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Review

Manure management and soil biodiversity: Towards more sustainable food systems in the EU

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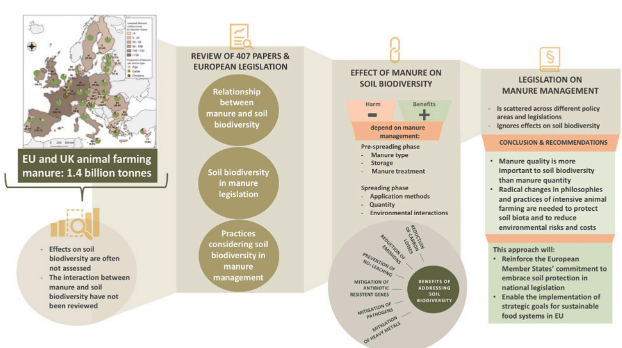
HIGHLIGHTS

- More than 1.4 billion t y^{-1} of manure are generated in the EU and UK and re-applied to soils with potential harmful effects.
- Current European regulations on manure disposal are scattered across different legislations and ignore soil biodiversity.
- This review of 407 documents revealed that manure quality is more important to soil biodiversity than manure quantity.
- To promote soil biodiversity by manure amendments, a radical transformation in the way agriculture is conducted is needed.
- This approach could reinforce EU MS' commitment to protect soils and improve future agricultural policies at the EU level.

GRAPHICAL ABSTRACT



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ABSTRACT

CONTEXT: In the European Union (EU-27) and UK, animal farming generated annually more than 1.4 billion tonnes of manure during the period 2016–2019. Of this, more than 90% is directly re-applied to soils as organic fertiliser. Manure promotes plant growth, provides nutritious food to soil organisms, adds genetic and functional diversity to soils and improves the chemical and physical soil properties. However, it can also cause pollution by introducing toxic elements (i.e., heavy metals, antibiotics, pathogens) and contribute to nutrient losses. Soil organisms play an essential role in manure transformation into the soil and the degradation of any potential toxic constitutes; however, manure management practices often neglect soil biodiversity.

OBJECTIVE: In this review, we explored the impact of manure from farmed animals on soil biodiversity by considering factors that determine the effects of manure and vice versa. By evaluating manure's potential to enhance soil biodiversity, but also its environmental risks, we assessed current and future EU policy and

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legislations with the ultimate aim of providing recommendations that can enable a more sustainable management of farm manures.
METHODS: This review explored the relationship between manure and soil biodiversity by considering 407 published papers and relevant legislative provisions. In addition, we evaluated whether benefits and risks on soil biodiversity are considered in manure management. Thereafter, we analysed the current legislation in the European Union relevant to manure, an important driver for its treatment, application and storage.
RESULTS AND CONCLUSIONS: This review found that coupling manure management with soil biodiversity can mitigate present and future environmental risks. Our analyses showed that manure quality is more important to soil biodiversity than manure quantity and therefore, agricultural practices that protect and promote soil biodiversity with the application of appropriate, high-quality manure or biostimulant preparations based on manure, could accelerate the move towards more sustainable food production systems. Soil biodiversity needs to be appropriately factored in when assessing manure amendments to provide better guidelines on the use of manure and to reduce costs and environmental risks. However, radical changes in current philosophies and practices are needed so that soil biodiversity can be enhanced by manure management.
SIGNIFICANCE: Manure quality in the EU requires greater attention, calling for more targeted policies. Our proposed approach could be applied by European Union Member States to include soil protection measures in national legislation, and at the EU level, can enable the implementation of strategic goals.

1. Introduction

1.1. Manure: Boon or bane?

In European legislation, there is no uniformly used definition for manure: While Regulation EC/1069/2009 on animal by-products defines manure as “any excrement and/or urine of farmed animals other than farmed fish, with or without litter”, being an organic fertiliser, the Nitrates Directive (Directive 91/676/EEC) defines manure from farmed animals as “waste products excreted by livestock: or a mixture of litter and waste products excreted by livestock, even in processed form”. The European Union (EU) produces a significant amount of organic fertiliser or waste - depending on the definition to be followed: 1.4 billion tonnes of manure from farmed animals were produced annually in the period 2016–2019 in the EU27 and UK (Eurostat, 2021a). Six large countries (DE, ES, FR, IT, PL, UK) produce ca. 68% of the total manure, while Ireland (84 million tonnes) and the Netherlands (73 million tonnes) also make important contributions. More than 75% of the produced manure derives from cattle, while pigs and chickens produced ca. 12% each (Fig. 1). However, the numbers of pigs and poultry/broilers increased significantly between 2010 and 2018, +25% and + 3%, respectivly (Eurostat, 2019). Due to increasing demand for meat and animal-based products, as well as an increase in the export of meat and dairy products, manure is increasingly generated in highly intensive farming systems (Bernal et al., 2015; Buckwell and Nadeu, 2016) and 4% of European farms produced 80% of the total amounts of manure in 2018 (Amann et al., 2018) (See Fig. 1 and Table 1).

The physicochemical properties of manure justify its wide use as a soil improver and organic fertiliser (Liang et al., 2014; Liu et al., 2010; Mandal et al., 2007; Saha et al., 2008). It improves soil physical and chemical properties (Kheyrodin and Antoun, 2011; Loro et al., 1997; Rayne and Aula, 2020; Unc and Goss, 2004), since it can slow down the rate of soil pH decline due to its high buffer capacity (Cai et al., 2015) and/or contribute to decrease aluminium toxicity (de la Luz Mora et al., 2017). Some studies also suggest that solid manure can raise soil pH due to the presence of potassium, sodium, magnesium and calcium (L'Herroux et al., 1997), calcium carbonates and bicarbonates (Whalen et al., 2000) and organic anions (Butterly et al., 2013), increasing the buffer and cation exchange capacities. However, manure's effects on soil pH depend on its initial value (Tang and Yu, 1999), the diet of animals (Butterly et al., 2013), the amount of manure applied (Hao and Chang, 2002), and its treatment prior to its application (Cavalli et al., 2016; Cavalli et al., 2017). Manure also provides essential mineral nutrients, such as inorganic nitrogen in the form of ammonium (NH_4^+; Geisseler et al., 2010), carbon (Francioli et al., 2016), phosphorus and sulphur (Liu et al., 2020), and metals such as zinc and copper (Bünemann et al., 2006; Delgado et al., 2012). Consequently, in a long term perspective, manure could potentially substitute part of mineral fertilisers, decrease farmers' costs and reduce the EU dependency on imports of phosphorus from other countries (Drangert et al., 2018; Garske et al., 2020). Importantly, manure substantily increases soil carbon stocks (Liu et al., 2020) and stabilises organic matter content and C/N ratios in a more stable fraction over the long term (Cui et al., 2018; Gong et al., 2009; Zhang et al., 2014b), which is the key driving factor for soil microbial diversity (Cui et al., 2018).

The nutrient added to soils via manure enhances enzyme (Liang et al., 2014; Watts et al., 2010) and microbial activity (Watts et al., 2010), as well as the abundance and biomass of soil fauna (Bengtsson et al., 2005; Birkhofer et al., 2008; van Eekeren et al., 2009). Benefits of increased fungal diversity by bovine manure application, for example, include minimized dry rot of potatoes (Gleń-Karolczyk et al., 2018). A meta-analysis found a more active and enhanced size of microbial populations in manures (pig, cattle, chicken, horse) controlling plant-feeding nematode populations (Liu et al., 2016). However, fertilising effects of manure and its influence on soil biodiversity are highly dependent on its chemical composition, which varies based on animal feed and species (Kerr et al., 2006) and treatment/processing methods (Jørgensen and Jensen, 2009; Risberg et al., 2017). Estimates from 2011 suggest that approximately 92.2% of nutrients in manure are being returned to fields with little or no processing (Foged et al., 2011). Direct application of manure is related to insufficient manure storage capacities (Buckwell and Nadeu, 2016). In 2010, only 31% of animal farms had storage facilities for manure and Germany, Italy, Belgium, Malta, Cyprus and the Netherlands exceeded recommended thresholds for animal density in relation to manure storage capacities (Eurostat, 2013). However, the amount of processed manure by waste category (for animal faeces, urine and manure) increased from 8.4 million tonnes in 2004 to 9.52 million tonnes in 2018 (Eurostat, 2021b), including manure recycled to compost or anaerobic digestates (Eurostat, 2010).

When the dry mass exceeds 20%, manure is considered solid, whereas slurry has a dry mass ranging between 4% and 20% and manure with a dry mass below 4% is considered liquid (Eurostat, 2021c). Since nitrogen (N) in solid manure is bound to organic matter, its mineralisation is a relatively slow process, persisting in soils for up to 5–10 years after its application (Webb et al., 2013). Liquid manure/urine from herbivores mainly provides mineral nitrogen in the form of ammonium (NH_4^+; Webb et al., 2013), which is readily plant-available especially in N-limited environments (Mooshammer et al., 2014). However, excessive manure applied during long periods will cause the accumulation of phosphorus and potassium and excessive N surpluses will cause high leaching rates of N (Geng et al., 2019). Particularly, when combined N application rates from manure and other N fertilisers exceed plant demand, N made available can exceed plant uptake capacities, resulting in increased pollution risks (Geng et al., 2019; Meng et al., 2005). Of all

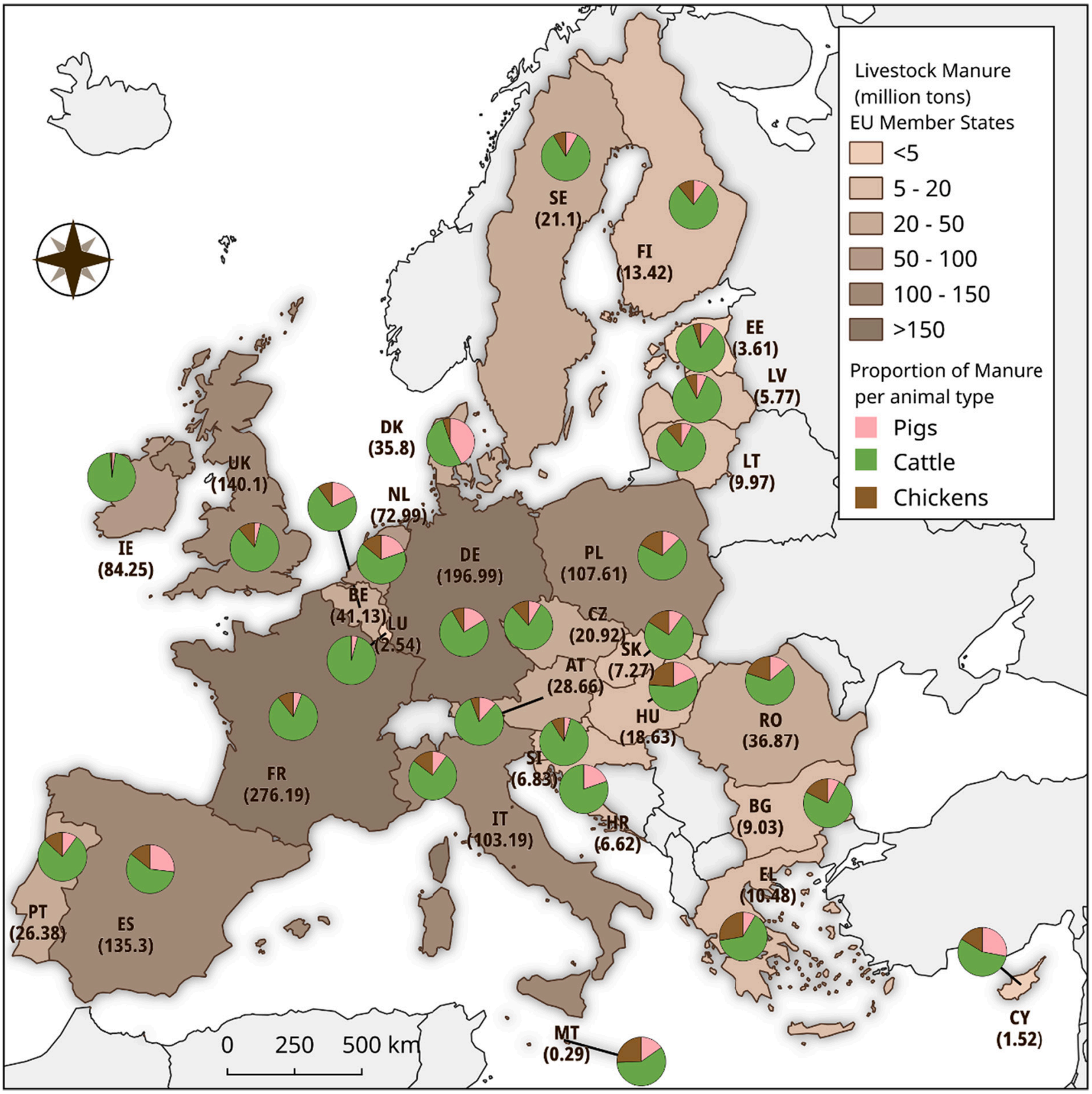

Fig. 1. Annual manure production (million tonnes) in the European Union and UK and distribution according to main animal types (Period: 2016–2019).

agricultural emissions, farmed animal manure contributed 18.6% of methane (CH_4) and 11.3% of nitrous oxide (N_2O) emissions in the EU27 + UK in 2015 (Eurostat, 2018). Agriculture contributes 93.3% of the overall ammonia emissions in the EU27 + UK in 2013, whereby animal manure contributed around 58.9% considering the capture, storage, treatment and use of animal manure (Eurostat, 2015).

