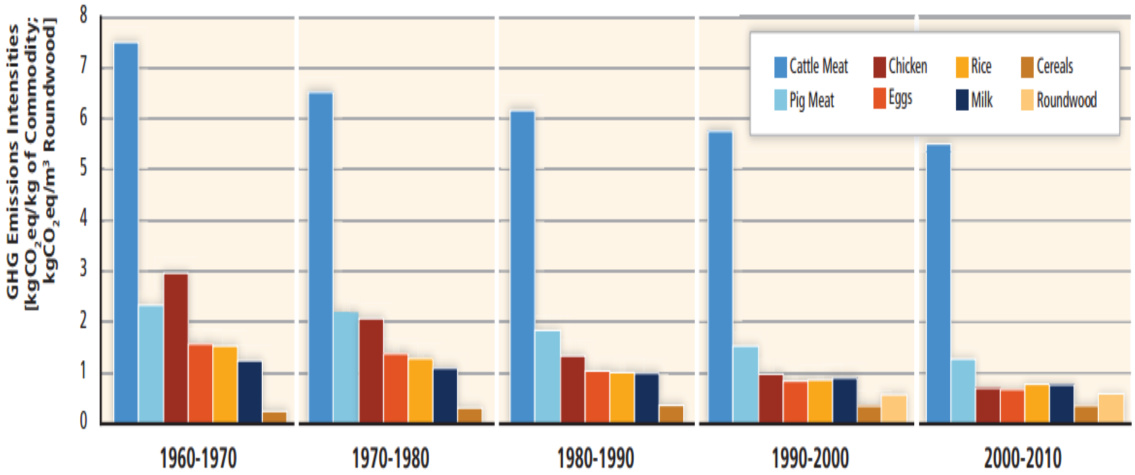
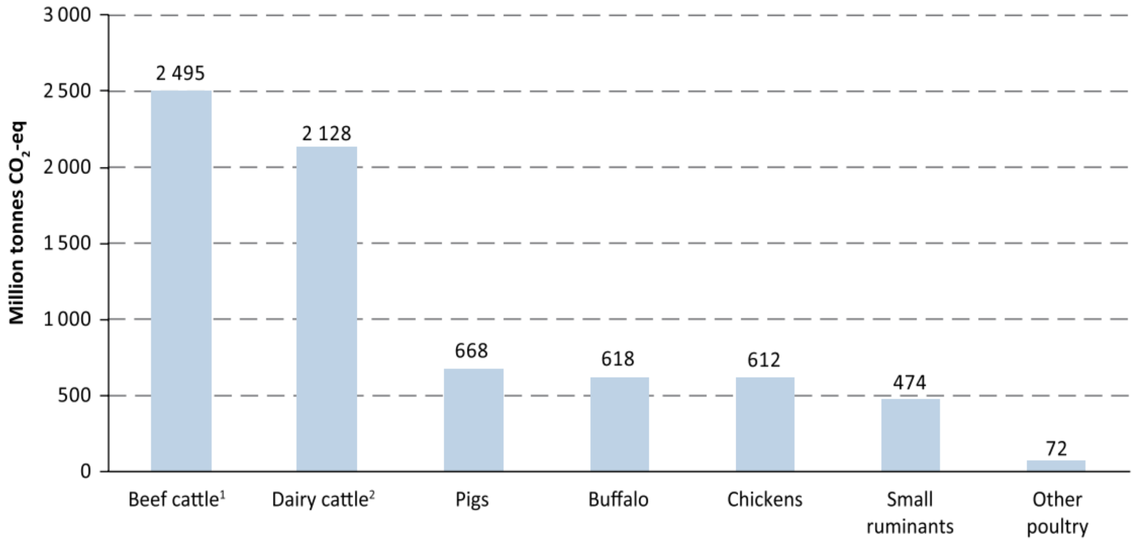


Figure 1. GHG emissions intensities of selected major Agriculture, Forestry and Other Land Use (AFOLU) commodities for decades 1960s – 2000s (Edenhofer et al., 2014)

(1) Cattle meat, defined as GHG (enteric fermentation + manure management of cattle, dairy and non-dairy) / meat produced;
(2) Pig meat, defined as GHG (enteric fermentation + manure management of swine, market and breeding) / meat produced;
(3) Chicken meat, defined as GHG (manure management of chickens) / meat produced;
(4) Milk, defined as GHG (enteric fermentation + manure management of cattle, dairy) / milk produced;
(5) Eggs, defined as GHG (manure management of chickens, layers) / egg produced;
(6) Rice, defined as GHG (rice cultivation) / rice produced;
(7) Cereals, defined as GHG (synthetic fertilizers) / cereals produced;
(8) Wood, defined as GHG (carbon loss from harvest) / roundwood produced.

This release of GHG emissions from animals, via enteric fermentation throughout normal digestive process, contributes to global warming (Broucek, 2014a). Among the GHG emitted by ruminants, enteric CH_4 is the most important contributor (Moss et al., 2000; de Haas et al., 2017), that increases GHG emissions and depletion of the ozone layer. Its global warming potential is 28 times than of CO_2 (Myhre et al., 2013). As shown in Figure 2, CH_4 is mainly produced by ruminants (dairy, beef, goats, and sheep) (Steinfeld et al., 2006; Broucek, 2014b). Between 87% and 90% of enteric CH_4 is produced in the rumen, and to a smaller extent (13% - 10%) in the large intestine (Broucek, 2014b). Ruminants can produce 250 to 500 liter of CH_4 per day (Johnson and Johnson, 1995; Lassey et al., 1997).



* Includes emissions attributed to edible products and to other goods and services, such as draught power and wool.
[1]Producing meat and non-edible outputs.
[2]Producing milk and meat as well as non-edible outputs.

Figure 2. Global estimates of emissions by specie (Gerber et al., 2013)

Broucek, (2014b) reviewed the literature sources and reported that cattle produce about 7 and 9 times as much CH_4 as sheep and goats, respectively. The GHGs emissions vary across life stages of cattle (Stackhouse et al., 2011). Lactating cows emit more CH_4 (353.8 g·d−1), compared to dry cows (268.8 g·d−1), and heifers (222.6 g·d−1). Within heifers, the amount of CH_4 emitted was higher in grazing fertilized pasture (222.6 g·day−1), than the unfertilized pasture (179.2 g·day−1) (Broucek, 2014b). The stage of lactation also influences the amount of CH_4 emission in dairy cattle, at peak lactation dairy cows produce about 430 g·d−1, down to 250 g·d−1 as milk yield decreases. Level of CH_4 emission depend on the differences in body weight, diet composition, feed intake, or milk yield. However, with the same diet and the same intake, some variation in emission of CH_4 remains between cows, suggesting a genetic effect and the existence of genetically low CH_4 emitting animals. Calves emit about 9.4 kg CH_4 per head and year (Broucek, 2014b).

Manure management is also a source of CH_4 in dairy farms. It contributes in a small percentage (10% of total emissions) (AEA Technology Environment, 1998) from deposition of feces of grazing animals, manure applied to field, and manure on barn floors. The predicted amount of CH_4 from feed intake was 6.4 kg/m^3 of slurry manure in storage. Globally CH_4 emission from manure management was 33.2 kg·head^{-1}·year^{-1} for dairy cattle (Broucek, 2014b).

3. Strategies for mitigating CH_4 in livestock

"Mitigation is a human intervention to reduce the sources or enhance the sinks of GHG" (Edenhofer et al., 2014). Actually, the high level of emissions produced by agriculture and livestock production opens up large opportunities for climate change mitigation through livestock actions (Steinfeld et al., 2006). In this sense, Herrero et al., (2016) argues that up to 50% of the global mitigation potential of the agriculture, land-use and forestry sector could be represented by the livestock sector. According to Johnson and Johnson, (1995), it is possible to reduce CH_4 emissions while maintaining or enhancing productivity, by improving diet quality, removing nutrient deficiencies, utilizing growth promotants appropriately and by selecting more efficient genotypes. National Livestock Methane Program, (2015) published different strategies for lowering CH_4 emissions in Australia. These strategies include the use of supplements, forages and genetic improvement by selecting low emitting animals. Developing strategies of reducing CH_4 production, quantifying cattle emission must be possible under a wide range of conditions (Johnson and Johnson, 1995). Figure **3** summarizes the effectiveness of the different practices for raising productivity and lowering CH_4, as well as an indicative cost for continued research into each. Despite the lower potential of genetic selection compared to other strategies, it is a low cost strategy that accumulates across the generations. Selective breeding for reduced CH_4 emissions, without lowering productivity, could represent a sustainable reduction in CH_4 emissions given selection pressure is maintained (Pickering et al., 2013a).

Nonetheless, it is not clear how to implement this strategy in the field. For instance, the increase in longevity and fertility in dairy systems, jointly with a reduction of milk volume (i.e., increase of fat and protein content), live weight, dry matter intake (DMI) and somatic cell count (SCC) can improve the net revenue and decreases the GHG emissions per cow and per unit product (Bell et al., 2013; Amer et al., 2017; Özkan Gülzari et al., 2018). However, these studies do not clarify whether total methane production (non per unit of product) would also decrease. Wall et al., (2010) proposed three routes that could reduce CH_4 emissions via genetic improvement by selection: (i) improving productivity and efficiency (e.g. residual feed intake, longevity), (ii) reducing wastage in the farming system, by lowering the age at first calving for example, which is considered as a paying off of the energy consumed and CH_4 emitted, and (iii) directly selecting on emissions, when the CH_4 or a correlated trait to it is measurable. Knapp et al., (2014), reviewed some strategies based on genetic selection, management of feeding and nutrition and rumen modifiers, then summarized the potential of reduction in CH_4 per energy-corrected milk (ECM), this potential ranges from 9 to 19% (Figure **5**).

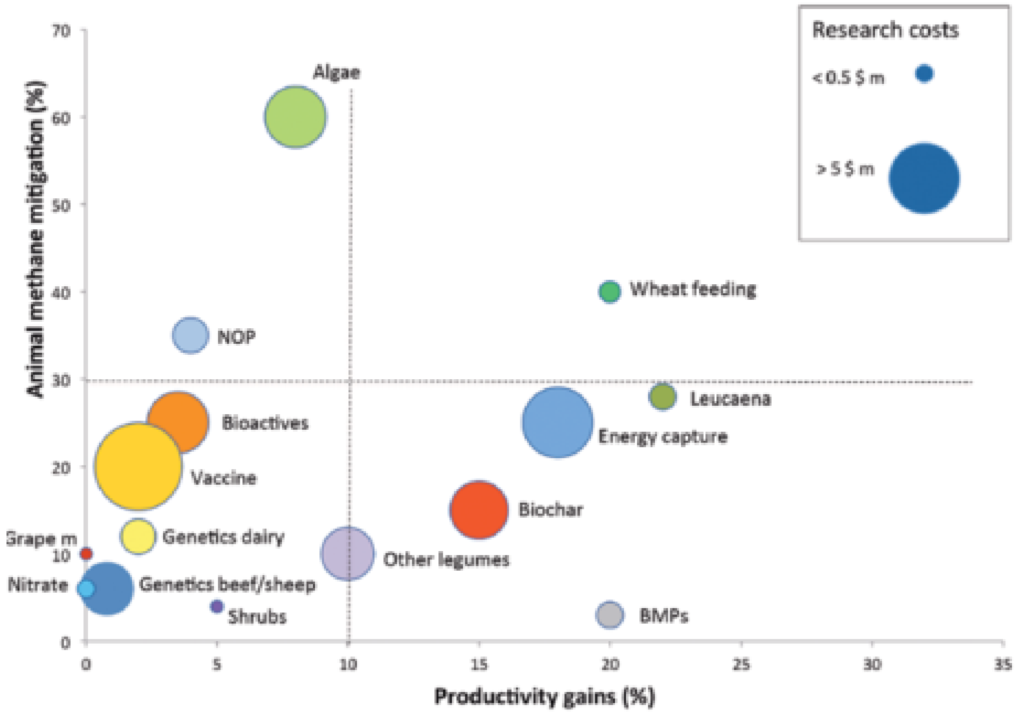


Figure 3. summary of effectiveness of CH_4 reduction practices (National Livestock Methane Program, 2015)

One way to improve feed efficiency (producing at the same level, with lower feed intake) is to minimize the energy losses that occur during the fermentation process and digestion of the animal (Garcia-Rodriguez and Gonzalez-Recio, 2017). Hegarty et al., (2007) conducted an experiment that consisted in selecting among 76 Angus steers chosen from divergent breed line for residual feed intake (RFI). Their results showed that use of RFI for improving livestock efficiency would also decrease enteric CH_4 emissions without affecting animal production (Figure 4). Furthermore, It had been approximated that there is potential for reduction in emissions of 8% per year after 20 years when selecting for low CH_4 emissions and for improved productivity (National Livestock Methane Program, 2015).