Table 1
Research protocol leading into research questions and keywords. The Research Questions in the first column will be addressed in the corresponding sections.

Research Question (RQ)	Aim	Method	Keywords in the Search
RQ1: Which factors regulate the direct and indirect effects of farm manures on soil biodiversity and their implications on the fate of manure additions? (Section 3.1)	To identify the factors determining the impact of manure on soil biodiversity including benefits and threats to soil biodiversity as well as the effects of soil biodiversity on the fate of manure	To perform a systematic literature review on the effects of manure on soil biodiversity and vice versa	Manure management and/or animal faeces and/or animal dung and/or animal urine, benefit and/or harm, soil biodiversity
RQ2: Which practices help to achieve sustainable manure management in the EU? (Section 3.2)	Recommend best practices for integrating soil biodiversity in manure management to enhance benefits of manure for soil biodiversity	To examine sustainable farming practices for the role and integration of manure	Manure management, and/or sustainable agriculture, and European Union
RQ3: What role and importance, if any, is attributed to soil biodiversity in current European legislation on manure management? (Section 3.3)	To investigate the extent to which European policy instruments integrate soil biodiversity and manure management	To examine the integration of manure management and soil biodiversity in legal frameworks	Manure management, and/or soil biodiversity, and/or policy instruments, and/or European Union
RQ4: Which shortcomings in regulations and practices, if any, currently prevent sustainable manure management in the EU? (Section 3.4)	To determine knowledge gaps and limitations in current manure management practices and regulations to recommend sustainable manure management in the EU	To evaluate and combine/match the findings derived from the two previous methods	Manure management, shortcomings and/or limitations, and/or sustainable agriculture, and European Union

Additional pollution risks from manure result from the substantial use of animal food supplements (e.g., copper and zinc supplements in pig and poultry farming) in intensive animal farming that end up in the manure applied to soils (Moral et al., 2008; Provolo et al., 2018). In the EU, 150 million pigs consume more than 6.2 million tonnes of copper through feed additives (Panagos et al., 2018). Continuous additions of heavy metals contaminate soils, groundwater and affect organisms living in soils, whereby microorganisms are the first organisms being harmed (He et al., 2005). Heavy metals become more mobile in acidic soils (He et al., 2005), which long term applications of mineral fertilisers contribute to (Czarnecki and Düring, 2015). Manure may contain pathogens, which is particularly the case in untreated slurry from pigs kept in high densities (Guan and Holley, 2003; Unc and Goss, 2004; Venglovsky et al., 2018). Via manure applied to soils, zoonotic pathogens may enter the human food chain (Bicudo and Goyal, 2003). Antibiotics, used to maintain animal health and to indirectly boost animal growth (Kümmerer, 2009), also reach soils via manure. Various antibiotics in manure are mobile (Heuer et al., 2011; Zhao et al., 2019), harming plants (Zhou et al., 2020) and invertebrates (Sengeløv et al., 2003; Žižek et al., 2011). The adsorption of antibiotics depends on the type of antibiotics and soil properties (pH, clay content, organic matter and cation exchange (Wang and Wang, 2015)). The widespread use of antibiotics increases antimicrobial-resistant genes (You and Silbergeld, 2014), which may disseminate in soil bacteria through horizontal gene transfer (Xie et al., 2017), jeopardizing ecosystem health (Zubair et al., 2020).

1.2. The relevance of soil biodiversity and the impact of intensive farming systems

Soil organisms and their diversity are critical drivers for ecosystem processes (Smith et al., 2015). Their functional diversity drives the multifunctionality of soil systems (Konopka, 2009; Wagg et al., 2014; Wardle et al., 2000). Within the soil food web, microorganisms and soil fauna (micro-, meso- and macrofauna) have a dominant role in shredding, transforming and decomposing soil organic matter (Brussaard, 2012; de Vries et al., 2013; Frouz, 2018). As such, soil organisms drive nutrient cycling (Osler and Sommerkorn, 2007), including N fixation (Singh et al., 2016), plant nutrient and water uptake (Geisseler et al., 2010), plant productivity (Van Der Heijden et al., 2008) and pathogen and antibiotic resistance (Chen et al., 2017; Podmirseg et al., 2019).

Intensive agricultural systems have been shown to cause profound alterations in soil biota abundance and activities (Orgiazzi et al., 2016a; Tsiafouli et al., 2015). In the EU, most agricultural soils suffer from soil biodiversity losses due to soil erosion, climate change, agricultural intensification and diffuse soil contamination (Jeffery and Gardi, 2010; Orgiazzi et al., 2016b). Recent studies have shown that the application of untreated slurry can harm soil biota by introducing antibiotic resistance genes (Chen et al., 2017; Pérez-Valera et al., 2019) and pathogens (Goberna et al., 2011) or broad-spectrum antiparasitic controls, thus reducing the diversity of dung fauna (Adler et al., 2016; Iglesias et al., 2006). Also, dietary supplements such as heavy metals (Qi et al., 2018) or hormones (and their hormone-like by-products such as bile-acids) have been shown to negatively impact soil organisms (Mendelski et al., 2019). Ammonium-based mineral fertilisers and urea could accelerate soil acidification (Goulding, 2016) and in turn, the mobility of metals (Czarnecki and Düring, 2015), and be responsible for the abrupt declines in microfauna biomass and diversity (Chen et al., 2019). In relation to this, Rousk et al. (2010) also found bacterial communities to be strongly impacted by decreases in soil pH. Mineral fertilisers also significantly alter the structural and functional community composition of microbes (Manoharan et al., 2017; Prashar and Shah, 2016; Wang et al., 2016). For example, they stimulate ammonia-oxidizing bacteria, which are predominantly responsible for N_2O emissions (Meinhardt et al., 2018), increase the abundance of aphid pests (Gagic et al., 2017) or decrease the activities of arbuscular mycorrhizas (de Souza and Freitas, 2018) and gut microbiota in collembolans (Ding et al., 2019b). Negative effects on soil biota increase when mineral fertiliser are applied over a long period (29 years), compared to the application of manure treatments consisting of composted farmyard manure and biodynamic preparations (Faust et al., 2017; Sradnick et al., 2013) and when they replace carbon-rich organic fertilisers, as was found for nematode community structure and functions (Liu et al., 2016). Declined soil biodiversity reduces long term nutrient availability to plants (Wagg et al., 2014); in fact, intensive farming systems often have low nutrient retention (Lago et al., 2019; Schrama et al., 2018). Mineral fertilisers do not add organic carbon to soils. Therefore, the decline of soil organic matter and soil biodiversity may cause a positive (self-accelerating) feedback loop with detrimental environmental consequences (Fig. 2).

Soil organic matter availability and soil biodiversity are closely interlinked (Thiele-Bruhn et al., 2012): Manure is a source of organic carbon feeding soil organisms, whereby ecosystem services provided by microorganisms increase organic carbon pool in the long term (Cui et al., 2018; Gong et al., 2009; Zhang et al., 2014b) and therefore, can contribute to more sustainable agriculture on the condition that certain factors are fulfilled, which this paper aims to review.

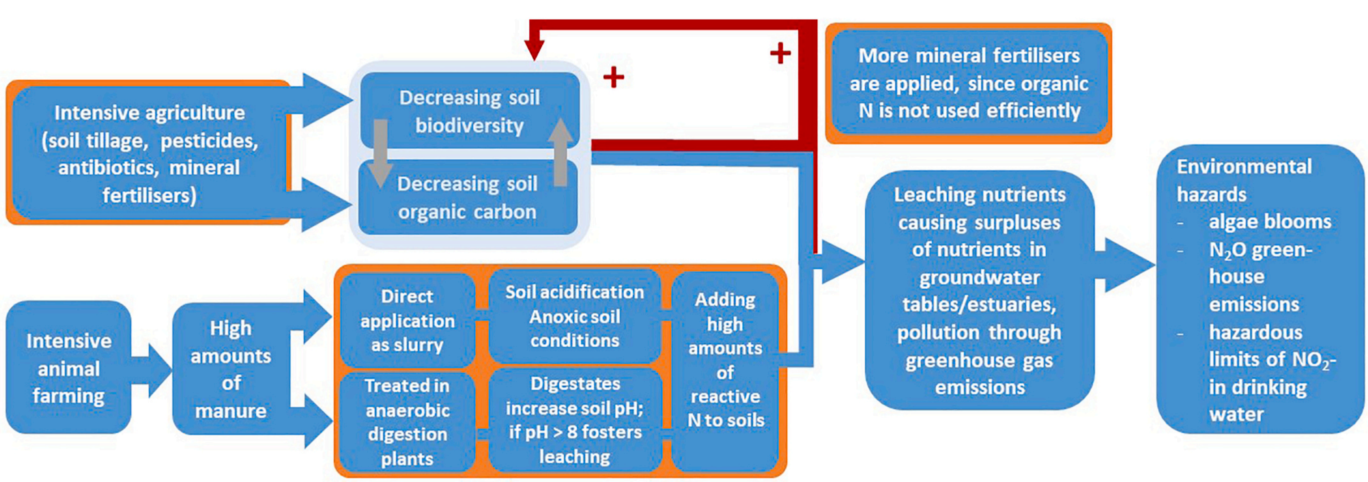

Fig. 2. The links between organic matter, nitrogen surpluses and soil biota - impact & consequences (simplification; dependent on various environmental characteristics such as soil properties, climate, type of organic fertiliser and land-use practices).

1.3. Manure and its management under current EU legislation

Solutions for manure management are among the priorities of the European Union (European Commission, 2020; European Commission, 2014). Currently, manure is not regulated under one single EU regulation or directive. Instead, various different directives, regulations and national Member State laws impact its management and quality. For example, the Nitrates Directive (Council Directive 91/676/EEC) requires Member States (MS) to implement national legislation governing the timing and amount of manure in nutrient vulnerable zones (Oenema et al., 2011), the Fertilising Products Regulation 2019/1009 indirectly lays down the treatment of manure impacting the quality of organic fertilisers and the Industrial Emission Directive requires MS to choose from a set of best available techniques for better handling pig and poultry manure (2010/75/EU). Although nutrient budgets (Klages et al., 2020) and other good agricultural practices set by MS (such as allowed application amount in nutrient vulnerable zones, storage practices) have improved the use efficiency of organic fertilisers, including manure from farmed animals (Dalgaard et al., 2014; Mihailescu et al., 2014; van Grinsven et al., 2015), the nitrogen surpluses did not decrease in several MS between 2010 and 2015 (European Environmental Agency, 2019). In 2013, 26.6% of the monitored sites in EU had increased nitrate values (European Commission, 2013a). The European Commission called for reducing nutrient losses by 50% in its Green Deal (in the Farm to Fork Strategy and the Biodiversity Strategy 2030) (European Commission, 2020d); therefore, more studies are investigating the recycling and nutrient recovery potentials of manure (Huygens et al., 2020; Huygens et al., 2019). Review work (Bloem et al., 2017), reports (Saveyn and Eder, 2014) and legislation (Fertilising Products Regulation EC 2019/1009) have focused on enhancing the effectiveness and safety of fertilising products, only including processed manure. Effects on soil biodiversity were not considered – mostly since the scope of legislation in place does not integrate soil biodiversity. While Blaustein et al. (2015) explored the release and removal of pathogenic microorganisms deposited with manure, excluding beneficial microorganisms and effects for soil fauna, the effects of manure on soil biodiversity and vice versa (Chen et al., 2020) have not been considered adequately in literature. Also, those interactions have not been put into a policy context. Recently, several publications looked at EU policy developments relevant to sustainable soil management (Montanarella and Panagos, 2021; Römbke et al., 2016). Sustainable manure management (Hou, 2016; Malomo et al., 2018) and practices for nutrient recovery from manure at the EU level (Huygens et al., 2020) gained more importance. Nonetheless, scientific papers on sustainable manure management (Hou et al., 2017; Malomo et al., 2018) and reports produced by the EU and European advisory boards on soil biodiversity neglect the risks and benefits of manure for soil biodiversity. They do not consider the importance of autochthonous soil biota that are added via manure (EASAC, 2018; Huygens et al., 2019, 2020; Orgiazzi et al., 2016a).

This review analyses the current directives and regulations in place for manure management in the European Union, assesses its impact on soil biodiversity and explores how the incorporation of soil biodiversity could enhance the solutions currently in place. The relationship between manure management and soil biodiversity will be explored, both in the published literature and European legislation, as a means of synthesizing the main findings and identifying critical limitations and recommendations. For the latter, we will investigate manure's potential to be a part of sustainable soil management enhancing soil biodiversity. Potentially, this includes guidelines to incorporate better manure management in future regulatory policies. In particular, the scope of the review is to focus only on livestock manure and does not include other organic wastes applied to agricultural lands even in smaller quantities. This review proposes creative solutions that integrate soil biodiversity promotion by addressing manure management at a European scale.