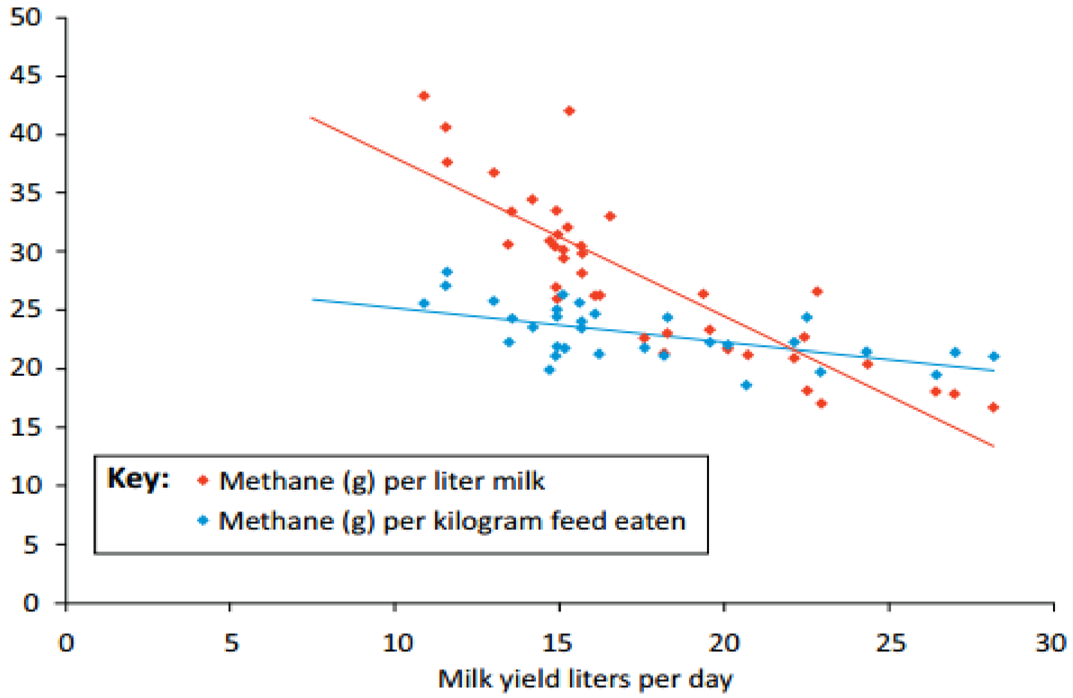


Figure 4. Selection for increased efficiency of production in ruminant livestock species tends to lower CH_4 emissions per unit of product (Hayes et al., 2013)

Individual variation in daily CH_4 emission between ruminants is explained by their microbiota at the latter stage. All strategies aim to modify this microbiota directly or indirectly (Lassey et al., 1997; Martin et al., 2010). We need animals that produce important quantities of milk, but with a microbial population that ferment the feed with lower emissions of CH_4.

Genomic breeding values are necessary to assist on the progress of genetic selection in the direction of permanent and cumulative decline in enteric CH_4 emissions (Pickering et al., 2015b; Hayes et al., 2016).

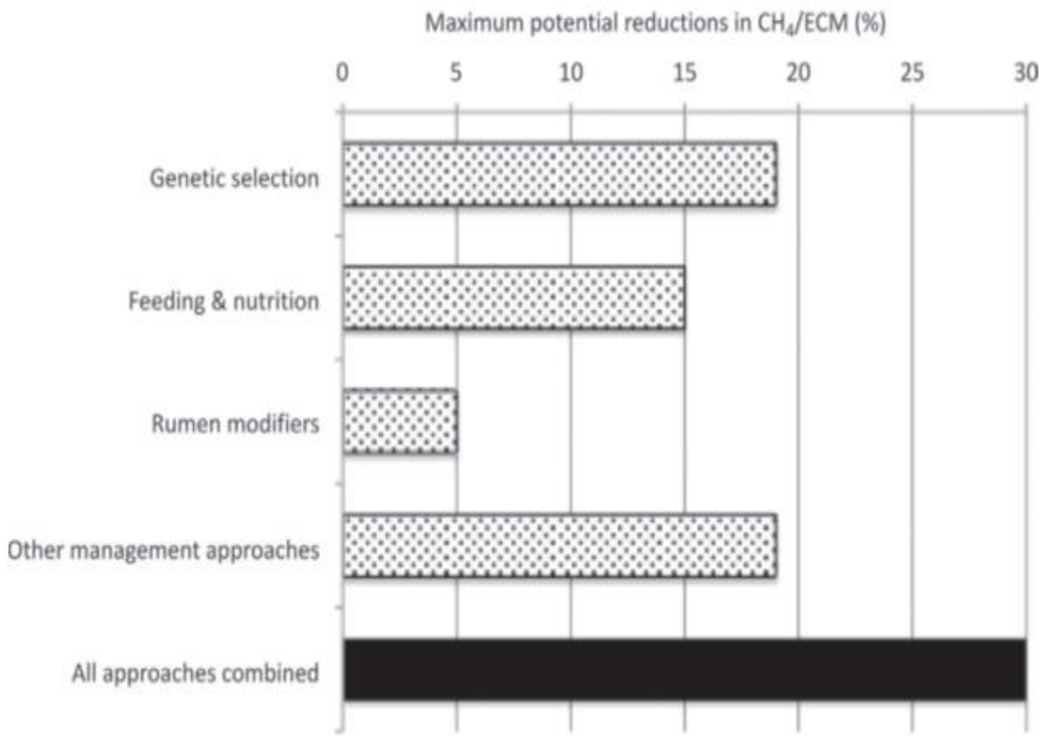


Figure 5. Estimated maximum impact of various approaches to mitigating CH_4 in intensive dairy production that have been demonstrated to be effective on an in vivo basis (Knapp et al., 2014)

It was predicted that classic genetic selection could reduce CH_4 emission (kg/lactation, grams/ FPCM) by 11 and 26% in 10 yrs. However, the greatest limitation for a breeding program is in measuring CH_4 or traits related to it (e.g., feed intake) on the progeny of sires. Genomic selection (GS) might provide an alternative to cope with this limitation (de Haas et al., 2011; Pickering et al., 2013a). GS consists in measuring a large reference population for MY or residual CH_4 production (RMP), genotyping this reference population for a large number of SNP markers, and then using the information to derive a genomic prediction equation, which can then be used to calculate genomic estimated breeding value (GEBV) for any selection candidate that is genotyped (Figure **6**) (Hayes et al., 2016). GS has the advantage over traditional selection (based on pedigree and phenotype alone) to select animals accurately early in life based on their genomic predictions, and also for traits that are difficult or expensive to quantify (e.g fertility, disease resistance, CH_4 emissions, and feed conversion) (Hayes et al., 2013). GS has been adopted for dairy industry worldwide, and is expected to double the genetic gains for milk production and other important traits (Hayes et al., 2013). It might, therefore, decrease CH_4 emissions given that some genetic variation between animals exist.

In conclusion, genetic and genomic selection for CH_4 emission reduction represents a potential option in dairy cattle. However attention needs to be directed to a number of issues related with brief and low cost measurements before it is to be implemented in practice (Pickering et al., 2013a).

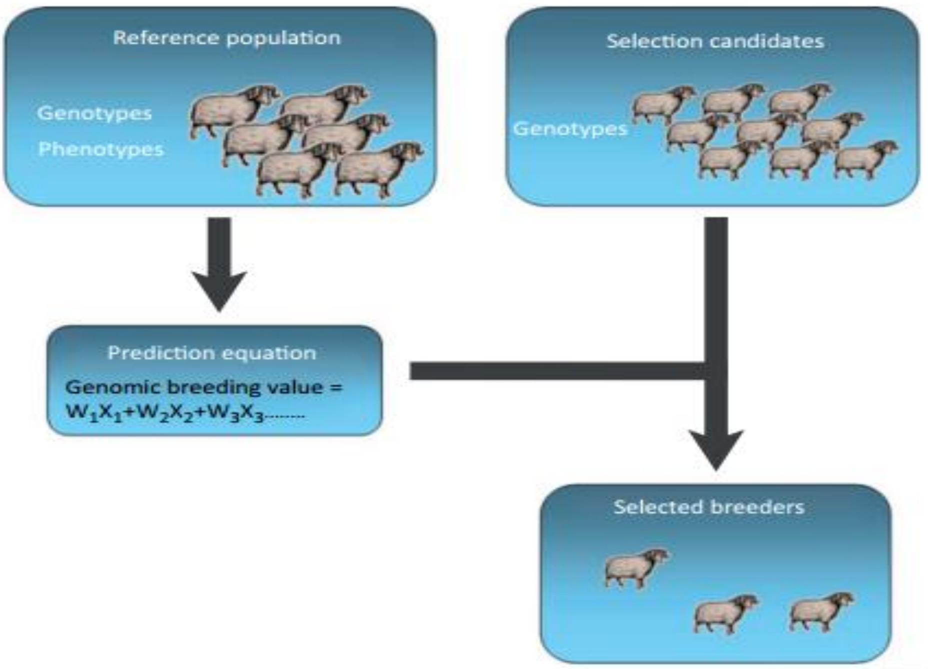


Figure 6. Genomic selection. A large number of individuals are measured for the trait and genotyped for the genome-wide markers (reference population) (Hayes et al., 2013).

4. Breeding objectives

The first step in the design of structured breeding programs should be the definition of breeding objectives (Kluyts et al., 2003). The maximization of profit can be the simplest (and most important) breeding goal. Several scientists have studied the concept of breeding objectives and adapted the principles to dairy cattle breeding (Groen, 1989; Kluyts et al., 2003; Wall et al., 2010; Laske et al., 2012; Hietala et al., 2014; Robinson and Oddy, 2016).

"The primary goal of most livestock producers is, very simply, to make money"

(Harris, 1970).

The development of a breeding goal can be discretized in the following phases (Hazel, 1943; Kluyts et al., 2003):

- Determination of the breeding, production and marketing system
- Specification of income and expense sources in commercial farms
- Specification of biological traits affecting income and expenses
- Derivation of economic weights
- Designation of selection criteria
- Estimation of genetic (heritability, genetic correlations) and phenotypic parameters (standard deviation, correlations)
- Estimating breeding values
- Dissemination of genes through selective mating

Developing breeding objectives that include environmental concerns is possible and suitable. New techniques for measuring direct and indirect emissions will improve the potential for reducing emissions by exploiting these measures in genetic selection (Wall et al., 2010). The importance of direct observation of methane emissions, as well as phenotypic and genetic correlation with its proxies was emphasized by Robinson and Oddy, (2016).

5. CH_4 measurements in dairy cattle

Precise and inexpensive direct methane measurements are needed for (co)variance estimation with other traits, and for genetic evaluation purposes (Storm et al., 2012, de Haas et al., 2017). Herd et al., (2013) suggested CH_4 production (L/d or g/d), CH_4 intensity (L/kg of milk or live weight), and CH_4 yield (L/kg DMI) as a possible direct phenotypes for CH_4 production. Residual CH_4 production (Observed minus predicted CH_4 production) can also be used (Manzanilla-Pech et al., 2016).

The main strengths and weaknesses of each phenotype are summarized in Table **1** (de Haas et al., 2017).

Table 1. Several methane phenotypes with their definitions, strengths, and weaknesses (de Haas et al., 2017; Methagene working group, 2017)

Trait	Definition	Strength	Weakness
Methane production	Methane production per day (L/d or g/d)	- The ultimate trait we want to improve - Easy to understand - Heritable trait	- Highly correlated with feed intake and production level
Methane intensity	Methane production related to output (e.g., per kg of milk, live weight, meat)	The phenotype of interest for the user	- Non standardized units (litters, energy, energy corrected,..) - Ratio trait, so selection can be difficult to incorporate properly - Needed to be clearly discussed by policy makers as a phenotype to select for.
Methane yield	Methane production related to input (e.g., kg of DMI)	The phenotype of interest for the user	- Depends on diet composition - Difficult to discriminate animals with different - Not really used by farmers or industry - Ratio trait, so selection can be difficult to incorporate properly
Residual methane Residual methane	Observed methane production minus predicted methane production	- Good statistical properties - corrected for traits that influence methane	- Correlation between observed and expected CH_4 production can be low, as proxies and predictors are not always accurate - Can be difficult to explain to users

5.1. Measurements techniques of CH_4 in vivo

Several techniques are available for measuring methane directly in farms. The convenience of each technique depends on many factors, like the cost, level of accuracy suited, and the scale and design of the experiments to be undertaken (Bhatta et al., 2007). Each one of the techniques has its advantages and disadvantages. A brief description of the main available techniques follows.

5.1.1. Respiration chambers (yield)

The chambers estimate CH_4 measurements by collecting exhaled breath from the animal and measuring gas concentration as the animal stays in the chamber usually for several days (e.g., the CH_4 concentration) (Storm et al., 2012; Pickering et al., 2015b). There are two types of chambers, the closed circuit and the open circuit. In the open-circuit, a known flow of air is drawn past the animal and its change in composition is measured; while in the closed-circuit the CO_2 is absorbed and weighed and oxygen is metered into the system. The open-circuit chamber has an exact counterpart in a mask method and the closed-circuit chamber an approximate counterpart in a mask-spirometer method (Turner and Thornton, 1966).The closed circuit system is hardly used and open circuit chambers are preferred (Broucek, 2014a). In Figure **7** an outline of an open-circuit system is represented. According to Hammond et al., (2016), two critical origin of variation for measurement of CH_4 emission through respiration chambers are airflow rate across the chamber and the dynamics of air mixing in the chambers, which defines response time.

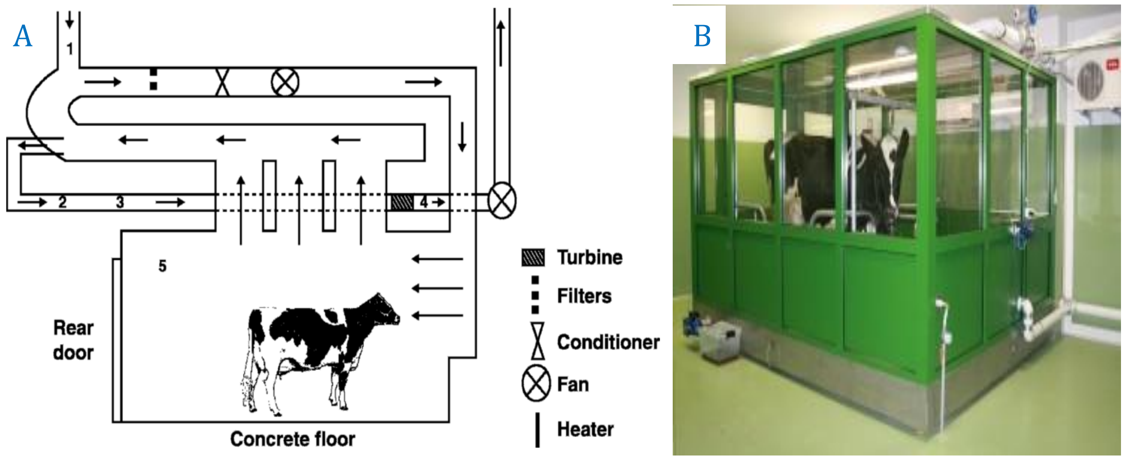


Figure 7. Respiration chamber;
A) Schematic of the open-circuit respiration chambers (Grainger et al., 2007)
B) Use of respiration chambers to measure methane emission, Poland (Source: http://globalresearchalliance.org/country/poland/; Accessed: January 11, 2018)

5.1.2. The SF6 technique (concentration)

SF6 tracer gas is delivered through a permeation tube (bolus), which is placed in the rumen. The CH_4 to SF6 ratio breath is measured over 24 hours (ie, a complete feeding cycle), and repeated over a period of five to eight days by exchanging the canister. The sample is corrected with respect to the background concentration. As such, if the tracer concentration is known, the rate of CH_4 production can be calculated (Figure **8**) (Berndt et al., 2014).

SF6 is a gas that is easily measurable and traceable at low concentrations. It is of synthetic origin (Hill et al., 2016). This technique is suitable for free ranging, penned and grazing animals (Hammond et al., 2016). However, SF6 technique is expensive and has not been widely adopted (Hill et al., 2016). Garnsworthy et al., (2012a) argues that, because of repetitive handling on animals, gas collection machines attached cows, and the insertion of the bolus in the rumen to release SF6, this technique may interfere with cow behavior. Further, the lifespan of the bolus is unknown and it may release SF6 for an unknown period, precluding repeated measurements for the same cows.

B

$$Methane\,(g/d) = SF_6\ release\ rate \times \frac{Animal\ sample\,[CH_4] - Background\ air\,[CH_4]}{Animal\ sample\,[SF_6] - Background\ air\,[SF_6]} \times \frac{Molecular\ weight\ CH_4}{Molecular\ weight\ SF_6} \times 1000$$

Figure 8. The Sulphur hexafluoride (SF6) technique.
A) Examples of sampling points for the collection of air samples(Berndt et al., 2014).
B) Calculation of daily CH_4 emission formula (Hammond et al., 2016)

5.1.3. GreenFeed system (automated head chambers)

The GreenFeed system consists on an extractor fan that draws air over the animal's head, to make it pass through the nose and mouth and then collect the air through an exhaust pipe (Figure 9). The collected air is mixed, filtered and the airflow rate is measured using a hot-film anemometer. The concentration of CH_4 (and CO_2 and O_2) in the sample is defined using non-dispersive infrared analysis (Hammond et al., 2016).

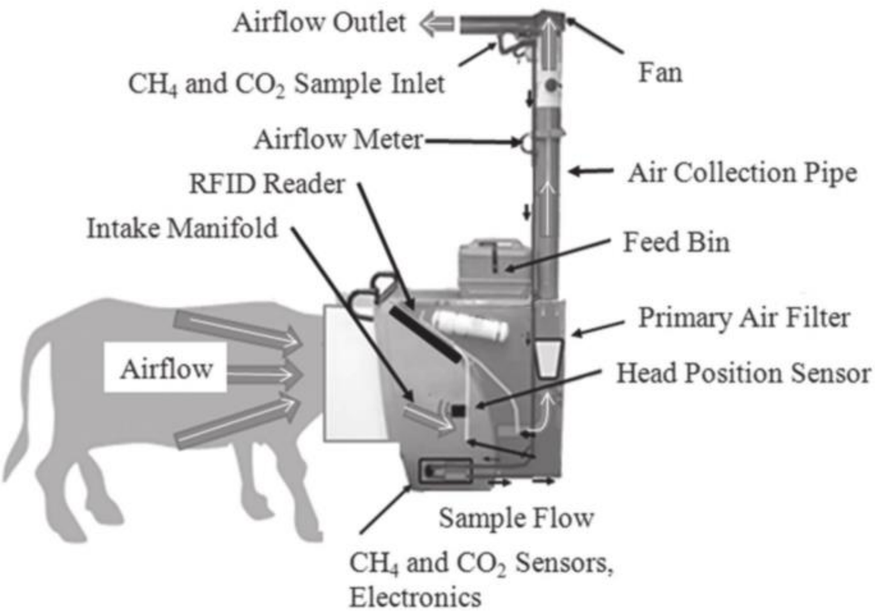


Figure 9. Layout of the GreenFeed (C-Lock Inc., Rapid City, SD) system (the flux method). RFID = radiofrequency identification (Huhtanen et al., 2015).

5.1.4. Sniffers

The sniffer type method (Figure **10**) of breath analysis during milking is a "brief measurement" (Pickering et al., 2013b) and a low cost method technique (Garnsworthy et al., 2012b) for estimating CH_4 production for a large number of animals, which allows estimating variation in daily emissions on farm (Hammond et al., 2016). In this method, a sampling inlet is placed in the feed manager trough of an automatic milking system to collect air emitted by animals during milking, and gas concentrations in exhaled air are continuously sampled, analyzed, and logged at 1-s intervals using data loggers to measure CH_4 and CO_2 concentrations in close proximity to the muzzle of the animal. Information on eructation frequency and CH_4 released per eructation are used to estimate CH_4 emission rate by individual animal during milking (Garnsworthy et al., 2012b).

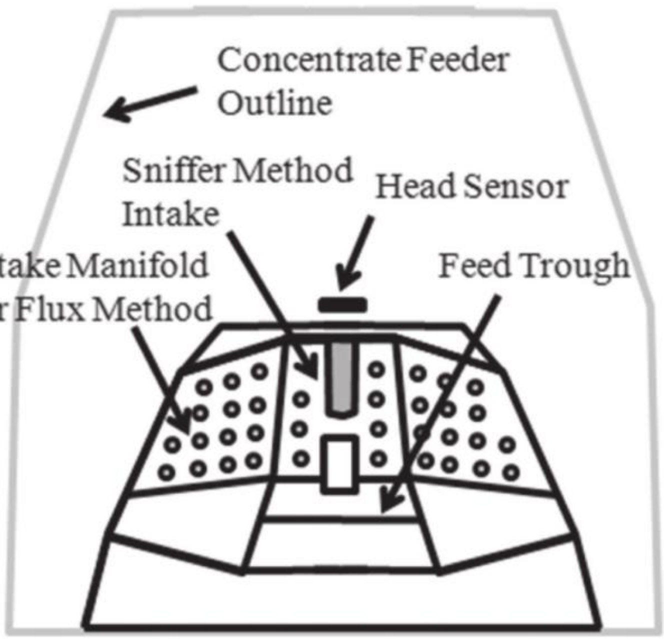


Figure 10. The sniffer-method configuration used in laboratory and farm studies (Huhtanen et al., 2015)

5.1.5. Laser devices (concentration)

The laser CH_4 detector has been suggested as a method to characterize enteric CH_4 emissions from animals in a natural environment (Ricci et al., 2014).Measurement of CH_4 by Laser CH_4 devices are taken manually by a potable apparatus from 1-3 m of distance from the cow. It is based on infrared absorption spectroscopy using a semiconductor laser as a collimated excitation source and employing the second harmonic detection of wavelength modulation spectroscopy to establish a CH_4 concentration measurement. The integrated concentration of CH_4 between the apparatus and the target point is then displayed. The measured value is expressed as CH_4 concentration while accounting for the thickness of any CH_4 plume. Hence, the measurements are expressed in parts per million-meter (ppm-m) (Chagunda et al., 2009).

A series of peaks, which represent the respiratory cycle of the animal are recorded. Often, only the peaks that reflects an increase in CH_4 concentration due to eructation are considered (Ricci et al., 2014) (Figure **11**).

Figure 11. Methane column density profiles from laser methane detector of three selected cows (Metalgen, 2017)

5.2. Proxies

Although several methods for measuring CH_4 in vivo have been suggested, most of those techniques do not fit to large-scale measurements at the farm level and are mostly expensive. Possible proxies including indicators or indirect traits for

CH_4 had been reviewed and described by Negussie et al., (2017), as a potent mitigation strategy. Those traits are mostly based on feed intake, rumen function and microbial communities, milk composition and modelling by combining proxies in prediction equations for CH_4.

Furthermore, Prediction equation of CH_4 production in dairy cows , from proxies had been developed by various authors (Mohammed et al., 2011; Ramin and Huhtanen, 2013; Charmley et al., 2015).

Table 2 presents a comparison between the main proxies used to estimate CH_4 emissions.

Table 2. Comparison of different proxies used in estimation of CH_4 emissions (Methagene working group, 2017)

Method	Easy	Accuracy	Cost	Invasive	Throughput
Milk MIR	easy	low	moderate	No	High
Rumen Fatty acids	difficult	low	Low-moderate	Yes	Low
Feed intake/ efficiency	moderate	medium	High	No	Low
Body weight	easy	medium	low	No	High
Milk yield	easy	Low	low	No	High
Rumination activity (sensors)	moderate	-	High	No	Low
Rumen microbiota	difficult	medium	High	Yes	Low
Faecal ether lipids	Difficult	Low	High	No	Low
Milk fatty acids	moderate	low	High	No	High

5.3. Conclusion

Hill et al., (2016) presented and compared different methods of CH_4 measurement. Table 4 highlights the main advantages, disadvantages, the tendency of the cost and the main characteristics of each method of measurement of CH_4. A poor correlation between Sniffer method and CH_4 flux measured by the GreenFeed system was reported by K.J. Hammond et al., (2015). A better correlation between sniffer method based on CH_4/CO_2 ratio and CH_4 flux was reported by Cabezas-Garcia, (2017). The study of Chagunda and Yan, 2011, showed a high positive correlation coefficient of the laser methane detector and the calorimeter chamber. Garnsworthy et al., 2012b reported a good relationship between the measurements of CH_4 emission using the RC technique and CH_4 emission rate using sniffer method. A high and positive correlation between SF6 technique and RC was described by Deighton et al., 2013 (Table 3).

Table 3. Correlation coefficient between CH_4 production, determined using SF6 technique, GreenFeed system, Sniffers, Laser devices; and respiration chamber techniques.

Method	Correlation with Respiration Chambers	Reference
SF6	0.84	(Deighton et al., 2013)
GreenFeed system	0.1043	(K.J. Hammond et al., 2015)
Sniffers	0.79	(Garnsworthy et al., 2012b)
Laser devices	0.80	(Chagunda and Yan, 2011)

Table 4. Comparison of Different Techniques for Measuring Enteric CH_4 Methods (Ricci et al., 2014; Hill et al., 2016; Cabezas-Garcia, 2017; Methagene working group, 2017)

Method	Respiration and accumulation chambers	Tracers/SF6	Field measurements /GreenFeed	Sniffers	Laser devices
Advantages	Highly accurate, controlled environment; information about individual animals	Accurate; few interferences by other gases; the animal can free range	Information about many animals; data produced in natural grazing environment	Measures CH_4 emissions on very large numbers of animals in farm conditions	LMD variables could be used to rank animals or investigate differences between diets
Disadvantages	Results different from free-range animals; configurations still vary from one research group to another; an animal adaptation period is required; every 2–3h accumulation chambers must release CO_2 that builds up	Rely on SF_6, which is a greenhouse gas itself; does not completely capture all tracer and, therefore, relies on spot concentration measurements; high contact with animal, which can disrupt normal behavior	Require expensive and accurate measurement approaches: data processing heavily influenced by microclimatic conditions; loss of data can be high	Does not measure CH4 emissions directly Highly dependent on the muzzle position of the animal	it measures only concentrations, and without a means of calibrating data out- put against daily CH_4 production, the main value of LMD data would be comparative between diets, animals, and systems in terms of ranking
Cost	High	Moderate	Moderate	Low	Low-moderate
Robustness	Moderate-high	Moderate	Moderate	Low- moderate	Moderate
Intrusiveness	moderate -high	Moderate	Low-moderate	Low-moderate	Low
Throughput	Low	Moderate	Moderate	Moderate-high	Moderate-high
Labour intensity	Moderate-high	High	Low-moderate	low	Moderate-high
Automated matching (animal Id mistake	No	No	Yes	Yes	No
Total time in life an animal can be recorded	Entire life	One time	Lactation period	Lactation period	Entire life
Flux/ concentration	Flux	Flux	Flux	Concentration	Concentration

6. Estimation of economic weights and profit function

Finding the relative economic value of each trait is the first step in defining the ideal towards to which the breeder must strive (Hazel, 1943). Correct economic values are required to achieve a genetically and socio-economically balanced selection in cattle for both production and functional traits (Groen et al., 1997). We hereby, present the key concepts for deriving economic values from a bio economic model (profit function) next.