2. Materials and methods

This review follows the method proposed by Torraco (2005) to analyse existing literature and legislative documents. Therefore, we divided the literature review process into three phases: planning (theoretical background), execution (search strategy) and result analysis. The first phase defines research questions and a transparent search strategy to select relevant studies. Thereafter, the relevant information is extracted by performing systematic research, which is then synthesized and discussed to develop policy recommendations.

2.1. Research questions

As farm manure additions can have both beneficial and harmful effects on soil biodiversity, it is necessary to study all environmental impacts of animal manures. Accordingly, this study includes both direct and indirect effects of manure on soil biodiversity, but also their feedback responses on the fate of manure. Based on this assumption, the research questions and keywords defined in this review work are listed in Table 1.

2.2. Data collection and search strategy

The search was done in well-known literature databases such as the Web of Science (WOS) and Scopus. Before 1990, only few articles addressed the effects of manure on soil biodiversity (the first article on this topic and recorded in WOS was published in 1991, whereas in Scopus the first published article appeared in 1997), therefore our literature search included the peer-reviewed literature published between 1990 and February 2021. We also selected peer-reviewed publications in JSTOR, Routledge (Taylor & Francis), Google Scholar and Research Gate. A Boolean search by keywords applied a topic search in WOS and a TITLE-ABS-KEY search in Scopus (see Keywords in Table 1). Additionally, relevant governmental documents and grey literature from organisational websites from the European Union were systematically screened, including reports and statistics published by the European Commission or by agencies of the European Commission.

Relevant articles were selected based on the following inclusion criteria: (i) articles referring to manure management in the European Union and published in peer-reviewed scientific journals between 1990 and 2021 as described above and (ii) grey literature by the European Commission or by agencies linked to the European Commission or on behalf of the European Union (e.g., consultancies, assessments); (iii) legislative documents relevant to manure. The final database included 407 articles (for the full list see supplementary Table 1). The majority of the inputs in the database are scientific articles (87%) followed by grey literature (8%) and legislative documents (5%) (Fig. 3A). There is a huge increase in documents (publications, grey literature, legislation) with reference to manure during the last decade as ¾ of the documents were published in the period 2011–2020 (Fig. 3B).

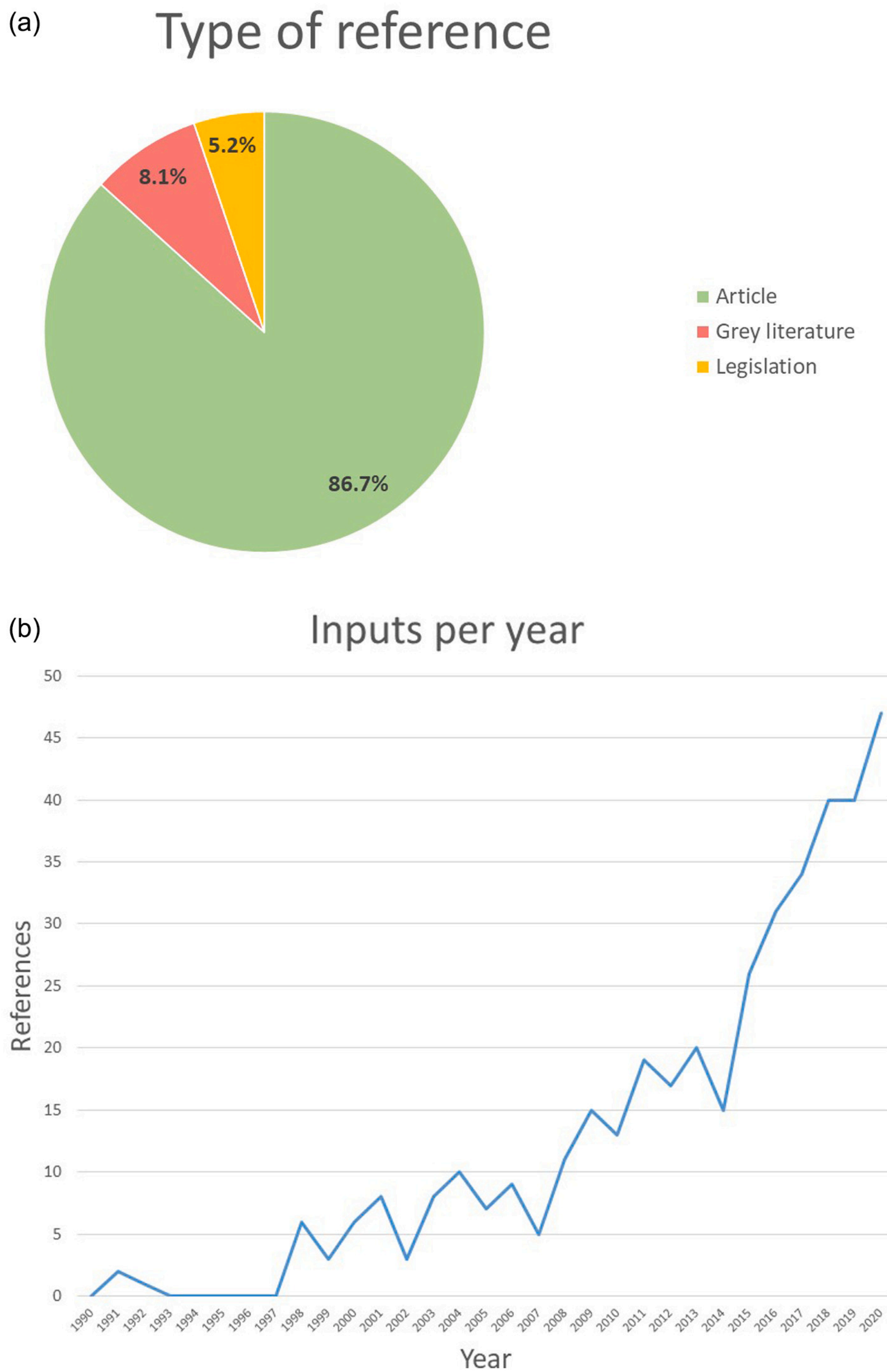

Fig. 3. A. Proportion of the inputs (Scientific Articles, Grey literature, Legislative documents) in our Database in the last 30 full years (1990–2021). B. The trend of inputs in our Database (Scientific Articles, Grey literature, Legislative documents) in the last 30 full years (1990–2020).

3. Results

3.1. *Factors determining the impact of manure additions to soil biodiversity in agroecosystems*

The main factors determining manure's impact on soil biodiversity are manure quality and its management (Blaustein et al., 2015; Bünemann et al., 2006). Both can either threaten or enhance soil biodiversity (Fig. 1). Variables impacting soil biodiversity are intertwined, therefore, focusing only on one parameter is not sufficient to predict effects. Various factors such as the dose, the composition of manure and the type of soil must be considered (Serafini et al., 2020). We divided variables depending on two management phases:

1) Management I considers variables managed in the pre-spreading phase (manure type, storage and treatment)
2) Management II considers variables managed in the spreading phase, including the application methods, quantity and environmental interactions.

3.1.1. Management I: pre-spreading phase

Decisions during the pre-spreading phase of manure include animal fodder, medical treatment given to animals, and storage and treatment of manure, considerably affect the quality of manure (Lekasi et al., 2003; Ndambi et al., 2019; Tittonell et al., 2010).

(i) Manure Type: The animal species from which the manure originates determine the diversity of microorganisms in manure. For example, bacteria isolated from ruminants are very distinct from swine manure (Whitehead and Cotta, 2004). Kim et al. (2014) found cattle manure to contain higher diversity of methanogens compared to pig manure. Loh et al. (2005) compared the effects of cattle and goat manure on the abundance and growth rate of earthworms in vermicompost, finding cattle manure to be more nutritious for earthworms. This indicated enhanced mineralisation, whereby the nitrogen content increased during the vermicomposting (Loh et al., 2005). A 13-year study found apart from pasture management, both poultry litter and cattle manure to increase soil microbial biodiversity (Yang et al., 2019a). Ansari and Mahmood (2017) found both goat manure and poultry manure with inoculated rhizobium to increase microbial population and enzyme. Murchie et al. (2015) found cattle slurry beneficial for earthworm populations, whereas pig slurry had no effect. However, Sun et al. (2016) found pig manure to diversify the fungal community more than cow manure when applied together with mineral fertilisers.

The content of organic carbon in manure varies by the animal species from which it originates. Moral et al. (2005) found horse, pig and rabbit manures to have the highest total organic carbon content, which was mostly determined by the organic matter concentration (manure consisted of faeces and straw from the bedding). The fraction of easily biodegradable organic compounds, which is also highly water-soluble, was highest in goat, chicken and horse manure. Amendments to manure enhance its organic matter content, which can either be added manually (Michel Jr et al., 2004) or by incorporating the bedding material such as straw, woodchips and sawdust (Petric et al., 2009). Replacing proteins with wheat in pig diets had no impact on tested Collembola and earthworm species (Serafini et al., 2020). Both cattle manure and swine manures add high amounts of organic carbon, but cattle manure contained higher amounts of dissolved organic carbon (Kim et al., 2014).

Feed supplements may be responsible for the presence of toxic contaminants in manure (Bolan et al., 2004) and heavy metals (Guan et al., 2011; Leclerc and Laurent, 2017), such as zinc and copper added in intensive piglets and poultry farming to promote their growth and prevent diarrhea (Gans et al., 2005; Jensen et al., 2016). A high concentration of copper and zinc can lead to their soil accumulation, particularly when the high density of pigs is combined with clay and alkaline soils (higher pH; Panagos et al., 2016). Once introduced to soils, heavy metals can alter the function of soil organisms, e.g., shifting from metal-sensitive to metal-resistant individuals (Giller et al., 1998). Depending on the initial state of soil biodiversity, metal stress may increase or decrease microbial diversity (Giller et al., 2009). However, Gans et al. (2005) found that already small heavy metal concentrations significantly harmed microbial diversity. Zinc, copper, arsenic, manganese and cadmium significantly influence the composition of microbiota (Li et al., 2020). An increasing concentration of copper sharply decreases bacterial richness and evenness (Nunes et al., 2016). Also, Xu et al. (2019) found metals to significantly inhibit the microbial activity but also the production of microbial biomass carbon. In the same study, the fungal population reacted more sensitively to metal toxicity Xu et al. (2019). In high amounts of pig manure amended to soils, the zinc and copper concentration reduced earthworms (Segat et al., 2019), possibly due to affected reproduction rates (Segat et al., 2015). Earthworms are the soil invertebrates most harmed by an elevated copper concentration (Naveed et al., 2014) and heavy metals were found to accumulate in earthworms after the application of pig manure, depending on the bioavailability of metals in pig manure (Li et al., 2010). Earthworms avoided soils spiked with copper and zinc (Lukkari et al., 2005) and concentrations of 272 and 156 mg kg^{-1} for Cu and Zn, respectively caused a 100% mortality. Domínguez-Crespo et al. (2012) found 323.11 mg kg^{-1} Cu and 84.12 mg kg^{-1} Zn to cause the death of 100% of earthworms. Apart from metals, also hormones, fed to cattle to enhance growth and milk production (Bartelt-Hunt et al., 2012), may end up in manure (Kjær et al., 2007), posing a potential risk for invertebrates (Mendelski et al., 2019).

Veterinary products fed to animals are often poorly metabolized, e. g., between 30 and 90% of antibiotics are excreted in urine and faeces (Alcock et al., 1999). When introduced to soils, antibiotics significantly alter soil microbial community biomass (Eguchi et al., 2012; Hammesfahr et al., 2011) and composition (Ding and He, 2010; Hammer et al., 2016). For example, the antibiotic sulfamethoxazole resulted in an initial decrease in bacterial growth rates, selecting more tolerant species (Demoling et al., 2009). Combining copper and the antibiotic oxytetracycline significantly declined functional microbial biodiversity (Kong et al., 2006). Also, antibiotics reduced earthworm gut-associated microbial diversity (Ma et al., 2019) and antibiotic resistance genes accumulated due to horizontal gene transfers (Ding et al., 2019a). Toxic effects of pig manure were significantly reduced in soils with a higher organic matter content (Segat et al., 2015).

(ii) Storage: Post defecation, manure is exposed to environmental conditions such as temperature, moisture, exposure to air and ultraviolet radiation, starting a natural biodegradation process altering bacterial community composition (Oliver et al., 2010) and organic carbon content (Moral et al., 2005). The storage of manure by composting or ageing impacts emissions and nutrients and its effect on soil biodiversity. With an increasing storage time, pathogens in poultry manure decrease, whereby reduced water content decreases microbial mobility (Brooks et al., 2009). Without manure storage, the antibiotic monensin levels exceeded thresholds for triggering effects on soil organisms (Žižek et al., 2014). The study also found composting to accelerate monensin degradation compared to manure ageing. However, storing manure containing the antibiotic sulfonamide for 60 days did not prevent sulfonamide-resistance genes (Heuer et al., 2008). After 175 days of storage, the abundance of resistance genes decreased and thus was faster declining after application to soils. Hammesfahr et al. (2011) found sulfonamide antibiotic compounds in pig manure to significantly influence the N cycle due to alteration in soil microbial community structure. Storing contaminated manure in dark at 20 °C for 6 months increased the concentration of sulfonamide antibiotic compounds due to their adsorption to manure compounds. When added to soils, antibiotic compounds are remobilized upon microbial degradation of manure. In the same study, nitrifying bacteria were inhibited in stored manure, possibly due to antibiotic resistance formed during storage. Applied to soils, stored contaminated manure reduced the bacterial biomass and inhibited nitrifying bacteria affected by antibiotics, while fungal biomass in manure increased during the storage (Hammesfahr et al., 2011).

(iii) Treatment: Chemical, thermal or physical treatments are used to reduce contamination risks of fresh manures (Kumar et al., 2013). Those treatments change manure composition and, therefore, nutrient content, emissions and leaching potentials (Sommer and Hutchings, 2001) and soil organisms (García-González et al., 2016). Based on the literature reviewed, we list the impact of different manure treatments on the environment, focusing on soils and soil biodiversity in Table 2 (for more details on effects of different manure treatments on the environment see supplementary Table 2).

Table 2
The impact of manure treatment on the environment (with focus on soils) and biodiversity (for low to medium amounts of manure, not exceeding 25 t ha^{-1}): ++ large positive impact; + positive impact; − negative impact; −- large negative impact; 0 neutral (neither positive nor negative impact); + − No clear position in literature; NA refers to no available studies. Techniques separating manure into solid and liquid fraction allowing their separate management are not covered in the table since the impact on the environment and biodiversity depends on the fraction and the technique.

Impact	Environmental impacts with focus on soils (see supplementary text in Appendix A for more details)						Soil Biodiversity			
Manure Treatments	NH_3 Loss	Heavy metal soil pollution	Salinisation	Antibiotics	Pathogens	Soil organic Carbon content	Microbial biomass	Genetic diversity	Soil fauna	Plant-parasitic nematodes
Raw application (from animals farmed in stables, excluding untreated manure by grazing animals)	−	−	−	−	−	+	+−	+−	−	+
Aerobic composting of the solid fractions (Aerobic microorganisms decompose organic matter, occurring naturally when manure is stored in heaps)	−	+	++	+	+	++	+	++	++	+−
Biostimulant Fermentation (Naturally-occurring acidification e.g., compost teas)	**NA**	−	+	+	++	++	++	++	++	++
Anaerobic digestion (Microbial degradation of organic matter to biogas, as methane and carbon dioxide)	−	−	++	−	−	+	**0**	+−	−	+
Additives and other pre/ treatments (e.g., acidification through the addition of chemical compounds such as sulfuric acid)	+	**NA**	**NA**	**NA**	−	**NA**	**NA**	**NA**	**0**	++

While heavy metals cannot be removed by treatments, antibiotics (Zhang et al., 2014a), antibiotic-resistant genes (Sengeløv et al., 2003) and hormones (Kjær et al., 2007) can be reduced via certain treatment. Contaminants in slurry were found toxic to earthworms and enchytraeids (Segat et al., 2019; Segat et al., 2020) and collembolans in subtropical soils (Segat et al., 2020). Walsh et al. (2012) found slurry to reduce microbial growth and fungal diversity. High amounts of ammonia had negative effects on soil organisms as found for cattle slurry on collembolans (Pommeresche et al., 2017), earthworms (Van Vliet and De Goede, 2006) and arbuscular mycorrhizal fungi (Kahiluoto et al., 2000; Wentzel and Joergensen, 2016). Compared to solid manure, the distribution of bacterial species in slurry was more evenly dominated by certain species when urine was mixed with faeces (Cotta et al., 2003). Several studies found raw liquid manure beneficial for soil organisms (Bosch-Serra et al., 2020; Plaza et al., 2004; Ponge et al., 2013; Wentzel and Joergensen, 2016), e.g., by increasing bacterial biodiversity (van der Bom et al., 2018). The application of raw liquid manure can lead to toxic conditions for plant-parasitic nematodes due to high amounts of organic acids, high ammonia compounds and low C/N ratios (Thoden et al., 2011). Griffiths et al. (1998) found cattle slurry to shift nematode diversity from plant-parasitic to bacterivorous species. Neufeld et al. (2017) found higher microbial biomass and a higher activity of cellulose-degrading enzymes after applying solid manure compared to the application of liquid dairy slurry. Increasing the dose of raw and composted pig manure (from 0 to 100 t ha^{-1}) enhances the toxicity of raw manure on collembolans (Maccari et al., 2020). Earthworms were more abundant after applying composted manure compared to untreated solid manure (Hurisso et al., 2011).

Composted amendments provide organic matter for soil organisms, introduce beneficial microbes (Semple et al., 2001) and reduce amounts of antibiotic resistant genes (Tien et al., 2017). Earthworms are more abundant if composted manure is added rather than when slurry is applied (Rollett et al., 2020). Farmyard manure increases bacterial biodiversity, particularly those better adapted to nutrient-rich environments (Francioli et al., 2016). However, larger amounts of composted manure did not increase the population, likely due to salinity stress (Hurisso et al., 2011). When compost is fermented (immersion-extraction-oxygenation) in a liquid (mostly water) for a certain period (from few hours up to two weeks), compost teas are produced (De Corato, 2020b), enhancing fertilising effects of manure (Pane et al., 2014). Compost teas can be produced under aeration (e.g., by manually stirring) or under anaerobic conditions (Scheuerell and Mahaffee, 2002) and additives may be added such as minerals (e.g., rock dust) and nutrient additives including kelp extract and/or humic acids (De Corato, 2020a; Pane et al., 2014). Depending on the conditions, the fermentation process enhances the content of matured, stabilised organic matter (humic substances) and soluble mineral nutrients (Pant et al., 2012). However, it also multiplies compost microbiota which effectively suppresses soil-borne plant diseases and pathogens (De Corato, 2020b). For example, a compost tea based on fermentation of amino acid fertiliser from rapeseed meal with compost from pig manure effectively controlled *Verticillium* wilt affecting cotton (Lang et al., 2012) and aerated compost tea with added nutrients (kelp extract, rock dust, and humic acid) contributed to control grey mold in *Geranium* (Scheuerell and Mahaffee, 2006). Pant et al. (2012) found the highest number of active microbial populations in compost teas based on aged vermicompost. The survival rate of grown strains depends on the quality of the inoculant (compost feedstocks, compost age, water ratio, fermentation time, and added nutrients), the soil environment (pH) and climatic factors (temperature). Consequently, the results of fermentations are variable and unpredictable (Scheuerell and Mahaffee, 2002). However, preparations based on local inputs that sustain indigenous soil biota are better adapted to the local environmental conditions. They are more likely to thrive under agricultural practices than the commercial inoculum sources frequently used in biostimulant products (Valliere et al., 2020). Further research is needed to improve the application and usability of compost teas (De Corato, 2020b). Due to its properties and because compost teas enhance plant growth (De Corato, 2020a; Pane et al., 2014), compost teas follow the definition of biostimulants suggested by du Jardin (2015).

For biogas production, organic material in manure is anaerobically digested by anaerobic microorganisms decomposing carbon in the bioreactor. Digestates are the "waste" product of the process, whereby effects to soil organisms differ. Compared to no fertiliser application and undigested slurry, anaerobic digestates stimulate bacterial growth while no effect on fungi was found (Walsh et al., 2012). In another study, Barduca et al. (2020) found anaerobic digestates to increase fungal activity while not changing microbial biomass. Also, no harmful effects have been observed on soil organisms after nine days (Johansen et al., 2013) or after one month of its application (Podmirseg et al., 2019). However, Westphal et al. (2016) found only a minimal impact of anaerobic digestion on nematode, bacterial and fungi communities in soil one month after application of digestates. Another six-week experiment comparing the fertilisation effect of anaerobic digestates of food waste with mineral N-fertiliser, compost and farmed animal manures showed that the high amount of NH_4^+ resulting from anaerobic digestion harmed earthworms (Rollett et al., 2020). Digestates are less decomposable than undigested manure (Cavalli et al., 2017) since more carbon sources and nutrients for soil organisms are limited after digestion (Möller, 2015). Other studies found negative effects on fungi (estimated as reduced concentrations of ergosterol (Walsh et al., 2012; Wentzel and Joergensen, 2016)) and a decreased carbon content compared to conventional slurry, which may affect soil biota (Ernst et al., 2008). Mukhuba et al. (2018) found anaerobic digestion to reduce the diversity of culturable bacteria. Anaerobic digestion does not reduce heavy metals, so that digestates often contain high amounts of heavy metals (Nkoa, 2014), which may harm bacterial diversity (Gans et al., 2005) and significantly decrease soil biodiversity (Anjum et al., 2017). Similarly, a study on Collembola found a significant drop in the density of collembolans three days after the application of anaerobically digested cattle slurry, which may be caused by high NH_4^+ contents. Seven weeks from the application, numbers did only slightly recover (Pommeresche et al., 2017). Compared to thermophilic digestates, leading to more rapid digestion under higher temperatures (around 55 °C), digestates of the slower mesophilic digestation at around 37 °C contained more microorganisms (pathogenic and beneficial; Qi et al., 2018).

Acidifying swine slurry increases the concentration of volatile fatty acids at low pH, limiting microbial metabolism and, hence, pathogen activity and effectively controlling plant-parasitic nematodes (Mahran et al., 2009). A recent study found a little overall impact of acidified slurry on collembolans, although a larger amount of slurry may create favourable habitats for these organisms (D'Annibale et al., 2019).

3.1.2. Management II: spreading phase

During the spreading phase of manure, decisions regarding the application techniques, the quantity of manure and environmental interactions (e.g., physiochemical properties and climate) may also impact soil biodiversity (Bünemann et al., 2006; Unc and Goss, 2004).

(i) Application: When manure is superficially applied on broad surfaces, e.g., with broadcasting methods allowing uniform manure application such as through box spreaders, tank wagons, tow hoses and irrigation systems (Eurostat, 2021c), the microorganisms contained are more likely exposed to UV light and desiccation, reducing their concentrations (Forslund et al., 2011). In the case of superficial manure application, the likelihood of run-off is higher (Lamba et al., 2013; Sistani et al., 2010), increasing the release and transport of pathogenic microorganisms. Subsurface application such as injection prevents run-off (Lamba et al., 2013) and increases the survival chances of microorganisms and the risk of groundwater contamination, especially when manure contains pathogens and is untreated (Forslund et al., 2011; Semenov et al., 2009). De Goede et al. (2003) found that the surface application of slurry enhanced soil respiration, while slit injection negatively impacted epigeic earthworms closer to the soil surface. However, the densities of deeper dwelling anecic and endogeic earthworms were equal or higher after slit injection compared to surface application on 12 different farms. The same authors estimated that slit injection damaged 15% of epigeic earthworms at depths 0–5 cm and 30% of epigeic earthworms at depths 0–10 cm. Van Vliet and De Goede (2006), comparing broadcaster and slit injected manure, found no impact on the abundance of Enchytraeidae and nematodes. However, one week after broadcaster manure application, nematodes in the stage of dauerlarvae disappeared. Slit cutting in the presence of manure had no impact on earthworm abundance, but when combined with manure, broadcasting increased the abundance of earthworms. However, in the absence of manure, slit cutting declined the abundance of earthworms by 30% one week after the application. Broadcasting affected particular juveniles possibly due to salts present in slurry.

Effects of manure application on the abundance of earthworms depend on soil moisture and season. Wet conditions negatively impacted epigeic earthworms (De Goede et al., 2003; Van Vliet and De Goede, 2006), most likely since they are attracted to the soil surface and thus easier damaged by the machines. Slurry applied together with chalk and clay minerals to 18 different fields resulted in a higher abundance of Collembola and Acari in broadcasted compared to slit injected fields. Microarthropods responded in particular to the slurry application method rather than slurry manure type (Jagers op Akkerhuis et al., 2008). Slurry is frequently applied using tractors, which poses a high risk of subsoil compaction (Thorsøe et al., 2019), negatively impacting soil biodiversity, the biomass of soil organisms and enzyme activity (Nawaz et al., 2013).

(ii) Quantity: The continuous application of manure increases microbial biomass, soil respiration, nutrients such as potassium, phosphorus, magnesium (Segat et al., 2019) and mineral and organic N pools in soils (Pang and Letey, 2000; Yu et al., 2021). When plant nutrient requirements are exceeded, manure can contribute to the soil accumulation of soluble salts, heavy metals, P and N (Huygens et al., 2019). Ammonium was found to be among the main toxicity factors in organic materials, reducing the survival and reproduction of invertebrates (Renaud et al., 2017) and collembolans (Maccari et al., 2020). When pig slurry is applied to high N demanding crops following their maximum demands (2 t dry matter ha^{-1}), soil biota is likely harmed (Domene et al., 2008).

High amounts of applied manure also increase the content of pathogenic bacteria concentrations (Hruby et al., 2016) and heavy metals such as cadmium (Belon et al., 2012; Carne et al., 2020), zinc and copper (Segat et al., 2019). In France, zinc in manure contributed to 78% of overall zinc contamination in soils (Belon et al., 2012). Testing various doses of pig manure on earthworms (ranging 0 to 300 t ha^{-1}) and enchytraeids (ranging 0 to 100 t ha^{-1}), Segat et al. (2020) found increasing doses to reduce juveniles of both groups significantly. Another study tested the effects of a single application of 20, 50 and 150 t ha^{-1} of pig manure on soil fauna, whereby the highest amount altered soil fauna composition significantly, increasing insect larvae, dipterans and mites while decreasing ants, collembolans, earthworms and enchytraeids (Segat et al., 2019).