6.1. Aggregate genotype and selection index

Hazel and Lush, (1942), reported three methods of selection according to its efficiency, and they demonstrated that the "total score" is the most efficient one. This method consists of weighted selection for all target traits simultaneously, using some net merit index. This index is constructed by adding the credits and penalties for each animal into one single figure, which accounts for the animal superiority or inferiority for each trait in comparison to the population average. The drawback of this approach is that the genetic progress expected for a particular trait when using an index of *n* traits is only a fraction of the progress that is obtained when only this trait is selected, this fraction is equal to $I/\sqrt{n}$ if the traits are independent and the product of their economic weight, their heritability and their standard deviation is the same. When setting up breeding programs, the choice of an aggregate genotype should be the starting point (Groen et al., 1997). The aggregate value of an animal is the sum of its genotypic value for each trait, weighted on the relative economic value. An animal's aggregate genotype is defined as the sum of the average (strictly additive) effects of the genotypic values of the traits that composed the breeding goal:

$$H = a_1 g_1 + a_2 g_2 + \cdots + a_n g_n \qquad \text{Eq. 1}$$

Where:

a_i: The economic value of the trait *i*.

g_i: The true additive genetic value for the trait *i*.

The selection index (I) aims to predict (H) from *m* measurable traits and is written as:

$$I = b_1x_1 + b_2x_2 + \ldots + b_mx_m \qquad \text{Eq. 2}$$

where x_j represent the phenotypic performance for the observed traits and b_j are the regression coefficients, calculated in order to maximize the correlation between the aggregate genotype and the selection index (r_{IH}) (Hazel, 1943), which is the same as maximizing the logarithm of r_{IH} or minimizing the variance of the prediction error (statistically equivalent to minimizing the sum of the squares of the deviations between H and I) (Weller, 1994). The variance of prediction error is as follow:

$$Var\,(I - H) = var\,(I) + var\,(H) - 2cov(I,H) \qquad \text{Eq. 3}$$

The correlation between H and I is written as:

$$r_{IH} = \frac{\text{cov (I,H)}}{\sqrt{var(I)var(H)}}$$

$$\log(r_{IH}) = \log cov(I,H) - \frac{1}{2}\log var(I) - \frac{1}{2}\log var(H) \qquad \text{Eq. 4}$$

b_i coefficients are estimated by minimizing Eq. *3* or maximizing Eq. *4* equating to zero the derivative of each of them with respect to b_i.

After a mathematical development we have the following equation (Groen et al., 1997):

$$\mathbf{P.b} = \mathbf{C.a} \qquad \text{Eq. 5}$$

Where:

b : an *m* x 1 vector with the regression coefficients of the index traits;
P : an *m* x *m* matrix with the covariances between *m* index traits;
C : an *m* x *n* matrix with the covariances between *n* genotype traits and *m* index traits.

a : Vector of economic weights of the *n* traits in the aggregate genotype
The variance of the selection index is :

$$\mathrm{var(I)} = var\left(b' X\right) = \ b' var(x) b = b' Pb \qquad \textit{Eq. 6}$$

The variance of the aggregate genotype:

$$\mathrm{var(H)} = var\left(a' g\right) = \ b' var(g) b = a' Ga$$

Where:

G: Matrix of *n* rows and *n* columns formed by the variances and covariances between the *n* traits that involve the aggregate genotype.

The covariance between the selection index and the aggregate genotype is:

$$\mathrm{cov\ (I, H)} = cov\left(b' X, H\right) = b' cov(X, g) a = \ b' Ca = b' Pb \qquad \textit{Eq. 7}$$

and, the correlation between H and I (r_{IH}), which is the accuracy of the selection index, is:

$$r_{IH} = \frac{\mathrm{cov\ (I,H)}}{\sigma_I \, \sigma_H} = \frac{\sigma_I}{\sigma_H} = \frac{\sqrt{b' PB}}{\sqrt{a' Ga}} \qquad \textit{Eq. 8}$$

The genetic response of the index I is as follows:

$$R_I \ = i\sigma_I = i \, r_{IH} \, \sigma_H \qquad \textit{Eq. 9}$$

6.2. Profit function

The modelling methods to derive economic weights can be divided into simulation, dynamic programming and profit functions (Kluyts et al., 2003). Profit function or "Efficiency of production", is a function of inputs and outputs of the production system. Inputs can be defined as the total of production-factors required for production within the system; outputs as the total of products resulting from production within the system (Groen, 1989). Harris, (1970) suggested three possibilities that can define the profit function:

- maximize benefits (= outputs - inputs);

- minimize costs per unit of product;
- maximize revenues/costs

In animal breeding, the first or second points have been mainly considered, although the most vital has been the former one (Smith et al., 1986; Gibson, 1989):

$$B = R - C$$

Where B is benefits, R is returns, and C costs. R and C are functions of any number (n) of traits.

6.3. Economic value estimation

Traditionally, traits in the breeding objective need to have an economic value in order to place genetic pressure on it (de Haas et al., 2017). This economic value expresses to which extent the economic efficiency of production is improved at increasing one unit of genetic superiority for such a trait (Groen, 1989). Brascamp et al., (1985) showed that the economic values derived from profit equations depend on the base used for the evaluation (e.g. per unit of investment, per breeding female, per individual or per unit of product). Thus, in order to remove uncertainty about the appropriate economic values in livestock, the profit equation has to be transformed by setting its outcomes to zero (profit as a cost of production), this is named "normal profit" in economics. Then the relative economic values are likewise for all bases of evaluation, which make it the appropriate basis for determining economic weights.

Economic weights are derived as partial derivatives of profit function with respect to the trait considered in order to define a linear aggregate breeding objective (Moav, 1973; Brascamp et al., 1985) . In other words, the economic weight of a given trait is the change in the benefit due to the increase of a unit in the genetic merit of such trait. This is usually calculated by comparing the difference in benefits between the current situation and the situation in which a trait increases one unit, keeping the all other traits constant.

Several studies in the literature have estimated economic values of production and functional traits over time and across countries in dairy cattle. In Dutch dairy cattle, economic values for veal, beef and milk production traits using profit equations, were derived by Bekman and van Arendonk, (1993). Charfeddine, (1997) developed a bio economic model of dairy cattle in Spain, and estimated economic values in a situation of free market and quota, for both productive and functional traits. Another study carried out by González-Recio et al., (2004) in Spanish Holstein cows, included fertility cost in a bio economic model, and estimated economic values for calving interval and number of inseminations per service period. Vargas et al., (2002), estimated economic values for both production and functional traits in Holstein cattle of Costa Rica, the results showed that improving genetically fertility, health and cow efficiency traits impact positively the profitability of Holstein cows. Wall et al., (2008), developed a framework to derive economic values for body tissue mobilization, to be added to a broader economic index in dairy cattle in United Kingdom. Economic values in three breeding perspectives for longevity and milk production traits were estimated in Holstein dairy cattle in Iran (Sadeghi-Sefidmazgi et al., 2009). Ghiasi et al., (2016) estimated economic values for fertility, stillbirth and milk production traits in Iranian Holstein dairy cows. Hietala et al., (2014) estimated economic values of production and functional traits, including residual feed intake, in Finnish milk production. Veerkamp et al., (2002) estimated economic values for milk, fat and protein yields, survival and calving interval for pasture-based systems in Ireland under different milk quota scenarios, and included it in the economic breeding index. Wolfová et al., (2007), calculated economic weights for dairy and beef sires in crossbreeding systems, and reported differences in relative economic weights between the purebred and crossbred dairy systems for some traits of the dairy sires.

The first study to derive the economic value of enteric CH_4 produced by ruminants was recently undertaken by Bell et al., (2016). This study estimated an economic value of -£1.68 per kg increase in CH_4 per lactation of enteric CH_4 (kilograms/lactation). Another study carried out by Bell et al., (2015) showed that increasing production efficiencies would increase profit and decrease emissions per cow and per kg MS of dairy systems. Recently, Amer et al., (2017), developed a methodological framework for deriving weightings to be incorporated in selection indices from genetic traits that impact GHG emissions intensities in Irish dairy cattle.

OBJECTIVES

1. Main objective

The main objective of this thesis is to include CH_4 emissions into the breeding goal of dairy cattle, under foreseen scenarios that aim to reduce the carbon footprint of dairy cattle in terms of lower methane emissions.

2. Specific objectives

- To determine the economic value of CH_4 under four different scenarios in the Spanish dairy cattle:

    i. Scenario 1: current situation as benchmark (without putting an economic value on CH_4 emissions)
    ii. Scenario 2: penalization of CH_4 emissions through a carbon tax.
    iii. Scenario 3: CH_4 emissions in a carbon quota.
    iv. Scenario 4: including CH_4 as a net energy loss cost.

- To estimate the variance components for CH_4

- To include CH_4 emissions in the selection index in each of the foreseen scenarios.

- To evaluate the expected response to selection under the scenarios tested.

MATERIAL & METHODS

1. Breeding goal

We assumed the current breeding goal that is applied in the Spanish Holstein population by CONAFE. It aims for larger benefits from milk yield, longer productive life, and reduced mastitis incidence and days open.

Here, the reduction in CH_4 production was included into the breeding objective as mentioned in the objective of this thesis

The current ICO index includes the following traits (http://www.conafe.com/VisorDocs.aspx?pdf=evaluaciones_Metodologia_y_requisitos.pdf) presented in Table 5 .

Table 5. Traits included in the current ICO

Traits		Units
Milk yield	(Milk)	kg
Milk fat yield	(Fat)	kg
Milk protein yield	(Prot)	kg
Feel and Leg Index[1]	(FLI)	-
Udder Composite Index	(UCI)	-
Longevity	(Long)	days
Somatic Cell Count	(SCC)	log(SCC)
Days Open	(DO)	days
Enteric Methane	(CH_4)	kg

2. Genetic parameters

The (co)variance matrix between the traits in the index were provided by CONAFE (Table 6).

The number of records observed per each trait for the progeny are based on CONAFE data. The number of observations was set to 50 for CH_4.

[1] From genetic evaluations of linear traits (conformation traits) recommended by the World Federation of Holstein-Frisian, and evaluated by INTERBUL, synthetic indices are calculated, such as the Feet and Leg Index (FLI) and the Udder Composite Index (UCI)

Table 6. Phenotypic (above diagonal), and genetic correlations (below diagonal), between traits, number of progeny observation (Num dau), heritabilities (h2), repeatabilities (r) and genetic standard deviation for the traits in the ICO

Trait	Milk	Fat	Prot	FLI	UCI	Long	SCC	DO	Num dau	h2	r	Genetic SD
Milk	1	0.59	0.92	0.20	0.20	0.20	-0.04	0.11	150	0.28	0.50	800.00
Fat	0.75	1.00	0.61	0.20	0.20	0.21	-0.01	0.02	150	0.28	0.50	24.00
Prot	0.93	0.83	1.00	0.20	0.20	0.22	0.04	0.08	150	0.28	0.50	22.00
FLI	0.47	0.45	0.50	1.00	0.10	0.40	0.00	0.00	150	0.16	0.16	1.00
UCI	0.53	0.49	0.56	0.58	1.00	0.40	0.00	0.00	150	0.39	0.39	1.00
Long	0.12	0.21	0.13	0.33	0.49	1.00	0.00	0.00	100	0.09	0.12	10.00
SCC	-0.01	-0.10	-0.01	-0.09	-0.24	-0.51	1.00	0.00	150	0.18	0.37	1.00
DO	0.51	0.41	0.46	0.20	0.18	-0.32	-0.26	1.00	150	0.04	0.07	10.00

FLI: Feet and Legs Index; UCI: Udder Composite Index; long: longevity, SCC: Somatic Cell Count; DO: Days Open; CH_4: Methane

CH_4 heritability and correlation (genetic and phenotypic) estimates with other traits were calculated, but in order to evaluate the response to selection, heritability of CH_4 was set to 0.25 (Kandel et al., 2017b), and repeatability to 0.41 (Haque et al., 2015), mainly because of our small data set .

Actually, Several recent studies attempted to measure individual CH_4 emissions in dairy cattle, and estimate genetic and phenotypic (co)variance components in their real environment (Haque et al., 2015; Pickering et al., 2015a; Breider et al., 2018; Zetouni et al., 2018). Other authors estimated those components from proxies, such as mid infra-red spectra and milk composition (de Haas et al., 2011; van Engelen et al., 2015; Lassen and Løvendahl, 2016; Kandel et al., 2017b; a).

In this study, variance components of CH_4 are estimated with a one trait model, using Bayesian regression. The genetic analysis is carried out using the TM software (Legarra et al., 2008), and were saved 270000 samples of the marginal distribution discarding the first 30000 as "burn-in". The model includes environmental effects of parity (Par), days in milk (DIM) , the interaction between herd (Hd), robot (Rt) and week of lactation (Wk), a permanent effect (Pe) and the additive effect (a). A total of 15379 animals in pedigree were included. The model could be described as follows:

$$CH_4 = \mu + Par + DIM + Hd * Wk * Rt + Pe + a + e$$

Where:

μ Mean of population trait

e Error

The data includes week mean of CH_4 concentration (ppm) measured by sniffers method on 439 Holstein cows. Phenotypic correlations between CH_4 and production traits were calculated based on this data set (Table 8). The same model was used including CH_4 as a trait expressed in (g/d).

Table 7 presents the mean, and number of observations and cows of phenotypic data. The average of CH_4 emissions in these data was 224 g / day, with a phenotypic standard deviation of 126.4.

Table 7. Summary statistics of phenotypic data

Herd	Mean(g/d)	Mean (ppm)	Number of records	Cows
1	507.4	3494.90	116	58
2	126	911.42	401	82
3	197.2	1366.72	190	65
4	307.8	2155.67	330	113
5	212.2	1482.43	349	121
Total	224.0	1630.09	1386	439

Table 8. Phenotypic correlations between CH_4 and production traits

Trait	Milk	Fat	Protein	CH_4
Milk yield	1	0.59	0.92	-0.07
Milk fat yield		1	0.61	-0.18
Milk protein			1	-0.13
CH_4				1

CH_4: **Methane**

Genetic correlations between CH_4 and all other traits in the ICO are calculated using official genetic evaluations from CONAFE (june 2018), and included 480 sires evaluated for all the traits in the ICO. Table 9 presents the number of sires used for this calculation per trait.

Table 9: number of sires used per trait to calculate genetic correlations between CH_4 and other traits in the ICO

Trait	Number of sires
Milk	480
Fat	480
Prot	480
FLI	466
UCI	466
Long	478
SCC	478
DO	478
CH_4	480

FLI: Feet and Legs Index; UCI: Udder Composite Index; long: longevity, SCC: Somatic Cell Count; DO: Days Open; CH_4: Methane

Genetic correlations are estimated according to the method of Blanchard et al., (1983) as follows:

$$\hat{r}_g = \frac{\sqrt{(\sum b_i)(\sum b_i')}}{\sum b_i b_i'} r_{(EBV,EBV')}$$

where $\hat{r}_g$ is an estimator of the genetic correlation, EBV is breeding value of CH_4, EBV' is the breeding value of a second trait obtained from CONAFE evaluation, $r_{(EBV,EBV')}$ is Pearson's correlation between sets of breeding values of the two analyzed traits, $\mathrm{b_i}$ is EBV reliability for CH_4 and $\mathrm{b_i'}$ is EBV reliability of a second trait obtained from CONAFE evaluation. Reliability of the estimated breeding values was calculated based on the predicted error variance provided by TM software.

3. The bio economic model

3.1. Input Parameters

Productive and economic parameters for the dairy system including the incomes and costs of alimentation, health, reproduction and workforce were obtained from Spanish Holstein Association (CONAFE). The average data were based on completed lactations in 2013, representing all regions of Spain (Table **10**, and Table *11*).

Table 10. Production traits included in the model for an average lactating cow

Parameter	Units	Average
Lactation Length	days	305
Milk yield	kg	9542
Milk fat yield	kg	348
Milk protein yield	kg	305
Fat%	%	3.65
Crude Protein%	%	3.20
Lactations	lactation	2.91
Coital Index	insemination	2.69
Liveweight	kg	600
Gestation lenght	days	283
Calf birth weight (kg)	kg	45
Replacement rate	%	34.4%
Mortality rate cows	%	6%
Culled cows	%	28.6%
Lifetime	year	6

The data includes milk production and composition for an average standardized lactation, number of lactations during the lifetime, replacement and mortality rate for an average heard, and the component parameters for revenues and costs included in the bio economic model.

Table 11. Economic parameters (in €/unit) for commercial dairy production applied in the bio-economic model

Parameter	Unit	€/unit (Average)
Revenues		
Milk volume	kg	0.034
Milk Fat	kg	3
Milk Protein	kg	5
Male calf	head	69
Female calf	head	400
Culled cow	kg Live weight	1.17
Costs		
UFL	UFL	0.18
Mcal	Mcal	0.11
Medicines	head/lactation	115
Veterinary	head/lactation	50
Mastitis	Case/lactation	35
Milking	head/lactation	103
Workforce	head/lactation	660
Artificial Insemination	dose/cow	22.0
Heifer replacement	head	1684

3.2. Description of the bio-economic model: the profit function

A detailed bio-economic model was computed in R for Statistical Computing to calculate the changes in benefits per cow per lactation annum, in response to changes in the biological traits of interest. The profit function was described as follows:

$$\mathrm{B}\left(\frac{\frac{€}{cow}}{yr}\right) = R - C - C_{CH_4} \qquad \text{Eq. 10}$$

B is the benefits expressed in euro per cow per year, R and C are revenues and costs for each trait, and C_{CH_4} is CH_4 emission cost. The revenues include incomes from milk volume, milk fat and milk protein, as well as from calves and culled cows sale (Figure 12).

The profit function can then be expressed as:

$$\mathrm{B}\left(\frac{\frac{€}{\mathrm{cow}}}{\mathrm{yr}}\right) = (\mathrm{R}-\mathrm{C})\mathrm{milk} + (\mathrm{R}-\mathrm{C})\mathrm{fat} + (\mathrm{R}-\mathrm{C})\mathrm{protein} + (\mathrm{R}-\mathrm{C})\mathrm{CH_4} + \text{meat revenue} \qquad \text{Eq. 11}$$

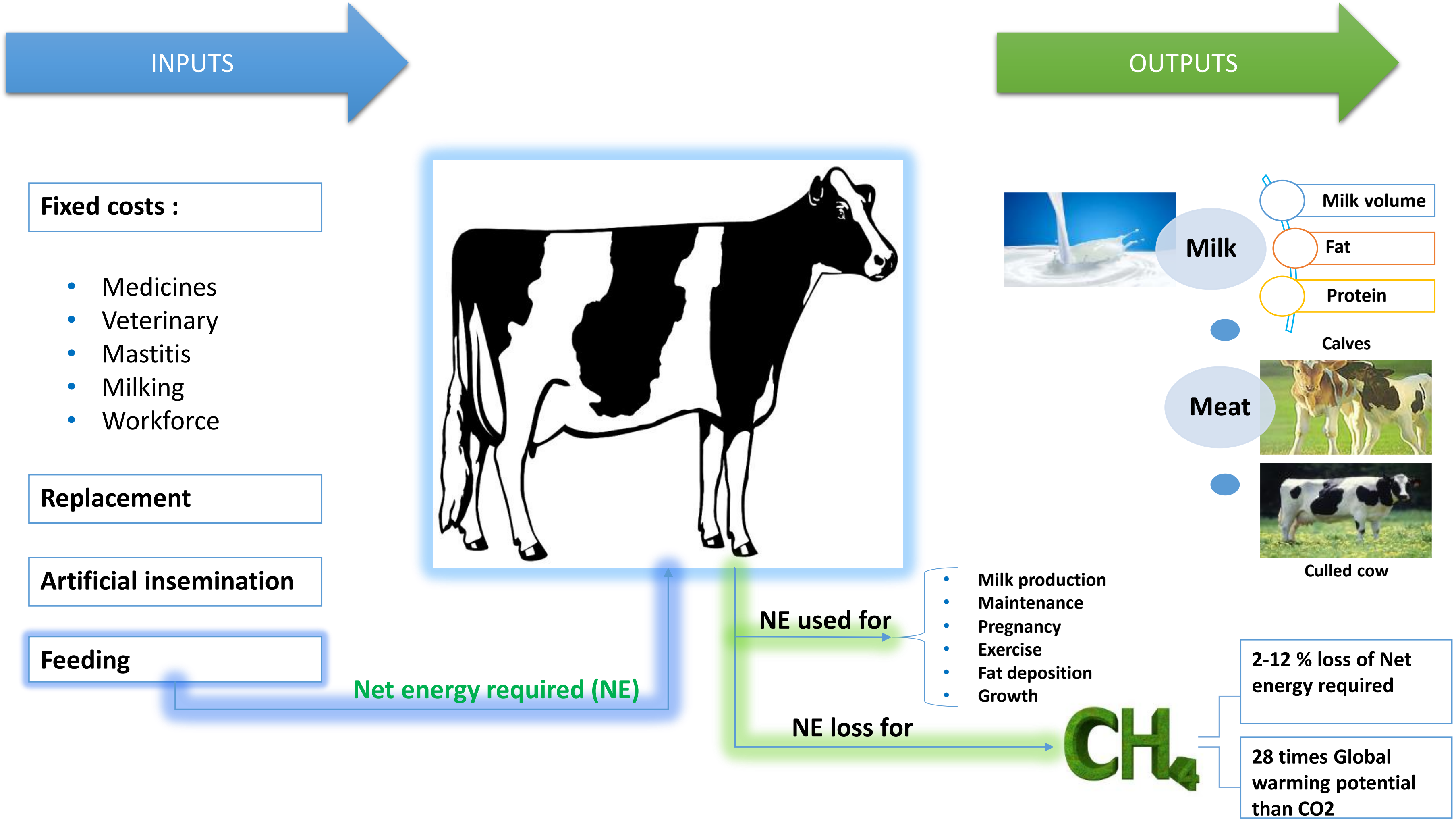


Figure 12. Graphic description of the bio economic model components

We assumed that the 50% of births are females, and 50% are males. The incomes from calf's sale were estimated as a function of replacement rate, male and female value (€/head), and mortality rate:

- Male (♂) calf meat income:

$$♂ Meat\ income \left(\frac{\frac{€}{calf}}{yr}\right) = (50\% + \%\,replacement) * (1 - \%mortality) * male\ \ value\ (€)$$

- Female (♀) calf meat income:

$$♀ Meat\ income \left(\frac{\frac{€}{calf}}{yr}\right) = (50\% - \%\,replacement) * (1 - \%mortality) * \ female\ value\ (€)$$

For culled cows, the meat income was estimated based on the following equation:

$$Culled\ cow\ meat\ income \left(\frac{\frac{€}{cow}}{yr}\right) = \frac{Culled\ cow\ value\ (€)}{average\ heard\ lifetime\ (yr)}$$

The costs in the bio economic model were estimated considering costs of:

- Feed for milk production
- Heifer rearing
- Artificial insemination (AI)
- Fixed costs: Medicines, veterinary, mastitis, milking and workforce.

For the AI cost, we assumed that all cows were artificially inseminated. We also assumed that 50% of AI are performed with genomic bulls, and 50% with proven bulls.

Based on the phenotype data , we considered an average emissions of 278.0 g·d−1 during lactating period ($Lact_{CH_4}$) and 250.0 g·d−1 on the dry period (Dry_{CH_4}). Hence, the estimation of total CH_4 emission per cow per year is as follow:

$$CH_4\, production\left(\frac{\frac{kg}{cow}}{yr}\right) = \left(Lact_{CH_4} * 305\right) + \left(Dry_{CH_4} * 60\right)$$

Where $Lact_{CH_4}$ is the average CH_4emissions during lactating period, and Dry_{CH_4} is the average CH_4 emissions during dry period. This results on an average emissions per cow per year of (278.0*305+ 250.0*60)/1000 = 99.8 kg/yr of CH_4.

The cost of CH_4 emission was estimated under the different scenarios as follow:

- Scenario 1:

No cost was applied to CH_4, as benchmark. However, CH_4 was incorporated in the selection index to test the response.

- Scenario 2:

In terms of environmental taxation, Spain has no explicit carbon tax (Donat et al., 2013). Hence, the estimation of the cost of CH_4, was based on the updated short-term carbon values reported by the Department for Business Energy & Industrial Strategy, (2017). We have estimated the cost of CH_4 based on the shadow price of CO_2 under three scenarios: low, moderate and high scenario. We have used the moderate scenario for 2028 (in 10 years). Assuming a global warming potential for CH_4, of 28 times that of CO_2 (Myhre et al., 2013), the cost of CH_4 emitted per cow per year was:

$$CH_4\, cost\left(\frac{\frac{€}{cow}}{yr}\right) = CO_2\, shadow\, price\left(\frac{€}{kg}\right) * 28 * CH_4\, production\left(\frac{kg}{yr}\right) \quad \text{Eq. 12}$$

Note: The values in the report were in pounds, and were converted to € for the actual exchange rate (1 £ = 1.14 €).

- Scenario 3:

In this scenario, a quota is supposed to be applied on CH_4 emissions by restricting the number of animals. Hence, no cost was calculated for CH_4, and the economic value was derived considering the number of animals as non constant in the profit function.

- Scenario 4:

CH_4 represents a loss of dietary energy in ruminants because 2% to 12% of the total net intake energy is lost as enteric eructated CH_4 (Johnson and Johnson, 1995; Lassey et al., 1997; de Haas et al., 2011). Therefore, we can estimate the cost of CH_4 based on net energy loss from CH_4 emissions. An average of 6% of net energy loss (TNEL) was considered to estimate the cost of CH_4 emissions in this study. First, total net energy required (TNER) per annum was estimated for the animal model (cf. Table **10**) based on the equations of National Research Council, (2001). Table **12** presents the summary of the estimation of total net energy required (in Mega calories) for maintenance, production of milk, and pregnancy for an average lactating cow.