The doses of unstabilised swine manure had a more significant impact on earthworms compared to the effects of varying diets and the use of antibiotic growth-promoting additives (Serafini et al., 2020). Also, for collembolans, toxicity increases with the quantity of pig manure, whereby doses of 50, 75 and 100 t ha^{-1} caused 100% mortality (Maccari et al., 2020). However, Porhajašová et al. (2018) found 25 or 50 t ha^{-1} to not impact the presence of beetles (Coleoptera). The applications of 600 kg N ha^{-1} of pig manure led to a higher abundance of nematodes, compared to 150 kg N ha^{-1}, possibly due to an increase in microbial biomass (Jiang et al., 2013).

(iii) Environmental interactions: Testing the application of swine manure in four different soils, Segat et al. (2015) found that the soil texture and organic matter alter the toxicity of swine manure for earthworms. Compared to total N and total carbon, Jiang et al. (2013) found pH to have the most considerable effect on nematodes and fungivores (the study does not specify considered species of fungivores). Bacterivores, plant parasites and microbial functional groups were primarily affected by total carbon. The pH and cation exchange capacity are the most critical parameters affecting the bioavailability of metals and thus their toxicity (Lock and Janssen, 2001). Extreme soil pH values, both acidic and alkaline soil conditions, could lead to pollution. Toxicity by high pH affected the reproduction of Collembola (Crouau et al., 1999) and pH above 9 caused a 100% mortality of the earthworm *Eisenia fetida* (Kaplan et al., 1980). However, the success rate of slurries against plant-parasitic nematodes depends on non-ionized organic acids which are predominantly found at low pH (Thoden et al., 2011), so that swine manure was more successful in reducing plant-parasitic nematodes in acidic soils compared to alkaline soils (Lazarovits et al., 2001). The mobility of microorganisms in soils applied via manure significantly increases with rain or irrigation (Brooks et al., 2009). Also, slurry applied to frozen (Rocard et al., 2018) or waterlogged soils (Weslien et al., 1998) is more susceptible to run-offs, increasing the risk of *Salmonella* spreading (Hruby et al., 2018). Both climate and soil properties impact the variation of soil organisms, for instance explaining respectively 17.3% and 24.7% in nematode community composition (Jiang et al., 2013).

3.1.3. Effects of soil biodiversity on manure management

Over the last years, edaphic conditions and environmental heterogeneity have been increasingly investigated for their role in determining the effects of manure additions on pollution, nutrient availability, and soil biodiversity. The abundance of native, resident soil microbes is crucial for pathogen suppression (Goberna et al., 2011; Pérez-Valera et al., 2019; Podmirseg et al., 2019). Soils with high numbers of native soil bacteria are more likely to outcompete allochthonous species (Pérez-Valera et al., 2019; van Elsas et al., 2012), preventing the dissemination of antibiotic resistance genes (Chen et al., 2017). Also, earthworms contribute to reduce the broad-spectrum antibiotic oxytetracycline in manure after its application to soil, possibly activating and introducing microbes that can degrade antibiotics (Ravindran and Mnkeni, 2017). Soil bacterial diversity reduces the colonisation ability of the most common animal faecal pathogens that cause infections, such as the toxin-producing *E. coli* bacteria (Jiang et al., 2002; Xing et al., 2020). Pathogenic suppression is much lower in perturbed soils, where their indigenous microflora has been severely reduced (Moynihan et al., 2013). Those effects were confirmed in sterilised soils (Xing et al., 2020). In undisturbed agricultural systems (e.g., no-tillage), local bacteria in the upper soil layer form a barrier against antibiotic resistance bacteria (Pérez-Valera et al., 2019), whereas tilled soils increase the risk of significantly decreasing earthworm populations, as reported by Briones and Schmidt (2017).

Soil organisms play a crucial role in detoxifying soils, such as bacteria using metals in their metabolism for their growth (Ahemad, 2019); the large biomass of fungi reducing them to fewer toxic forms through chemical oxidation or reduction processes (Pinedo-Rivilla et al., 2009; Siddiquee et al., 2015); or earthworms immobilising them by adsorbing them to their cell walls (Ayangbenro and Babalola, 2017), which may be boosted by vermicomposting (Swati and Hait, 2017). The diversity of soil microorganisms are crucial to remove or detoxify contaminants (Garbisu et al., 2017). When soils' biodiversity was reduced through fumigation practices, soils were less resilient to persistent stress by copper contamination (Griffiths et al., 2000). Increased soil fauna diversity enhances plant nutrient uptake and reduces NH_3 losses (Geisseler et al., 2010; Singh et al., 2016; Van Der Heijden et al., 2008). Earthworms crucially improve soil structure and stabilise soil organic matter, thus enhancing the retention of carbon and N (Jouquet et al., 2011; Lago et al., 2019). Moreover, through their interactions with microorganisms, earthworms reduce ammonia volatilisation and increase soil N and P availability (Maldonado et al., 2019). This has led Lago et al. (2019) to conclude that more diverse food webs are critical for N retention in agricultural soils. Without this wide variety of biological activities, fewer nutrients are retained within the soil matrix and end up leached into groundwaters, as it has been found when soils are sterilised (Carswell et al., 2018) or when earthworms are excluded (Butenschoen et al., 2009). Also, for the immobilization of heavy metals, indigenous microorganisms play a crucial role as potential bioremediators (Li et al., 2020).

Soil biodiversity increases carbon stabilisation by stimulating the activity of microbial communities (Dignac et al., 2017), which enhances the mineralisation of organic matter in the short term and the sequestration of carbon in organo-mineral complexes in the long term (Bradford et al., 2013; Six et al., 2006). Earthworms' fast incorporation of organic matter into cast aggregates prevents decomposition (Vidal et al., 2016). Changes in community composition may have large impacts on carbon dynamics (Nielsen et al., 2011), e.g., the losses of nematodes declined carbon cycling significantly (Barrett et al., 2008). The loss of microbial diversity decreases carbon cycling, for instance, by reducing decomposition and carbon respiration (De Graaff et al., 2015). In a laboratory approach, a decreasing microbial diversity highly affected carbon cycling, whereby the significance of the diversity effect increased with nutrient availability (Maron et al., 2018).

Finally, soil biology plays a crucial role in preventing environmental threats related to manure application (see Table 3). To enhance the safety of soil organic amendments, Renaud et al. (2017) proposed to combine chemical and ecotoxicological data for an assessment of the toxicity of amendments. The study recommends to consider varying sensitivities and contaminant thresholds for various organisms but also to consider diverse routes of exposure during the assessment of toxicity

Table 3
The impact of soil biodiversity on the environmental threats caused by manure application: ++ large positive impact; + positive impact; − negative impact; –- large negative impact; 0 neutral (neither positive nor negative impact); + − No clear position in literature; NA refers to not available studies. Techniques separating manure into solid and liquid fraction allowing their separate management is not covered in the table since the impact on the environment and biodiversity depends on the fraction and the technique.

	Environmental Threats (relevant to soils)					
Impact of biodiversity	Emissions	NH_3 Leaching	Heavy Metal soil contamination	Pathogens (*Salmonella*)	Antibiotic resistance genes	Carbon losses
Microbial biomass	+−	++	++	++	++	+−
Genetic diversity	+	+	+	++	++	++
Soil fauna	+	+	+	++	++	++

of organic amendments, including manure.

3.2. Best practices for integrating soil biodiversity in managing manure

3.2.1. The role of biostimulants, biodynamic farming and agroecological practices

Soil fauna and microbial metabolism can be significantly accelerated by using fewer pesticides and mineral fertilisers (Birkhofer et al., 2008; Hartmann et al., 2015; Yang et al., 2019b), avoiding tillage (Briones and Schmidt, 2017) or by providing high-quality sources of nutrients to soil organisms (organic fertilisers/mulching; Carrillo et al., 2011). Also, adding external organisms may enhance the local genetic diversity (Brussaard et al., 2007; du Jardin, 2015; Sangiorgio et al., 2020). In organic farming systems (Bengtsson et al., 2005; Bünemann et al., 2006), manure plays a crucial role as a direct source of nutrients for soil organisms. It also adds genetic biodiversity and beneficial microorganisms to soils (Marschner et al., 2003; Zhen et al., 2014). The biodiversity in manure fertilised fields exceeds the number of mineral fertilised soils by 50% (excluding non-predatory insects and pests) (Bengtsson et al., 2005). The mean number of earthworm species was 126% higher and the number of individuals 156% higher in a diverse organic system than in a monocrop conventional field, applying mineral fertilisers (Feledyn-Szewczyk et al., 2019).

In organic viticulture, soil microorganisms were three to four-fold higher than in conventional viticulture (Karimi et al., 2020) mostly due to more available organic carbon applied by farmyard manure for soil organisms (Okur et al., 2016) - despite the usage of copper-based fungicides and bactericides, applied against downy-mildew. These treatments are currently still allowed in organic farming due to the lack of alternatives. While extremely high copper concentrations were found to negatively affect soil biodiversity, allowed amounts (< 4 kg per ha) are considered to not harm soil biodiversity (Karimi et al., 2021). However, other studies found that even low copper contamination can alter the spatial distribution of soil bacteria and fungi (Mackie et al., 2013) and cause severe damages on yeasts (Grangeteau et al., 2017). More extensive comparative studies and research for alternatives are needed (Karimi et al., 2020).

In ancient traditional knowledge systems in India (Biswas, 2020; Randhawa and Kullar, 2011), Europe (Bogaard et al., 2013; Krausmann, 2004) and South America (Birk et al., 2011), manure has been used as a main organic fertiliser or even as an ingredient for biostimulants. Traditional methods emphasise (i) quality of manure (e.g., source, including feed of the animal whose manure is used), (ii) method used for its treatment, including proportions of various ingredients used for such treatment and (iii) its management, i.e., the duration for which it can be stored, the amount to be used, and time/frequency of application. Such practices are applied, for example, in Natural Farming practices in India. Natural Farming refers to agro-ecological farming practices based on Indian traditional knowledge and combines organic farming practices with farm-made biostiumulant preparations (Das et al., 2020).

Most formulations used in Natural Farming have (cow) manure (particularly manure from indigenous breeds) as a critical ingredient. These formulations transform manure via fermentation into a potent biofertiliser that significantly enhances soil biodiversity. Apart from cow manure, farmer-made biostimulants are based on local, site-specific inputs such as sugar (e.g., ripe fruits), proteins (e.g., pea flour) and local soil. The application of these formulations improves the soil physical, chemical, and biological properties (Das et al., 2020; Devarinti, 2016; Liao et al., 2019), which are crucial to extract phosphorous from organic wastes (Szogi et al., 2015). Natural Farming biofertilisers have been shown to increase the availability of nutrients for sunflowers while decreasing the contaminants' concentration, such as chloride and sulfate (Iftikhar et al., 2019).

In the EU, organic production, that includes also the use of Biodynamic preparations ((EC) No 834/2007), allows the use of manures and composts complemented with compost teas based on manure (Mäder et al., 2002). The effectiveness of biodynamic preparations developed into a controversial scientific debate (Faust et al., 2017). Some studies find these preparations to enhance plants' self-regulating processes (Morau et al., 2020) and increase soil organisms' activity (Giannattasio et al., 2013; Spaccini et al., 2012). Zaller and Köpke (2004) found the decomposition significantly enhanced after 100 days in plots that received biodynamic preparations based on manure mixed with various flowers, compared to other farmyard manure treatments. By contrast, others find no difference in the effects between composted cattle manure and biodynamic preparations (Faust et al., 2017; Hendgen et al., 2018; Krauss et al., 2020; Sradnick et al., 2013). However, these studies often focus on microbial biomass (Faust et al., 2017) or considered the microbial functional diversity only (Hendgen et al., 2018; Sradnick et al., 2013). Mäder et al. (2002) considered the overall soil biodiversity, stating a higher diversity in biodynamic farming. Nevertheless, the trials conducted on organic and biodynamic practices sourced manure from different sources, limiting the significance of the results (Faust et al., 2017).

Both Natural Farming in India and some organic farming practices in the EU combine the addition of microorganisms gained from manure with more sustainable agricultural practices that link ecology with food production, the so-called agroecological practices (Chavarria et al., 2018; Migliorini and Wezel, 2017; Thiele-Bruhn et al., 2012). Agroecological practices are recommended due to enhanced resilience to ecologic pressure, such as pest attacks and weather extremes (Altieri et al., 2015) and economic and social pressure on farmers (Van der Ploeg et al., 2019). The same study provides empirical evidence for the economic potential of agroecological practices in the EU by simultanously sustaining employment levels and increasing incomes. Since agroecological farming systems operate on a regional basis, direct distribution, marketing and fairer prices increase consumer trust (Dumont et al., 2016) as well as (small) farmer profits. Parallel to agroecological movements (bottom-up movements, e.g., in Belgium; Stassart et al., 2018), there is increasing political support for these practices and institutional dynamics are driving the process: France has integrated agroecological practices at the national environmental policy level (Ajates Gonzalez et al., 2018; Moudrý et al., 2018), and the EU mentions explicitly agroecological farming practices in its Biodiversity Strategy for 2030 as a possible solution to maintain productivity while increasing soil fertility and biodiversity.

3.2.2. Recommendations for sustainable manure management

To prevent environmental threats relevant to soil biodiversity, we synthesised the optimal sequence of management steps that should be considered in manure management that consider the response on soil biodiversity (see the left side in Fig. 4).