Table 12. Total energy (in mega calories) for the average lactating cow for maintenance, milk production and pregnancy per year

Maintenance	3,539.94
Milk production	6,737.84
Pregnancy	306.41
Total (Mcalories)	10,584.20

The cost of CH_4 emission due to energy loss can then be estimated as:

$$CH_4\ cost\left(\frac{€}{\frac{cow}{yr}}\right) = TNEL\,(\%) * TNER\,(Mcal) * 1\ Mcalorie\ cost\left(\frac{€}{Mcal}\right) \quad \text{Eq. 13}$$

where: TNEL is total net energy loss and TNER is total net energy required per year (standardized per year). As 1 UFL is equivalent to 1.73 Mcal of net energy

(Vermorel, 1978). Hence, the cost of one Mcalorie is assumed to be 0.11 €, giving that the cost of 1 UFL is 0.18€.

4. Economic values

Economic values (i.e. profit or loss (P) = revenue (R) – cost (C)) per cow per lactation, were estimated as a unit variation of each trait, while maintaining the other traits constants.

Economic values were derived only for production traits (milk volume, fat and protein yields) and CH_4. For the remaining traits, the economic values were kept as they are in the current ICO, and were (€/unit) 0.20, -2.06, and -1.58, respectively for longevity, SCC and days open.

For CH_4, economic values were derived in relation to each scenario that was considered in this study as follows:

Scenario 1:

In the current scenario, no economic value was derived for CH_4. The economic values were estimated only for production traits: Milk, Fat, and Protein.

Economic values (ev_{xi}), for a given trait (x_i), was derived from the profit function as follow:

$$ev_{xi} = \frac{\partial(B)}{\partial_{xi}} = \frac{\partial(R)}{\partial_{xi}} - \frac{\partial(C)}{\partial_{xi}}$$

where R and C are the revenue and costs of each trait in the bio economic model.

Scenario 2:

In this scenario, the economic value is derived considering a tax per kg of CH_4 emitted and is calculated as follow:

$$ev_{xi} = \frac{\partial(B)}{\partial_{xi}} = \frac{\partial(R)}{\partial_{xi}} - \frac{\partial(C)}{\partial_{xi}} - \frac{\partial(C_{CH_4})}{\partial_{xi}}$$

The economic value was derived considering the CH_4 emission cost estimated for scenario 2 (Cf. Eq. 12).

Scenario 3: Quota of CH_4 emissions

In the case of restriction in CH_4 production, the number of animals (n) is not constant. Therefore, the ev_{xi} of a trait x_i was calculated by the following equation (Adapted from Charfeddine, 1997):

$$ev_{xi} = \frac{1}{\mathrm{n}} \frac{\partial(B)}{\partial_{xi}} = \frac{\partial(R)}{\partial_{xi}} - \frac{\partial(C)}{\partial_{xi}} + \frac{1}{n}(R - C)\frac{\partial(n)}{\partial_{xi}}$$

The CH_4 produce quota, which defines the number of cows (n) per herd and lactation, was calculated as follow:

$$Q_{CH_4} = \mathrm{n} * CH_{4pro}$$

where Q_{CH_4} is CH_4 produce quota, n is the number of lactating cows, and CH_{4pro} is CH_4 production (kg/cow /yr).

The derivative of Q_{CH_4}, in a situation of quota with respect to CH_{4pro} emissions and its components is by definition, equal to zero:

$$\frac{\partial\left(Q_{CH_4}\right)}{\partial_{xi}} = \frac{\partial(n)}{\partial_{xi}}\left(CH_{4pro}\right) + n\frac{\partial\left(CH_{4pro}\right)}{\partial_{xi}} = 0$$

So,

$$\frac{\partial(n)}{\partial_{xi}} = -\frac{n}{CH_{4pro}}\frac{\partial\left(CH_{4pro}\right)}{\partial_{xi}}$$

Hence, the $\boldsymbol{ev_{xi}}$ of CH_4, in case of restriction, is:

$$ev_{xi} = \frac{\partial(R)}{\partial_{xi}} - \frac{\partial(C)}{\partial_{xi}} + \frac{1}{n}(R - C)\,(-\frac{n}{CH_{4pro}}\frac{\partial\left(CH_{4pro}\right)}{\partial_{xi}})$$

And hence:

$$ev_{xi} = \frac{\partial(R)}{\partial_{xi}} - \frac{\partial(C)}{\partial_{xi}} - \frac{1}{CH_{4pro}}(R - C)\frac{\partial\left(CH_{4pro}\right)}{\partial_{xi}}$$

Scenario 4: CH_4 as net energy loss

The economic value is derived considering a 6% of total net energy loss due to CH_4 emission and is derived considering the CH_4 emission cost estimated for scenario 4 (Cf. Eq. 13) and is calculated as follow:

$$ev_{xi} = \frac{\partial(B)}{\partial_{xi}} = \frac{\partial(R)}{\partial_{xi}} - \frac{\partial(C)}{\partial_{xi}} - \frac{\partial(C_{CH_4})}{\partial_{xi}}$$

1. Economic importance of traits in selection index

Giving that marginal economic values are expressed in different units, it is difficult to compare directly the economic importance between different traits. Therefore the economic importance of the traits in the selection index can be expressed in various ways. This importance was calculated for the traits in the selection index ignoring the economic weight of conformation traits, because they do not actually have an estimated economic weight , which is too complicated to define.

- The relative importance of traits

The relative importance of a trait is the result of the product of the weighing factor with the genetic standard deviation of the trait and then divided by the total of all traits, expressed in percentage (CRV, 2015).

- The relative weight of traits

The relative weight of a trait is calculated by multiplying the weighing factor with the response of the trait and then divided by the total of all traits, expressed in percentage (CRV, 2015) .

5. Response to selection

The response to selection for the indices under the different scenarios was assessed using a multi-trait selection index including the current traits in the ICO and CH_4. Giving genetic parameters , the number of observations of the progeny, and economic values of the traits in the aggregate genotype (Hazel, 1943) , the annual response to selection was predicted for each scenario using selection index theory, implemented in the software developed by J. van der Werf (https://jvanderw.une.edu.au/software.htm) , according to the following equation:

$$R_I = \frac{i\, r_{IH}\, \sigma_H}{t}$$

Where i is the selection intensity (i=1), r_{IH} is the correlation between the traits of the index I and the aggregate genotype H, σ_H is the square root of the additive genetic variance of the population, and t is generation interval. In this study, we have considered a t=3.6 (García-Ruiz et al., 2016) as generation interval.

The annual economic response to selection (R_e) is calculated by multiplying the genetic response per its correspondent economic value (ev_i) per unit of the trait in the selection index:

$$R_e = \frac{i\, r_{IH}\, \sigma_H\, ev_i}{t}$$

6. Sensitivity analysis

The sensitivity of response to selection was assessed considering different situations in order to understand the consequences of selection including CH_4 emissions:

i. Varying by 50% the genetic correlations between CH_4 and the traits in the selection index.

ii. Varying by 50% the economic weights of the CH_4 in the selection index. For CO_2 tax scenario, this coincides with the low and high scenario for the estimation of the shadow price of CO_2 (cf page 37), therefore this scenario was analysed apart.

iii. Considering the low and high scenario for the shadow price of CO_2 in carbon tax scenario, this coincides to a high extent with changing ±50% the economic weights of carbon tax scenario.

RESULTS & DISCUSSION

1. Genetic parameters

The average daily CH_4 production per cow was 224 g/day, with a standard deviation of 126.4. This mean is lower than the one reported in other studies conducted in other countries (Kandel et al., 2017b; Breider et al., 2018), which was around 400 g/day. This difference could be explained by the differences between the production systems and feeding strategies across countries and also by the differences between methods of measurements. In our study the data was collected using sniffer method that measures concentration, but not the flux of CH_4.

In this study, the estimated heritability is 0.38, showing that enteric CH_4 is very heritable trait in dairy cattle (Table 13). This value is a little higher than the one reported in other recent studies, probably because of the sample size of the dataset : 0.33 (Breider et al., 2018), 0.13 (Pickering et al., 2015a) , 0.25 (Kandel et al., 2017b), 0.25 (Brito et al., 2018). The genetic variance of CH_4 of 2281.12 (g/d) shows that there is high genetic differences between cows for daily CH_4 production, thus there is a potential in selecting for lower CH_4 emitting cows.

Table 13. Additive genetic variance (σ^2_a) and heritability (h^2) for CH_4 trait

Trait	$\sigma^2 a$	h^2
CH_4 (ppm/day)	111656.81 ± 50271.26	0.38 ± 0.16
CH_4 (g/day)	2281.12 ± 880.45	0.39 ± 0.13

CH_4: methane

The following table shows the genetic correlations between the EBVs of CH_4 production and the different traits of the ICO for 480 sires, evaluated in our data with BLUP methodology with pedigree.

Table 14. Genetic correlation ($rgCH_4$), between CH_4 and other traits in the ICO

Trait	$rgCH_4$
Milk	-0.17
Fat	-0.10
Protein	-0.14
FLI	-0.19
UCI	-0.21
Long	-0.09
SCC	0.08
DO	-0.07

FLI: Feet and Legs Index; UCI: Udder Composite Index; long: longevity, SCC: Somatic Cell Count; DO: Days Open; CH_4: Methane

It is observed that the genetic correlations are generally low and negatives (Table 14), showing that there is a large margin for improvement in emissions without harming the traits of interest of the ICO. Furthermore, the general trend suggests that we would obtain a lower production of CH_4 from lactating cows when we select based on the production traits, as well as when we select for better lifespan. These results are in concordance with those reported by (Kandel et al., 2017b) , who obtained negative and low genetic correlations between CH_4 and production traits (milk, milk fat and milk protein yields). However, other studies reported a positive correlation between CH_4 and production traits (Breider et al., 2018), and between CH_4 and conformation and fertility traits (Zetouni et al., 2018). This could be explained by the fact that some authors (de Haas et al., 2011) ,indicated that the genetic correlation between CH_4 emissions and production traits, changes throughout the weeks of lactation, with contrary signs to the start and the end of lactation. From a biological point of view this makes sense, as in early lactation cows have a negative energy balance. Therefore, the correlation between milk yield and CH_4 is expected to be negative. Later in lactation energy for milk production comes from dry matter intake and hence a positive correlation between production traits and CH_4 production is expected. It is possible that when obtaining one calculation for the whole lactation the negative correlation present for early lactation dominates the dataset.

The correlations with UCI and SCC suggests that selecting for lower emitting cows, results in better udder health . The very low and negative correlation between CH_4 and longevity, causes to think that the average emissions per cow and day do not determine the lifespan of the animal, as we are not dealing with the total emissions per year, but the average per week measured by the robot per animal.

Moreover, the correlation between CH_4 and the total merit index (ICO), which is -0.21, shows that more profitable cows produce less CH_4. Hence, to some extent selecting for better production and efficiency leads to select more efficient cows. Indeed , Amer et al., (2017) showed that selecting for better efficiency leads to selecting less emitting cows per unit of product (i.e emissions intensity), and stresses that emissions intensity, which is total GHG emissions generated per unit of product output could be used as an appropriate trait to reduce methane emissions in dairy cattle and to select for better efficiency. In this sense, Koenen et al., (2013) also concluded that past and current animal breeding strategies for improved production efficiency have indirectly also significantly reduced emissions. Our study aligns with these results, as the genetic response for CH_4 trait was negative in the benchmark scenario showing that we are actually selecting more efficient cows, but incorporating CH_4 directly into the breeding goal will allow to select directly for better efficiency.

However, selecting more efficient cows seems to impair fertility, probably because more efficient cows mobilize more reserves and have negative energy balance. Indeed, during years, selection for better production impaired fertility. However, lately, fertility has been recuperated because it was included in the selection indices (García-Ruiz et al., 2016). Thus, actually we have sires that are good for both production and fertility traits, and probably selecting more efficient cows could penalize slightly the genetic gain achieved in terms of fertility. In this sense, Zetouni et al., (2018), also reported that more fertile cows emit more emissions.

In summary, the low and negative genetic correlation between daily CH_4 production and the traits currently included in the Spanish breeding goal, suggests that selecting for better production and efficiency leads to a decrease in CH_4 production.

2. Economic values

The economic values that have been derived for production traits from the bio economic model (Table 15), are all positives (€/kg): 0.01, 1.94 and 4.48 respectively for milk volume, milk fat and milk protein yields. The economic values for production traits were different but not far from those calculated by Charfeddine and Pérez-Cabal, (2014) in Spanish dairy cattle for the actualization of the ICO: 0.01, 1.94 and 4.48 (€/kg) respectively for milk volume, milk fat and milk protein yields. The economic values of the three components of production are different due to the differences in the marginal production costs of each one and the milk payment system.

For CH_4, the economic values derived differed for each scenario. In the case of application of a tax for CO_2, the estimated economic value is -1.21 €/kg of CH_4. In a situation of restriction in CH_4 production (i.e quota of CH_4 emissions), the economic value of CH_4 was estimated to be -9.32 €/kg of CH_4 increased. When CH_4 economic value is derived based on its relation to net energy loss, the value is -0.67 €/kg of CH_4.

The remaining economic values for other traits were kept as they are in the current ICO.