Firstly, manure quality must be assessed and only applied without further treatment when:

(i) heavy metals that have been added to the animal feed do not exceed the given tresholds,

(ii) no antibiotics have been used in the previous week(s),

(iii) the local climatic/edaphic conditions are favourable (soils are not frozen or extremely wet after heavy rainfall events), and.

(iv) the quantities applied are not excessive.

However, if animals show any signs of illness or have been treated with antibiotics or hormones, manure must be treated before being applied to soils. Here it must be noted that also just storing manure without further treatment does not reduce antibiotic compounds in manure adequately (Hammesfahr et al., 2011). Depending on the type of pollutant agent, different treatment strategies need to be adopted.

Manure as fertiliser should only be anaerobically digested if no antibiotics were added to the diet of farmed animals, othewise antibiotic-resistant genes may remain in the digestates (Tien et al., 2017). Keeping ill animals separately from healthy animals prevents the mass-usage of antibiotics, while permitting the separate storage of contaminated and uncontaminated manure. This, in turn, permits the selective usage of

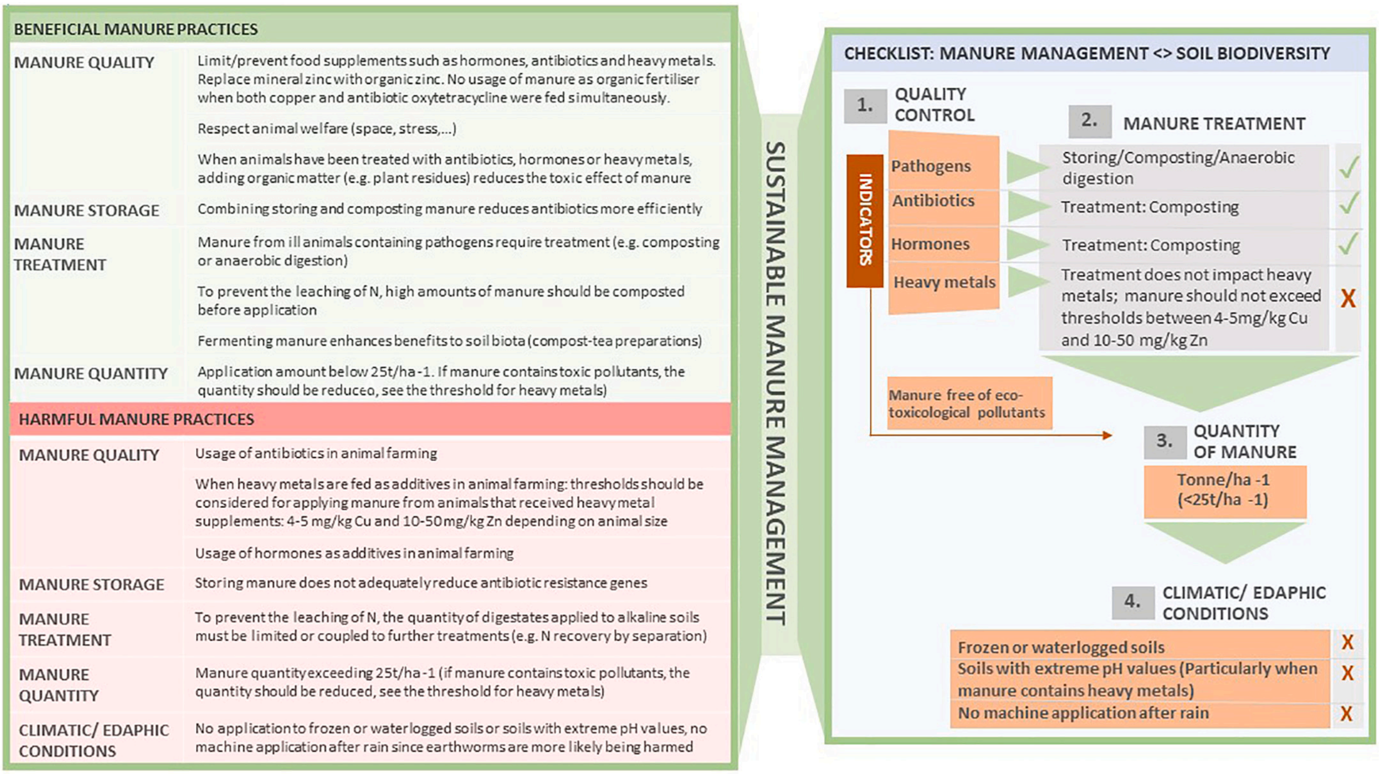

Fig. 4. Beneficial and harmful manure practices for soil biodiversity (on the left) result in sustainable manure management if the checklist provided on the right is followed.

manure from healthy animals as an organic fertiliser. Also, to reduce the toxic effects of pig manure in soils, organic matter should be added (e.g., straw or other plant residues; Segat et al., 2015).

Manures from animals treated with heavy metals (copper/zinc) should not exceed the permited limits of copper in organic farming (<4 kg per ha). Ding et al. (2021) determined dosages of copper and zinc as thresholds that should not be exceeded: 5 mg/kg Cu and 50 mg/kg Zn for animals in the range of 9–15 kg; 4 mg/kg Cu and 50 mg/kg Zn for animals between 15 and 25 kg and 4 mg/kg Cu and 10 mg/kg Zn for animals in the range of 25–60 kg. Since heavy metals cannot be removed from manure by any currently known treatment methods, the amount of copper needs to be calculated based on dietary additions and threshold limits. Also, mineral zinc may be replaced by organic zinc – resulting in improved faecal scores (Bouwhuis et al., 2017). The incidence of diarrhea of piglets can be reduced by decreasing stress among animals (e.g., by reducing the animal counts, more space and access to the outdoor; Martínez-Miró et al., 2016).

Careful attention should also be paid to the amounts of manure added since several studies have highlighted the negative correlation between manure quantities and soil biodiversity (Maccari et al., 2020; Segat et al., 2019), which, in turn, is closely associated with the increased concentration of pollutants in manure. Accordingly, when the type and concentrations of these pollutants are unknown, applied manure should not exceed 25 t ha^{-1} (Segat et al., 2019). Applying less amounts of manure might reduce other benefits of this organic fertiliser on soil quality, such as C sequestration. However, recent studies have shown that the positive effects of manure application (in large quantities) on soil carbon storage are strongly dependent on agricultural management (e.g., tillage, additional mineral fertilisation) and climatic and soil abiotic condtions (e.g., soil pH and initial C content) (Gross and Glaser, 2021). Indeed, climatic and edaphic conditions also need to be considered before manure application. Particularly, clay-rich alkaline soils are prone to metal accumulation, while acidic soils pose additional environmental risks because enhanced metal mobility can lead to groundwater pollution. Similarly, as there is a high amount of ammonia in the digestates, manure should not be applied to alkaline soils without adequate pre-treatment to prevent ammonium (NH_4) emissions.

In the context of manure application methods, the separation of the solid and liquid segments of manure through osmosis and scrabbing permits better recovery of N from post-processing (Finzi et al., 2020). Manure treatment practices must be included in subsidy systems to ensure the economic feasibility of the practices. Manure should not be applied by means of slit or broadcasting methods after raining events to prevent harmful effects on earthworms (De Goede et al., 2003; Van Vliet and De Goede, 2006).

In summary, to avoid ecotoxicological effects, thresholds must be set that consider both the quantity of manure and the local climatic and edaphic conditions. These thresholds should be based on quantitative assessments, including the responses of various soil organisms (ranging from micro to meso- and macro-fauna) to different manure management practices under different soil types, agricultural practices and climatic conditions. Manure management recommendations should meet sustainable farming methods (see 3.2.1) to fully protect and promote soil biodiversity and to achieve sustainable manure management (Fig. 4). Only sustainable manure management can counteract any potential environmental risks posed by farmyard manure additions.

Despite the potential of these recommendations to achieve more sustainable farming systems, in order to be fully adopted and implemented by farmers, we would need a radical transformation in the way agriculture is conducted. Coupling better manure quality and increasing soil taxonomical and functional diversity could provide a self-regulating system that maintains current production levels (Thiele-Bruhn et al., 2012; Lavelle et al., 2016).

3.3. The relevance of soil biodiversity in European legislation on manure management

The main instrument that drives agricultural production in the EU is the Common Agricultural Policy (CAP), combining economic incentives with regulatory requirements (Paleari, 2017). For example, the Nitrates Directive is a Statutory Management Requirement, which is part of the conditionality of the CAP. This needs to be fulfilled by farmers to receive subsidies. Within the conditionality of the CAP, Member States (MS) must decide certain Good Agriculture Environmental Conditions (GAEC), which then are mandatory for farmers. The post-2020 CAP introduces in GAEC 5, the Farm Sustainability Tool (FaST). Based on information of selected farms, the type of crops, the number of animals and the amount of manure, the tool provides data for nutrient management, including "data on soil, the proximity of protected areas and legal limits on the use of nutrients" (European Commission, 2019). The tool aims to facilitate nutrient management and adapt it to local site conditions (European Commission, 2019). Apart from its conditionality, the CAP provides mostly voluntary options (such as eco-schemes and agri-environment-climate payments) to protect and enhance soil biodiversity.

Various other regulations (e.g., Regulation 1069/2009 for the transport, trade and disposal of manure), apply directly to all MS while directives are implemented at the MS-level into national legislation, e.g., the Nitrates Directive, the Air Quality and National Emission Ceilings Directive and the Industrial Emissions Directive. For the implementation at the MS-level, MS decide on best practices for manure management and application based on the agronomic context (European Commission, 2014), for example, including nutrient balances (European Commission, 2000) and preferences for manure treatment (Hou et al., 2018). In legislations targeting manure, there is no generally applicable definition of manure quality. Different legislations address various aspects related to manure (e.g., air emissions, nutrient losses to water, manure application and management), but there is no overarching piece of legislation that bundles all different aspects related to manure management. An analysis of relevant legislation found in total eight policies (Table 4), impacting

Table 4
Manure management in EU legislation, including Directives and Regulations.

Legislation	Scope of the legislation in terms of manure management
Animal By-Products and Derived Products Regulation 1069/2009	Allows the direct soil application of unprocessed manure (that is not placed on the market) to soil, notwithstanding the quality of manure (type, storage, treatment). Manure application must follow the precautionary principle, considering the particular environmental condition. The regulation also sets rules for the disposal, collection and transport of manure, allowing the transfer of manure from one farm to another within the same MS without a health certificate tracking the manure's origin or quality. Sterilisation and production in a certified plant are needed to put fertilisers based on manure on the market.
The Common Agricultural Policy (CAP) Regulation (EU) No 1306/2013	The majority of direct payments of the CAP encourages quantity over quality, favouring vast amounts of manure which on the other hand might lead to "inappropriate disposal practices", including the application of raw manure or its application not being well-timed (Buckwell and Nadeu, 2016; Lazcano et al., 2008). Also, the CAP offers Member States the option of a voluntary coupled support for animal-based food (dairy products, beef, veal, sheepmeal and goat meat (European Commission, 2021a) aiming to prevent the import of animal products that were produced under sub-optimal social and environmental conditions. The rural development fund of Pillar II allows MS to spend up to 30% of their budget in activities, such as the support of manure processing or storage, including anaerobic digesters (European Commission, 2017). Also, subsidies for organic farming are part of Pillar II (see below). With the CAP 2021–2027, MS can decide upon eco-schemes. The list of recommended eco-schemes includes "other practices related to GHG emissions" including improved manure management and storage (European Commission, 2021b). Within the conditionality of pillar I, the Good Agricultural Environmental Conditions (GAEC), introduces GAEC 5, the Farm Sustainability Tool (FaST) (European Commission, 2018). Nutrient management requires data "such as data on soil, the proximity of protected areas and legal limits on the use of nutrients" (European Commission, 2019). FaST takes into account soil information obtained using on-site tests (soil and manure samples) and an inventory of all nutrient sources (e.g., crop residues), application techniques and handling and storage of manure (PwC, 2019). When MS decide for GAEC 5, the nutrient management with the FaST tool is compulsory for any agricultural land, not only for Nitrates Vulnerable Zones (European Commission, 2018), aiming to make nutrient management more adapted to local site conditions (EASAC, 2018) and to reduce "nutrient leakage in groundwater and rivers, as well as positively contributing to soil quality and reducing greenhouse gas emissions" (European Commission, 2019).
Air Quality and National Emission Ceilings Directive 2016/2284	MS may establish good agricultural practices by including manure to improve the nutrient status and soil structure, addressing animal husbandry practice, livestock housing, manure storage and spreading techniques, and how to improve nutrient management, for example, by promoting (a) investments for low emission animal housing systems, (b) investments for low emission manure storage systems, (c) feed additives to reduce methane emissions, (d) investment for low-emission manure spreading techniques and (e) investment for on-farm bio-digesters.
EU Nitrates Directive 91/676/EEC	The most critical EU regulation for managing manure and its impact on the environment (Van Grinsven et al., 2012). The Directive is Statutory Management Requirement 1 of the conditionality of the CAP – a prerequisite for direct payments that are either area- or animal-based (EU Commission). The Directive aims to protect water quality by designating nutrient vulnerable zones, requiring MS to propose Good Agricultural Practices in high-risk zones to prevent eutrophication measures, i.e., by considering amounts, storage, time of the year and local weather for manure application (Musacchio et al., 2020; Serebrennikov et al., 2020). The limits of applicable N in high-risk zones is max. 170 kg/ha of N and in certain zones of France, manure needs to be treated before its application to soils (Loyon, 2017). Some MS provide farmers with exemptions if manure disposal does not harm ecosystems (Serebrennikov et al., 2020; Svanbäck et al., 2019). Time limits for manure application per hectare according to climatic and geological characteristics prohibits the application of manures in winter or on steep slopes (Buckwell and Nadeu, 2016; Louwagie et al., 2011). The implementation of the Directive varies greatly, for example, whether digestates from anaerobic digestions are included and thus regulated (European Commission, 2013).
Organic Production Schemes Council Regulation (EEC) No 2092/91	Prohibits using manure obtained from factory-farmed animals in organic agriculture, but the definition is left to MS or organic control agencies (Løes et al., 2017). The limited availability of certain essential nutrients such as P allows organic farmers to apply mineral P, manure from conventional agricultural systems, organic residues if composted or anaerobically digested and other animal residues, including bones and meat (Løes et al., 2017). Varying regulations and limits of permitted conventional manure complicate the comparisons within MS, making consumer education difficult. Antibiotics in Organic Farming can only be used if alternative treatments fail, they must be approved by a veterinarian, and no animal can receive more than three treatments per year (Duval et al., 2020), except for vaccinations, treatments against parasites and compulsory eradication schemes. If the animal's life cycle is less than one year, only one treatment is possible. Stricter regulations decreased the use of antibiotics significantly (Wagenaar et al., 2011). While the usage of copper-based fungicides was reapproved in 2018, the usage is limited to 4 kg $year^{-1}$ and incldudes copper on the list of candidates for substitution, which means once an alternative is found, copper will be banned (Droz et al., 2021; EU 2018/1981). The amount of permitted copper was reduced by a factor of ten compared to the 1980–1990 period (Karimi et al., 2020).