Table 15. Economic values (€) derived for one unit of increase in production traits (milk, fat and protein yield) and CH_4, for the four scenarios. The economic values of the rest of traits are current values of the ICO

Traits	Units	Benchmark scenario	CO_2 tax	CH_4 quota	NE loss
Milk	kg	0.01	0.01	0.01	0.01
Fat	kg	1.94	1.94	1.94	1.94
Prot	kg	4.48	4.48	4.48	4.48
FLI	-	-	-	-	-
UCI	-	-	-	-	-
Long	days	0.20	0.20	0.20	0.20
SCC	log(SCC)	-2.06	-2.06	-2.06	-2.06
DO	days	-1.58	-1.58	-1.58	-1.58
CH_4	kg	0.00	-1.21	-9.32	-0.67

FLI: Feet and Legs Index; UCI: Udder Composite Index; long: longevity, SCC: Somatic Cell Count; DO: Days Open; CH_4: Methane

In beef cattle, similar studies have been developed. López-Paredes, (2018) developed a bio economic model that included CH_4 emissions into the breeding goal, and investigated different strategies for mitigating these emissions in beef cattle in Spain. Quinton et al., (2017) predicted the effects of beef selection indices on greenhouse gas emissions and showed that genetic improvement in production efficiency traits can also drive reduction in greenhouse gas emissions.

In dairy cattle , Bell et al., (2016) in Australia and Breider et al., (2018) in the United Kingdom have estimated the economic value of enteric methane emissions in dairy cattle and investigated its relationship with other biological traits. The economic value estimated in our study in the case of a CO_2 tax is close to the value reported by Bell et al., (2016), which is -1.68 £ per kg of CH_4 (≃ 1.77 €/kg of CH_4) .

3. Economic importance of traits in selection index

The relative importance of the traits expressed in percentage of economic importance in the selection index under each scenario are shown in Figure 13, and **¡Error! No se encuentra el origen de la referencia.** presents the relative weights of traits in selection index based on the response to selection under the four scenarios. This economic importance was calculated ignoring the economic weight of conformation traits (FLI and UCI), which are too complicated to estimate, and therefore we assumed that they do not have an economic weight.

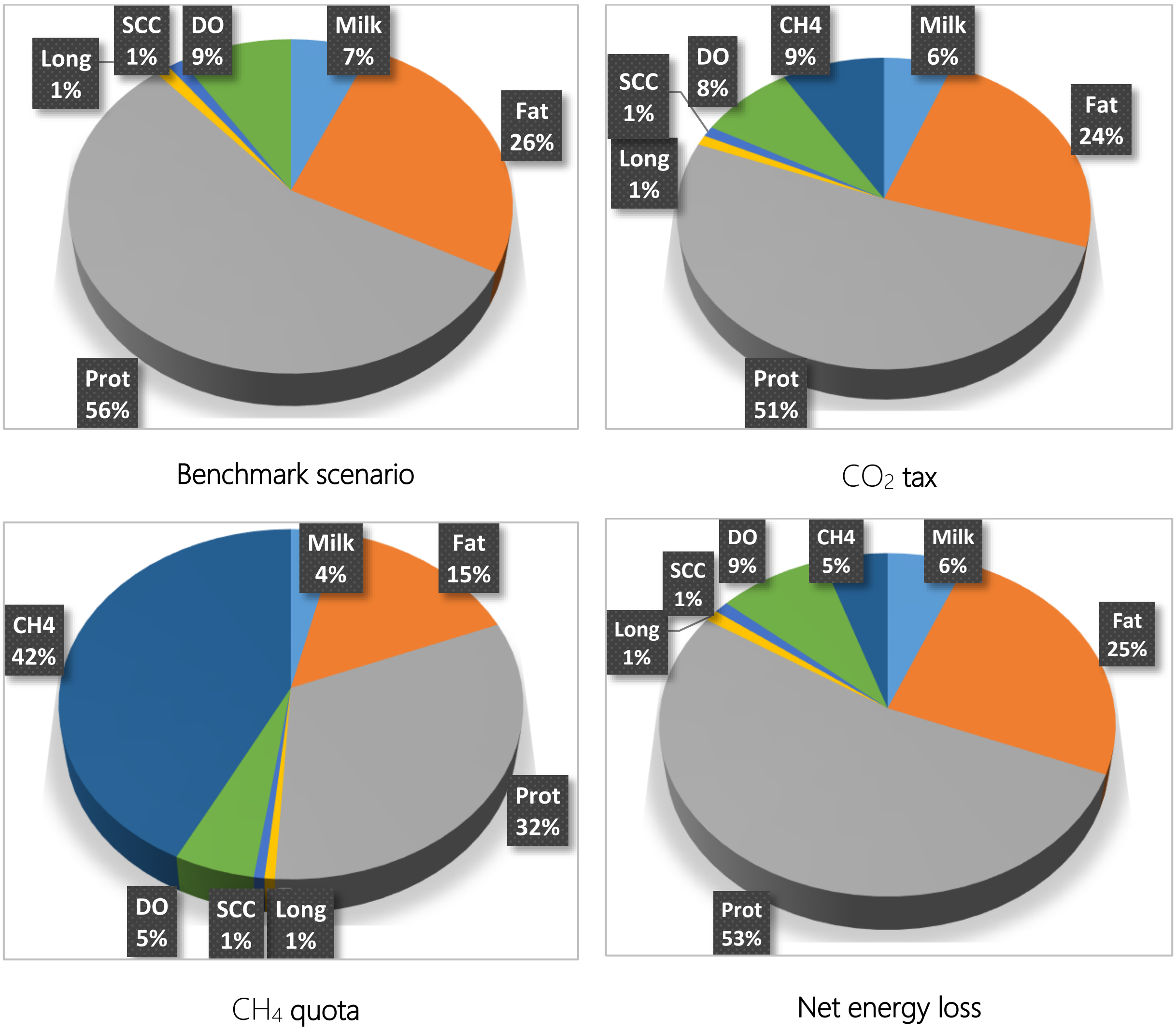


Figure 13. Relative economic weights (%) of the production, functional and CH_4 traits included in the index for scenario 1 (Benchmark), scenario 2 (carbon tax), scenario 3 (CH_4 quota) and scenario 4 (net energy loss)

FLI: Feet and Legs Index; UCI: Udder Composite Index; long: longevity, SCC: Somatic Cell Count; DO: Days Open; CH_4: Methane

The protein is the most important trait in all scenarios, except in the case of quota. The relative importance of CH_4 varies from in NE loss scenario to in a situation of quota. In this case, CH_4 tends to be the first important trait in the index.

The estimated economic values for CH_4 depends on the adopted strategy. Therefore, the economic importance of CH_4 in the selection index varies between the scenarios.

In Scenario 1 (including CH_4 emissions without an economic value on it), the functional and production accounted for 89% and 11% of the index, respectively. The inclusion of CH_4 as a carbon tax (Scenario 2) and net energy loss (scenario 4) in the index, decreased slightly the economic importance of the production and functional traits and CH_4 trait was included in the index with a 9% and 5% importance respectively for scenario 2 et 4. On the other hand, the CH_4 quota results in a decrease in the importance of the production traits to 51% and functional traits to 6% and arises the importance of CH_4 to 43%, giving more importance CH_4 trait to become the most important one in the index. Whilst in all other scenarios, the most important trait is milk protein yield. As a consequence of quota restrictions, changes in production levels, reduce the economic weight of productive traits, and to a higher extent, the economic weight of the protein and increases the economic weight of CH_4.

4. Selection indices and response to selection

Wall et al., (2010) proposed three routes that could reduce CH_4 emissions via genetic improvement by selection: (i) improving productivity and efficiency (e.g. residual feed intake, longevity), (ii) reducing wastage in the farming system, and (iii) directly selecting on emissions, when the CH_4 or a proxy is measurable. In this study, we focused on the third point which is selecting directly for less CH_4 emitting cows. Therefore CH_4 was included as a measurable trait into the selection index. Four scenarios have been investigated including one current scenario and three environmental indices (i.e. Carbon tax, CH_4 quota and net energy loss scenarios).

After including the genetic parameters and economic values in the selection index, we have obtained the results of the expected overall genetic and economic responses under the benchmark scenario and the three environmental indices expressed in units of the traits and in €/cow/year (Table *16*). The genetic response (in units) for each trait included in the selection index under the four scenarios is highlighted in Figure 14. The total economic impact of each scenario is shown in the total benefit expressed in € /cow/year. Indeed, in order to compare the generated benefit between scenarios, we multiplied the genetic responses of each scenario by the economic values of the given scenario which are compared to. Thus we obtained 4 situations under each scenario. The results are presented in comparison with the baseline scenario.

Selection under the current benchmark scenario generates an increase in benefits of 38.35 € per cow and annum, and results in a response to selection for CH_4 emissions without any economic value on it of -0.51 kg/cow/annum. The negative genetic response of CH_4 in the baseline scenario shows that, we are actually selecting for better efficiency in dairy cattle. To some extent these emissions are expected to be reduced as an indirect outcome of selecting for improved efficiency of the production system (e.g. fertility, longevity, feed efficiency) as it was stressed by Amer et al., (2017).

Incorporating CH_4 with an economic value on it in the index, leads to a reduction in CH_4 emissions (kg/cow/year) in all the environmental indices and the responses of CH_4 became (kg/cow/year) -0.86 , -2.41 and -0.70 respectively for CO_2 tax, CH_4 quota and net energy loss scenarios, with respect to the baseline scenario. We can infer from this that the higher the economic weight of CH_4 in the index, the higher the decrease in CH_4 emissions per cow and year.

Penalizing for CH_4 emissions through a carbon tax resulted in a reduction in the annual response for milk fat yield by -1%, , an improvement in genetic gain of FLI and UCI (+2%), and better longevity (+3%) . The genetic gain of the remaining traits was similar to the baseline scenario and no change was observed.

In the case of a CH_4 quota (i.e. scenario 3), all the traits tend to improve slower. Actually, Restricting CH_4 emissions reduces the economic responses for all the productive and functional traits in the selection index from -3% up to -21%, except for longevity and SCC which gave a favorable genetic response. The largest change in genetic gain is expected in days open and milk production traits in comparison to the baseline scenario.

No change in genetic gain was found in the production and functional traits when CH_4 was included as a net energy loss cost in the selection index. Besides, this index resulted in a better longevity (+2%) with respect to the benchmark scenario.

On the other hand, there was differences in the generated benefits between the three environmental indices with respect to the baseline scenario. If we select for a CO_2 tax on CH_4 emissions, the benefit will increase by about 1% with respect to the baseline index, thus, selecting under this scenario results in more profitable cows if a CO_2 tax is applied on CH_4 emissions in the future. When CH_4 was incorporated as a net energy loss, no change in total benefit was observed with respect to the reference scenario. The CH_4 quota scenario reduces to -21% the economic gain generated by the reference scenario.

In summary scenario 2 (i.e. CO_2 tax) would select for more efficient cows that have less CH_4 emissions with respect to baseline scenario, with better udder health and lifespan. Scenario 3 (CH_4 quota) results in selecting cows that are low CH_4 emitting and fertile, but less productive with reduced quality of conformation traits. Scenario 4 (i.e. net energy loss) leads to selecting more productive and efficient cows, that have a slow reduction in CH_4 emissions compared to scenario 2 and 3, but better than the benchmark scenario. Furthermore, generally the differences between the baseline index and the three environmental indices weights are more attenuated in the net energy loss index.

Table 16. Expected annual responses to selection (in units of traits and €/cow/year) under the four scenarios

Item	Units	Benchmark scenario		CO_2 tax		CH_4 quota		Net energy loss	
		Units	€/cow/yr	Units	€/cow/yr	Units	€/cow/yr	Units	€/cow/yr
Milk	kg	193.00	2.741	193.19	2.74	162.82	2.31	193.37	2.75
Fat	kg	5.89	11.412	5.84	11.31	4.64	8.98	5.87	11.37
Prot	kg	5.77	25.848	5.74	25.73	4.69	20.99	5.76	25.82
FLI	-	0.14	0.000	0.14	0.00	0.14	0.00	0.14	0.00
UCI	-	0.16	0.000	0.17	0.00	0.16	0.00	0.16	0.00
Long	days	0.54	0.107	0.56	0.11	0.56	0.11	0.55	0.11
SCC	log(SCC)	-0.01	0.02	-0.01	0.03	-0.02	0.05	-0.01	0.03
DO	days	1.13	-1.781	1.13	-1.78	0.94	-1.49	1.13	-1.78
CH_4	kg	-0.51	0.00	-0.86	1.04	-2.41	22.41	-0.70	0.47
Total benefit /scenario 1			38.35		38.14		30.95		38.28
Total benefit /scenario 2			38.96		39.18		33.87		39.14
Total benefit /scenario 3			43.06		39.18		33.87		39.14
Total benefit /scenario 4			38.69		38.72		32.57		38.76

FLI: Feet and Legs Index; UCI: Udder Composite Index; long: longevity, SCC: Somatic Cell Count; DO: Days Open; CH_4: Methane

Total benefit scenario 1: the generated benefit by the responses of each scenario using economic weights of scenario 1; Total benefit scenario 2: the generated benefit by the responses of each scenario using economic eights of scenario 2; Total benefit scenario 3: the generated benefit by the responses of each scenario using economic weights of scenario 3; Total benefit scenario 4: the generated benefit by the responses of each scenario using economic weights of scenario 4

Figure 14. Genetic response per annum for the traits included in the selection index, under the four scenarios.