(continued on next page)

Table 4 (*continued*)

Legislation	Scope of the legislation in terms of manure management
Fertilising Products Regulation (EC) No 2003/2003 replaced by Regulation (EU) 2019/1009	It lays down rules for EU fertilising products to access markets. Since Regulation 2003/2003 only covers inorganic fertilisers, standards such as composting, standards and quality widely differ between MS (Bernal et al., 2017; Cesaro et al., 2015). This regulation will be replaced by the Fertilising Products Regulation 2019/1009 in July 2022 (European Commission, 2019), which will impose stricter limits for mineral fertilisers (for example cadmium content) and will allow fertilising products from recycled bio-wastes and other secondary raw materials (Huygens et al., 2019; Paungfoo-Lonhienne et al., 2019), aiming to reduce the pressure on biological resources through the recycling of biowastes (Bell et al., 2018; Paungfoo-Lonhienne et al., 2019). Waste Products, according to 2008/98/EG (including manure), will lose their status as waste as soon as they are transformed into other products (Directive 2008/98/EC). For example, its treatment (such as fermenting or composting) will convert manure into a fertilising product. Labelled as a Circular Economy product, the Regulation allows for the recycling of manure as a secondary waste product. This will increase the access to fertilising products based on manure. The update will introduce limits for pollutants (such as copper and zinc) in composts and digestates (the proposal does not regulate raw manure). For the anaerobic digestion of manure, temperature-time profiles for thermophilic digestion, glass/metal and plastic contents are regulated and limit heavy metals, polycyclic aromatic hydrocarbons and pathogens. According to the regulation, any given organic soil improver must have a dry matter content higher than 20% and the content of organic carbon must exceed 7.5% of the total dry mass (Rodrigues et al., 2020).
Veterinary Medicinal Directive 2001/82/EC replaced by Regulation EU/2019/6	Foresees the authorisation, testing, documentation and traceability of veterinary medicinal products, including, e.g., broad-spectrum anti-parasitics such as Ivermectin (Adler et al., 2016) but also hormones and antibiotics. The regulation EU/2019/6 will replace the Directive 2001/82/EC in January 2022 (European Medicine Agency, 2019). Among the main objectives of the updated regulation (EU/2019/6) antimicrobial resistance should be tackled through prudent usage. As a measure, for example, certain antimicrobials may be restricted for use in animals, prioritising them for human treatment. It requires MS to collect data on antimicrobial medicinal products used in animals.
Industrial Emissions Directive (2010/75/EU)	The Directive regulates manure management for larger pig and poultry farms. To obtain a permit, the farms must comply with a series of conditions related to animal housing and manure management (storage, processing techniques, application techniques) to reduce overall adverse environmental impacts.

manure management and consequently soil biodiversity (see Table 4; for an overview of existing legislation on manure management and soil biodiversity; see supplementary Table 3). Only the Organic Production Scheme Regulation (2092/91) currently combines manure management with farming practices that may enhance soil biodiversity.

Currently, there is no unified plan of N budgeting in the EU, and indicators for nutrient management (such as the calculation for N surpluses), water quality (Klages et al., 2020) and calculations for manure-N availability (Webb et al., 2011) greatly vary at farm level. For example, for the calculation of the nutrient use efficiency, only 13 MS (out of EU27 + UK) consider soil type, 14 MS consider crop type, 13 MS consider the time of application, 8 MS consider climate, 7 MS consider the method of application, 23 MS consider manure type and 7 MS ensure the long term availability of the indicator (Webb et al., 2011). Similarly, MS include digestates differently generated through anaerobic digestion. For example, Spain and Italy do not address digestates specifically, while Austria, Belgium, Germany, Denmark, France, Lithuania, the Netherlands and Poland handle digestates as fertiliser, soil amendment or soil improver. The limitation set by the Nitrates Directive of 170 kg/ha of N applicable to nitrate vulnerable zones includes digestates in Ireland, Belgium, Denmark, Estonia and the Netherlands (if the input material consists of at least 50% of manure) and Sweden (if input material consists of 100% of manure). Austria, Bulgaria, Germany, Finland, France, Italy, Latvia, and Slovenia omit digestates in their calculation; the remnant MS did not answer the survey by the Joint Research Centre (European Commission, 2013b). Some MS use very high-efficiency rates for digestates (Wallonia in Belgium calculated a 100% efficiency; (Webb et al., 2011)), which is not consistent with research results (Möller and Müller, 2012) and may increase leaching risks. Bloem et al. (2017) recommended not to calculate the amount of applicable digestates or manure based on its N content because of the low N:P ratios that favour P surpluses and possible runoff. Few MS set maximum values of applicable P such as Sweden (Bloem et al., 2017), Denmark (Wall et al., 2011), Belgium and the Netherlands (Amery and Schoumans, 2014).

Also, the importance attributed to soil biodiversity varies among MS. Although several countries collect data on soil organisms independently (Römbke et al., 2016), there is a lack of a standardised methodology at the EU level (van Leeuwen et al., 2017). To overcome some of these challenges, the European Commission has launched the EU Soil Observatory (Montanarella and Panagos, 2021). Also, the research activities of the Joint Research Centre (JRC) in the European Commission aims at standardising soil biodiversity monitoring at the EU level (Orgiazzi et al., 2018). A soil biodiversity module has been introduced in LUCAS topsoil survey and almost 1000 soil samples have been analyzed for soil biodiversity through metabarcoding and metagenomics (Orgiazzi et al., 2018). In particular, in 630 samples, including agricultural soils, metagenome was also sequenced. This permitted investigation into the impact of management practices on soil functional genes (e.g., antimicrobial resistance genes). While the proposal for a Soil Framework Directive, including threats to soil biodiversity, was rejected in 2007 due to the blocking of five Member States (MS), soil protection is currently regulated at the MS level following the principle of subsidiarity (Römbke et al., 2016). However, the protection of soil biodiversity has recently, gained greater prominence and was directly addressed in the Biodiversity Strategy for 2030, within the EU Green Deal (Montanarella and Panagos, 2021). Recently, the first global soil biodiversity assessment was presented by the Food and Agriculture Organisation (FAO, I, et al., 2020). The EU has expressed the intention to improve sustainable soil management that may promote, amongst other, soil biodiversity: the "Farm to Fork Strategy" aims to reduce pesticide usage by 50%, fertiliser application by 20% and nutrient losses by at least 50%, while the Biodiversity Strategy 2030 targets to increase high-diversity landscape features by at least 10% and organic farming from 7.2% in 2019 to 25% in 2030 (European Commission, 2020d).

3.4. Analysis of manure-relevant EU legislation for potential integration of soil biodiversity issues

Analysis of the legislation in place revealed the following limitations that complicate sustainable manure management in the EU:

- Currently, the concentration of manure production is too high to be absorbed by the local environment, leading to losses and unsustainable management. Apart from approaching the unsustainable sizes of animal farmed in the EU, prevailing manure practices such as the application of raw manure and the often subsidised anaerobic digestion must be further examined for their potential negative effects as soil amendments due to the quality of nitrogen sources, the risk of contamination with heavy metals and antibiotics and the potential nutrient leaching due to changed soil chemistry. Challenges posed by a declining quality and an increasing amount of manure are not adequately addressed in existing legislations, which also do not include the potential negative effects on soil biodiversity (see supplementary Table 3). Because policy frameworks regulating manure management are scattered across various legislations, the complexity of achieving sustainable soil management is increased (Thorsøe et al., 2019).

- Only Organic Production Schemes regulate manure quality in combination with agricultural practices that are beneficial for soil organisms. Other legislation does not consider soil biodiversity in their scope and consequently neither in their recommended practices. For example, regulations permit the immediate application of high amounts of slurry despite the lack of long-term studies on its effects on soil organisms (Podmirseg et al., 2019). Furthermore, even in organic farming, the application of conventional manure is permitted, potentially introducing hormones, heavy metals, microplastics and antibiotic gene resistance to organic soils, thereby compromising environmental goals and quality standards of Organic Production Schemes (Løes et al., 2017). The recycling of conventional manure in organic agriculture remains an issue that needs to be addressed in future policy developments. To address drug residues, upstream actions (drug administration to animals) may be most suitable, which will be introduced in the update of the Veterinary Medicinal Regulation EU/2019/6 coming into place in 2022.
- From 2022 onwards, the Fertilising Products Regulation will come into place aiming to facilitate cross-border transport of high-quality products. The Regulation will imply stricter quality controls for organic soil improvers (e.g., maximum amounts of copper (300 mg/kg dry matter) and zinc (800 mg/kg dry matter), minimum carbon content) featuring the potential of pre-treated manure as a safe, hygienic fertiliser product. However, thresholds have not been calculated based on soil biota taxonomic data and no similar thresholds for the application of raw slurry are available. This Regulation will facilitate access to markets for organic fertilisers based on manure, however, it will not overrule national legislation since the Regulation is based on the principle of optional harmonisation. While an additional material class is under development which group all animal by-products, currently, the regulation only considers composting and anaerobic digestion, both of which are time and resource-intensive (i.e., requiring a large surface area for logistics and high investments; (Alegbeleye and Sant'Ana, 2020). Likely, farms with small amounts of manure apply anaerobic digestion less frequently (Foged et al., 2011), but many MS subsidizes anaerobic digestion (Banja et al., 2019; European Commission, 2012).
- The EU prioritises inputs from animal farming for biogas production (European Commission, 2012) to limit indirect land-use changes and prevent the conversion of abandoned or set-aside land providing biomass for biogas production (Tamburini et al., 2020). For example, the Renewable Energy – Recast to 2030 (RED II) restricts feedstocks with "high indirect land-use change-risk" (Directive (EU) 2018/2001). However, research on anaerobic digestion does not consider the long term effects on local soil conditions and often ignores the potential risks of pollutants in digestates (Grando et al., 2017; Meyer and Edwards, 2014; Podmirseg et al., 2019).
- The Nitrates Directive is currently the most critical tool to mitigate nutrient leaching from manure. Despite its standards set to reduce N losses, full compliance has not yet been achieved (Buckwell and Nadeu, 2016). For example, a stakeholder network analysis in the Lombardy plain finds steady or increasing N surpluses calling for better governance and monitoring due to farmers' attitudes towards inadequate N management (Musacchio et al., 2020). In France, in the Seine basin, actions to reduce nitrates in water tables were not effective and diffuse agricultural pollution has not decreased since the 1990s. Since 2013, the number of water tables that have been downgraded due to exceeding nitrate thresholds has doubled (Bouleau et al., 2020). The European Commission urged France (European Commission, 2020c), Italy (European Commission, 2020a), Belgium and Spain (European Commission, 2020b) to comply with nitrate thresholds for losses to water tables set in the Nitrates Directive. If thresholds are not met, MS are brought in front of the Court of Justice of the European Union, as was the case for Germany (European Ombudsman, 2019). Legal processes led to stricter rules for applying fertilisers (Bouleau et al., 2020) and Germany implemented a new national fertiliser law in 2020 (Schulz, 2020).
- The Industrial Emissions Directive regulates manure management for larger pig and poultry farms, including nutritional management of animals, the collection, storage, treatment and land application of manure (Directive 2010/75/EU), aiming to reduce overall adverse environmental impacts. It does not set limits for heavy metals, which pose a particular threat to soil biodiversity and the functions provided.
- While certain flexibility at the MS level is crucial to consider spatial characteristics, certain standards should be applied EU-wide. Currently, climate and soil conditions are not yet adequately considered for manure management at the MS level. For example, only a few MS consider the spatial soil properties and long term effects in calculating manure-N availability (Webb et al., 2011). Estimates of nitrogen budgets provided by MS do not consider nutrient accumulation and leaching to harm soil biodiversity (Klages et al., 2020). Also, the effects of pre-treatment on nutrient availability are ignored (Bloem et al., 2017) and the recovery of nutrients besides N (e.g., in GAEC conditionality) is not yet included in current EU regulations (Garske et al., 2020).
- Amendments adopted by the European Parliament on 24 October 2017 on the proposal for a regulation laying down rules on the making available on the market of CE marked fertilising products ((EC) No 1069/2009 and (EC) No 1107/2009 (COM(2016)0157 –C8-0123/2016–2016/0084(COD)), recommended the inclusion of recital 5a: "(5a) To ensure effective use of animal manure and on-farm compost, farmers should use those products which follow the spirit of 'responsible agriculture', favoring local distribution channels, good agronomic and environmental practice and in compliance with union environmental law, … The preferential use of fertilisers produced on-site and in neighboring agricultural undertakings should be encouraged." Such a provision can yield benefits rather than further environmental losses, if farmers are adequately trained for on-farm treatment and application of manure based natural fertilizers and biostimulants. Unfortunately, no such farmer training curriculums have been designed and this proposal has been dropped by EU 2019/2009.

4. The way forward in future EU policies of manure management

At present, integrated manure management that addresses both the risks and benefits for soil biodiversity sits beyond the scope of legislation, despite increasing evidence drawing attention to the link between both components (as highlighted in this review). Since manure management is a balancing act on the crossroad of conflicting interests, socially acceptable solutions (Burton, 2007) require the commitment from the scientific community, policymakers, farmers and other stakeholders involved (Gebrezgabher et al., 2014).

The scientific community emphasises the importance of considering the environmental impacts of the entire manure management process because integrated manure management may reduce environmental hazards by 65% while doubling nutrient efficiency (from 33 to 70%; De Vries et al., 2015). However, besides addressing climatic emissions, holistic manure management also needs to include the effects on belowground biodiversity. This review has shown that solutions combining manure management and soil biodiversity enhancement simultaneously could lower environmental risks. For example, applying raw manure with biofertiliser or biostimulants may enhance plants' nutrient uptake and decreases the contamination risks derived from the nutrients surplus (Paungfoo-Lonhienne et al., 2019; Schoebitz and Vidal, 2016; Thonar et al., 2017). However, recommendations often focus on emissions only. For example, the recommended manure incorporation practices to reduce NH_3 emissions (Amann et al., 2018; UNECE, 2015) do not consider the harmful effects on soil organisms. This shortcoming could be overcome by coupling manure injection with climatic conditions (prevent injection after raining events) to protect soil-surface dwelling organisms.

Furthermore, since manure quality greatly diminishes with increasing numbers of farmed animals (due to usage of antibiotics, heavy metals), reducing the animal stocks is a key factor for reducing undesired environmental impacts, including those on soil biodiversity. While certain consequences of heavy animal farming can be filtered by better treatment methods (e.g., pathogens), heavy metals cannot be filtered by treatment methods and remaining antibiotic-resistant genes have been found in digestates (Tien et al., 2017). Therefore, priorities should focus on quality rather than quantity. This goes hand in hand with reducing the consumption of meat and dairy products with more environmental-friendly and nutritious plant-based diets, which simultaneously contribute to food security (De Boer and Aiking, 2011). To prevent the replacement of farmed animals in the EU with cheaply imported meat and dairy products, education and awareness campaigns need to support innovation of food culture.

In addition to reducing animal density, better quality manures can be obtained through a better monitoring of unprocessed manure, as well as better manure application practices – comparable to those existing for food composition, fertiliser and biostimulants formulations. A proposal would be to better inform farmers by adding relevant information on packaging (labelling), including the nutrient content, the source (type of animal, place of origin, year), the quality as well as management of manure that will be placed on the market using the Fertilising Product Regulation. Standardising manure from highest to lowest quality based on source, treatment method and expected impact on beneficial soil organisms would support more mindful manure application by farmers – reducing negative impacts of low-quality manure and maximising positive impacts of high-quality manure. While organic fertilisers currently still occupy a niche market (horticulture, floriculture, gardens), with an increasing share of organic farms (EU goal of 25% until 2030), the market is likely to grow. The grading and labelling of manure (ingredients) can help to better monitor the effects of short- and long-term application of specific types of manure in various agricultural conditions (soil, crop, climate). Such standardised procedures will allow tracking the relocation of manure and its nutrients (Bernal et al., 2017; Musacchio et al., 2020), enabling redistribution of nutrients from surplus areas to arable areas (Hanserud et al., 2017), as applied in the Netherlands (Van der Straeten et al., 2011). Also, a transparent, standardised manure quality will facilitate the transport to nutrient-deficient regions (Luostarinen et al., 2018). However, control and monitoring mechanisms require extensive financial resources. As a trade-off, local, traceable manure chains must be created. In Austria, for example, a standardisation for the manufacturing and labelling applies for compost, classifying compost depending on input materials, including heavy metals (RIS Austria, 2001).

Soil biodiversity needs to be appropriately included when budgeting manure amendments to provide better guidelines on the use of manure and to reduce costs and environmental risks. Recommended values and thresholds of manure amendments must consider the state of soil biodiversity according to different land management practices. To move towards integrated manure practices, alternative manure treatment methods such as acidification of manure (Fangueiro et al., 2015; Ti et al., 2019), or by combining anaerobic digestion with solid–liquid separation and N removal (Finzi et al., 2020), need to be expanded while considering the long term effects of manure management on soil organisms (Bünemann et al., 2006). Further, farming systems based on traditional knowledge (TK) such as biodynamic farming or Indian Natural Farming that adopt a systems approach to managing and enhancing soil and seed biodiversity need to be studied more closely. Additionally, research must address potential threats in manure management (pathogens, antibiotic resistance genes, leaching of nutrients and heavy metals) as well as benefits (acting as biofertiliser, enhancing soil biodiversity, adding organic matter), while considering soil-specific characteristics and local status of soil biodiversity.

In parallel, standardised monitoring schemes of soil biodiversity (such as LUCAS) will help to better understand the effects of land-use practices, including manure practices, on soil biota. Monitoring soil biodiversity needs to incorporate taxonomic and functional soil biodiversity to obtain information on the effects of land-use practices on ecosystem services (Felipe-Lucia et al., 2020). Coupling a harmonised methodology to determine the quality, amounts and treatment of manure and manure-based fertiliser products with soil biodiversity data could be integrated into the Farm Sustainability Tool for Nutrients (FaST) or similar nutrient management tools for monitoring nutrient budgets. The FaST proposed in the framework of the CAP Good Agricultural and Environmental Conditions (GAECs), aims to facilitate sustainable use of fertilisers for all farmers in the EU while boosting the digitalisation. The mandatory usage of the FaST Nutrient Management Tool from 2021 onwards brings the opportunity to move beyond the amount and timing of manure field application by integrating manure quality, its origin and site characteristics (Buckwell and Nadeu, 2016). FaST can play a role in accounting for the amount of manure produced by farm and should also incorporate manure management in the nutrient budget. For example, recommended values and thresholds of manure amendments must consider the state of soil biodiversity according to different land management practices and provide better guidelines on the use of manure to reduce costs and environmental risks. The tool also offers a great potential to educate farmers in linking soil biodiversity and manure management and enhance soil biodiversity functions in the next CAP update. The FaST is part of the post-2020 Common Agriculture Policy, which gives more flexibility and responsibility to MS in regulating manure management and soil biodiversity according to their national needs. However, it is unknown how MS will respond towards voluntary actions such as eco-schemes.

Data provided by the LUCAS would allow establishing thresholds and indicators to prevent threats posed by inappropriate manure practices. Indicators are crucial as quantitative targets in regulatory instruments, and could contribute to better manure treatment and nutrient management (Daxini et al., 2018; Savage and Ribaudo, 2013). Therefore, either the scope of current legislations need to be expanded, or a new legislation is required that considers the impact of manure on soil biodiversity. Both strategies require the same extent of the legal process and would allow implementing the direction set at a strategic European political level, where the importance of protecting soil biodiversity is increasing. At a strategic level, the EU is increasingly aware of the importance of protecting soil biodiversity: for example, the Biodiversity Strategy 2030 explicitly mentions the critical role of soil biodiversity. Moreover, the aims of both the Farm to Fork Strategy and Circular Economy Action Plan indirectly protect and promote soil biodiversity. For example, the Biodiversity Strategy 2030 explicitly mentions the critical role of soil biodiversity.

Since the Fertilising Product Regulation opens a market for manure and biostimulant products and will also more strictly regulate mineral fertilisers from 2022 onwards, the access and value of organic fertilisers are likely to increase. From an economic point of view, sustainable manure management may contribute to partially replace mineral fertilisers by increasing soil biodiversity and, consequently, nutrient efficiency. This replacement could decrease the dependencies on imports of mineral P (European Commission, 2018; Svanbäck et al., 2019) and N; therefore, it will request fewer fossil energy sources (European Commission, 2013c; Smil, 2004). A trade-off of replacing mineral fertilisers with manure will require wider knowledge-management as well as knowledge dissemination. For implementing manure management practices, farmers' awareness of soil biodiversity is crucial (Serebrennikov et al., 2020). A study in Spain, Italy, Denmark and the Netherlands found that legislations are the key factor contributing to manure treatment (Hou et al., 2018). However, too strict regulations or a lack of knowledge prevent the implementation of composting practices (Viaene et al., 2016). Knowledge of on-farm composting or other manure treatment methods needs to be spread, e.g., through training and agricultural extension work (De Corato, 2020a).

Such a replacement would impose positive long-term effects and sustainable soil practices replacing short-term gains derived from mineral fertiliser usage. At the farm level, change in agricultural practice is either driven by personal motivation, economic incentives outweighting the costs for adaption or by regulatory instruments such as fines, loss of payments or reputation as a threat of potential consequences of non-compliance (Prager and Posthumus, 2010). Consequently, lower prices and higher efficiency of bio-based fertilisers could counteract the lack of trust towards such products and convince farmers to substitute (part of) mineral with organic fertilisers (Tur-Cardona et al., 2018). Also, current behavioural adaptations of European farmers often focus on economic aspects (Dessart et al., 2019), neglecting psychological characteristics such as attitudes, norms, perceived control and resources that positively influence the intentions of farmers (Daxini et al., 2018). For more effective agri-environmental policies, the socio-economic aspects of farmers' decision-making must be addressed (Dessart et al., 2019; Siebert et al., 2006).

5. Conclusions

Considering soil biodiversity in manure management provides a win-win solution for enhancing agricultural productivity, reducing farmers' costs and enabling positive environmental effects (fewer fertilisers, an increase of soil organic carbon, mitigation of contamination risks and improved nutrient efficiency rates, particularly when compared to mineral fertilisers as shown in two recent meta-analysis (Luo et al., 2018; Zhang et al., 2020)). All these together can accelerate the move towards sustainable food systems in the EU. Soil biodiversity can be increased by implementing more sustainable farming practices such as high diversity farming, limiting mechanisation and pollutants and by adding microorganisms cultivated in manure (e.g., as done in biodynamic farming practices). Microorganisms in manure that are adapted to local environmental and soil properties are found to promote soil biodiversity in the long term, to the extent that soil biodiversity in manure fertilised soils exceeds that of inorganically fertilised soils by 50% (Bengtsson et al., 2005).

Since manure can also threaten soil biodiversity, potential risks should be addressed, e.g., by treating contaminated manure or by avoiding the application of manure containing heavy metals. This review provides an overview of beneficial and harmful manure practices vis-à-vis soil biodiversity. These practices should be selectively applied, monitored and considered by policy makers to better protect and meaningful enhance soil biodiversity. Our findings may contribute to policy developments as projections for 2030 predict an increase in liquid slurry application (European Commission, 2014). This, in turn, may cause significant harm to soil biodiversity due to high concentration of heavy metals, pathogens and antibiotics. As a first step to tackle the EU's manure relevant problems, it is important to prioritise the manure quality over quantity. Putting quantity over quality harms soil biodiversity and therefore, sustainable food systems. Soil biodiversity needs to be appropriately included when budgeting manure amendments to provide better guidelines on manure use and to reduce costs and environmental risks. To counteract current mismanagement, increased monitoring and integrated research are needed.

At a strategic level, the EU is increasingly aware of the importance of protecting soil biodiversity. However, to achieve the ambitious goals of the Farm to Fork Strategy and the Circular Economy Action Plan, new/updated regulatory instruments are needed. At present, holistic manure management measures that address both the risks and benefits for soil biodiversity are beyond the scope of existing legislation. Either the scope of current legislations needs to be expanded, or new legislation is required that considers the impact of manure on soil biodiversity.

Due to the high amounts of manure that are currently being produced, their disposal is prioritised in policy agendas rather than recognising and building on its multiple benefits. It is time to adopt a more holistic approach that seizes the benefits of farm manures for soil biodiversity, as an additional strategy for soil protection in light of current and future environmental challenges.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

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