FLI: Feet and Legs Index; UCI: Udder Composite Index; long: longevity, SCC: Somatic Cell Count; DO: Days Open; CH_4: Methane

5. Sensitivity analysis

Bekman and van Arendonk, (1993), showed that future economic values might change dependent on level of output and prices of economic weights to changes in price and production circumstances. In the case of this study, in order to understand the consequences of selection including CH_4 emissions when some key parameters were changed, the sensitivity of response to selection was assessed considering different situations (Cf. 6. Sensitivity analysis). The expected genetic responses in several traits when selecting under each of the considered situations of the sensitivity analysis are presented in Table 17 , Table *18*, and Table 19.

Table 17. Changes in genetic responses when genetic correlations between CH_4 and the traits in the selection index were changed by 50%, for the four scenario

Traits	Units	Genetic Standard deviation	Variation	Benchmark scenario		CH_4 quota		Net energy loss	
				Units	€	Units	€	Units	€
Milk	kg	800	+50%	193.00	2.74	169.32	2.40	193.70	2.75
			-50%	193.01	2.74	155.96	2.21	193.02	2.74
Fat	kg	24	+50%	5.89	11.41	4.70	9.11	5.87	11.36
			-50%	5.89	11.41	4.57	8.85	5.88	11.38
Prot	kg	22	+50%	5.77	25.85	4.81	21.53	5.76	25.82
			-50%	5.77	25.85	4.56	20.43	5.76	25.81
FLI	-	1	+50%	0.14	0.00	0.15	0.00	0.14	0.00
			-50%	0.14	0.00	0.12	0.00	0.14	0.00
UCI	-	1	+50%	0.16	0.00	0.17	0.00	0.17	0.00
			-50%	0.16	0.00	0.14	0.00	0.16	0.00
Long	days	10	+50%	0.54	0.11	0.62	0.12	0.55	0.11
			-50%	0.54	0.11	0.49	0.10	0.54	0.11
SCC	log(SCC)	1	+50%	-0.01	0.02	-0.03	0.06	-0.01	0.03
			-50%	-0.01	0.02	-0.02	0.03	-0.01	0.02
DO	days	10	+50%	1.13	-1.78	0.98	-1.55	1.13	-1.79
			-50%	1.13	-1.78	0.91	-1.43	1.13	-1.78
CH_4	kg	14	+50%	-0.78	0.00	-2.53	23.55	-0.97	0.65
			-50%	-0.23	0.00	-2.28	21.26	-0.45	0.31

FLI: Feet and Legs Index; UCI: Udder Composite Index; long: longevity, SCC: Somatic Cell Count; DO: Days Open; CH_4: Methane

The results showed that in all the tested situations, the total benefit was insensitive, except for CH_4 quota scenario, in which the profit was moderately sensitive when the economic weights and genetic correlations between CH_4 and other traits were increased by 50%. The most sensitive traits in terms of response to selection were CH_4, longevity and to a small extent somatic cell count for all the scenarios, but this response was highly sensitive in CH_4 quota scenario. Response to selection for production traits (Milk volume, milk fat and milk protein yields) and functional traits (UCI and FLI) was insensitive to the changes, except for scenario 3 where the response ranges from -1% up to +28% with respect to the same response in the baseline index. Hence, we can infer that the moderate economic weight of CH_4 in CO_2 tax and net energy loss scenarios if varied ±50% does not affect genetic responses of other traits in selection index. However when the economic importance of CH_4 in the selection index is high as in the case of CH_4 quota, any changes can affect the genetic responses of other traits up and down.

Table 18. Changes in genetic responses when the economic values of CH_4 in the selection index were changed by 50%, for the four scenarios

Traits	Units	Genetic Standard deviation	Variation	Benchmark scenario		CH_4 quota		Net energy loss	
				Units	€	Units	€	Units	€
Milk	kg		+50%	193.00	2.74	142.35	2.02	193.31	2.74
		800	-50%	193.00	2.74	184.31	2.62	193.27	2.74
Fat	kg		+50%	5.89	11.41	3.94	7.63	5.85	11.33
		24	-50%	5.89	11.41	5.42	10.50	5.89	11.40
Prot	kg		+50%	5.77	25.85	4.03	18.07	5.75	25.77
		22	-50%	5.77	25.85	5.40	24.19	5.77	25.84
FLI			+50%	0.14	0.00	0.13	0.00	0.14	0.00
	-	1	-50%	0.14	0.00	0.15	0.00	0.14	0.00
UCI			+50%	0.16	0.00	0.14	0.00	0.17	0.00
	-	1	-50%	0.16	0.00	0.17	0.00	0.16	0.00
Long	days		+50%	0.54	0.11	0.52	0.10	0.55	0.11
		10	-50%	0.54	0.11	0.58	0.12	0.54	0.11
SCC			+50%	-0.01	0.02	-0.03	0.05	-0.01	0.03
	log(SCC)	1	-50%	-0.01	0.02	-0.02	0.04	-0.01	0.02
DO	days		+50%	1.13	-1.78	0.82	-1.30	1.13	-1.78
		10	-50%	1.13	-1.78	1.07	-1.70	1.13	-1.78
CH_4	kg		+50%	-0.51	0.00	-2.79	38.98	-0.80	0.81
		14	-50%	-0.51	0.00	-1.69	28.38	-0.61	0.20

FLI: Feet and Legs Index; UCI: Udder Composite Index; long: longevity, SCC: Somatic Cell Count; DO: Days Open; CH_4: Methane

Table 19. Changes in genetic responses considering the low and high scenario for the shadow price of CO_2 in CO_2 tax scenario

Traits	Units	Genetic Standard deviation	Variation	CO_2 tax	
				Units	€
Milk	kg	800	low scenario	193.35	2.75
			high scenario	192.68	2.74
Fat	kg	24	low scenario	5.88	11.38
			high scenario	5.80	11.23
Prot	kg	22	low scenario	5.77	25.83
			high scenario	5.71	25.60
FLI	-	1	low scenario	0.14	0.00
			high scenario	0.15	0.00
UCI	-	1	low scenario	0.16	0.00
			high scenario	0.17	0.00
Long	days	10	low scenario	0.55	0.11
			high scenario	0.56	0.11
SCC	log(SCC)	1	low scenario	-0.01	0.03
			high scenario	-0.01	0.03
DO	days	10	low scenario	1.13	-1.78
			high scenario	1.12	-1.78
CH_4	kg	14	low scenario	-0.66	0.35
			high scenario	-1.00	1.71

FLI: Feet and Legs Index; UCI: Udder Composite Index; long: longevity, SCC: Somatic Cell Count; DO: Days Open; CH_4: Methane

6. Genetic trends

In order to evaluate the genetic trends of each strategy, we have simulated the expected genetic trends of CH_4 emissions in Spain for the next 10 years, in terms of genetic response of CH_4 in the selection index under the four scenarios, considering two situations. In the first situation the censes were considered as constant, and in the second situation, the censes was considered to decrease by 1% each year according to the previous tendency of past 10 years (scenario and CH4 quota. The decline of the censes of cows by 1% each year (Figure 15 A) results in a difference of +11% in CH4 emissions reduction with respect to the situation that considered the censes to be constants through the years (Figure 15 B).

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The general tendency showed that in both tested situations, if we maintain selection under the current scenario, CH_4 emissions would decrease slightly, as result of the selection for better efficiency. Selection for the CO_2 tax index, leads to reduction in CH_4 emissions as well, but the decrease is much higher with respect to the reference scenario. CH_4 emissions tends to fall in the quota scenario, which is due to restricting the number of animals. And when selecting for the net energy loss scenario, we notice a reduction in CH_4 emissions with respect to the baseline scenario, but this decrease is reduced with respect to the carbon tax scenario and CH_4 quota. The decline of the censes of cows by 1% each year (Figure 15 A) results in a difference of +11% in CH_4 emissions reduction with respect to the situation that considered the censes to be constants through the years (Figure 15 B).

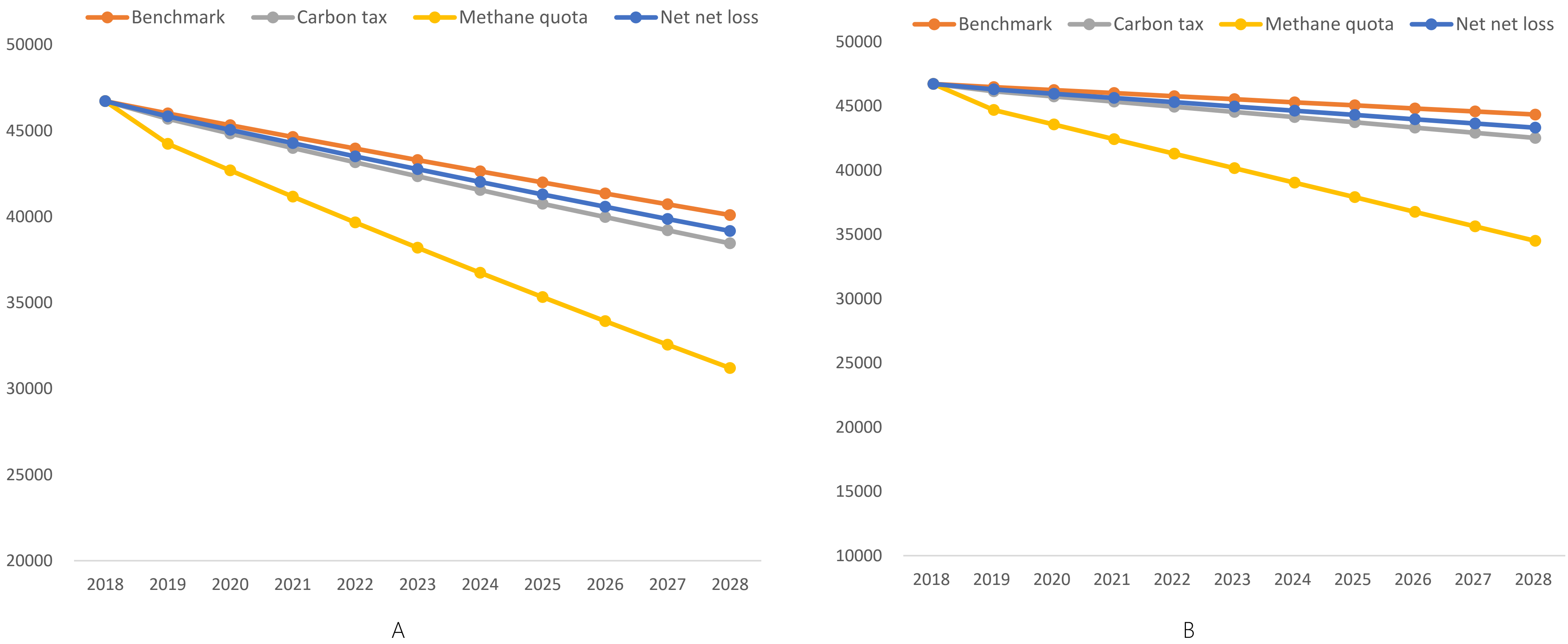

Figure 15. Expected methane emissions produced in Spain based on genetic gain in methane emissions in tones per annum under the four scenarios (i.e. number of cows*CH_4 genetic gain*time/1000); A: constant evolution of animal census thought the years; B: decrease of 1% in census each year. Source of census data: (http://www.conafe.com/VisorDocs.aspx?pdf=estadisticas_CENSO_DE_ANIMALES.pdf)

CONCLUSIONS

01 The genetic analysis showed that CH_4 emissions is a heritable trait in dairy cattle,

02 Selection for better efficiency leads to lower CH_4 emissions, and the most profitable cows emit less CH_4,

03 The expected genetic and economic responses in traits between the CO_2 tax and net energy loss indices with respect to the baseline index were negligible, whilst the quota system reduces the marginal benefits generated by the genetic improvement of a breeding program by -21% in Spanish dairy cattle,

04 A small impact on production and functional traits was observed when CH_4 was included as a carbon tax and as a net energy loss into the breeding objective,

05 It is possible to select directly for CH_4 production trait, and the adopted strategy for the incorporation of CH_4 in the selection indices, would determine to a large extent the type of the future,

06 This study showed that there is a potential in mitigating CH_4 emissions by genetic selection in dairy cattle while remaining profitable, but this potential can only be fully exploited if breeders are fully committed to effective implementation.

For future studies we recommend:

- As genetic correlation estimates that we obtain between methane and other traits are low with large confidence interval, and genetic gain depend on other traits, which depends on the sign of this genetic correlation. Therefore, we need robust genetic correlation estimates with other biological traits in the selection index with large dataset, in order to reduce the uncertainty of the confidence interval. Moreover, as new traits have been recorded and included in genetic evaluations, such as feet health information for the prevention and control of lameness in dairy cattle (CONAFE, 2018b). Hence, further genetic correlations with other new biological traits of economic interest needs to be estimated.
- To conduct surveys and sociological studies with breeders to investigate dairy cattle producers breeding decisions and to what extent they are aware of environmental objectives such as climate change mitigation whilst remaining profitable. We think that this is the first step before any implementation of any policy that aims to reduce the carbon footprint in dairy cattle via genetic selection.

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APPENDICES

Appendix 1. Input and output costs calculated for the animal model

	€/cow/lactation
Incomes	
Milk	324.4
Fat	1044.0
Protein	1525.0
calves	113.5
Culled cow	200.8
Total incomes	**3207.7**
Costs	
Alimentation	717.8
Rearing cost (Heifer)	578.7
A. Insemination cost	59.0
Medicines	115.0
Veterinary	50.0
Mastitis	35.0
Milking	103.0
Workforce	660.0
Total costs	**2318.5**
Gross Margin	**889.2**