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Jeanne Maréchal

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Spécialité : « *Ecologie et Evolution* »

Par

**Jeanne MARÉCHAL**

## **Sols fertiles et Trame Brune en milieu urbain**

Impacts de l'ingénierie pédologique sur les sols et les communautés lombriciennes

**Thèse présentée et soutenue à Rennes, le 09/03/2022**

**Unité de recherche : UMR 6553 - ECOBIO**

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*Reconstituting soils for nature in cities  
Able to support strong and healthy trees  
Starts like old epic stories  
Once upon a time earthworm communities*

*Let me introduce the setting  
Roundabouts and roadside plantings  
Environments full of barriers  
Where maybe survive some soil engineers?*

*Made of stripped and transported soil material  
It's a whole world that humans try to be managerial  
Desert islands or ideal places  
What are for our heroes these new green spaces?*

*Without being emotional  
We need earthworms to make them functional  
By building structures like aggregates and galleries  
They embody adjuvants of urban territories*

*So, just a thesis to consider  
What is happening under  
Just a minute to convince  
That we should talk about this*

*J. Maréchal, 2019*

## Liste des articles

- I. Maréchal, J., Hoeffner, K., Piron, D., Pommier, T., Durand, M., Michot, D., Villenave, C., Chauvin, C., Marié, X., Cluzeau, D. Characterisation of the physical, chemical, and biological fertility of 4-year-old and 20-year-old reconstructed Anthrosoils in urban areas.
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- III. Maréchal, J., Hoeffner, K., Marié, X., Cluzeau, D. Earthworm communities of reconstructed soils in urban areas are impacted by soil engineering processes and asphalt surfaces.
- IV. Maréchal, J., Hoeffner, K., Marié, X., Cluzeau, D., 2021. Response of earthworm communities to soil engineering and soil isolation in urban landscapes. *Ecological Engineering* 169, 106307, doi : 10.1016/j.ecoleng.2021.106307.



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# INTRODUCTION

## 1. Sols fonctionnels et biodiversité

### *1.1. Contributions de la biodiversité aux fonctions des sols*

Le sol, ou couverture pédologique, est défini en pédologie comme « la couche supérieure de la croûte terrestre transformée par des processus climatiques, physicochimiques et biologiques, composée de particules minérales, de matière organique, d'eau, d'air et d'organismes vivants organisés en horizons de sols génétiques » (AFNOR, 2015). Le sol est subdivisé en volumes considérés comme homogènes appelés horizons (Baize and Girard, 2008). La formation des sols naturels résulte de l'altération de la roche-mère par l'action de l'eau, de l'air et d'êtres vivants, engendrant des transformations bio-physico-chimiques. Ce processus est très lent, nécessitant des siècles voire des milliers d'années, et dépend, en plus du temps, de la nature de la roche-mère, du climat et de la topographie. Compte-tenu de la diversité de ces situations, les sols naturels sont très variés en termes de profondeur, couleur, texture, structure, et présentent donc des propriétés physiques, chimiques et biologiques variables (Bispo et al., 2016). La World Reference Base (WRB, 2015) distingue en effet 32 groupes de sols de référence représentatifs de régions pédologiques majeures, et qui peuvent se différencier à l'issue de processus pédogénétiques secondaires. Le Référentiel pédologique français détaille ainsi 110 références de sols et plus de 320 qualificatifs (Baize and Girard, 2008).

Cette grande diversité de sols abrite de très nombreux organismes vivants<sup>1</sup> qui constituent la biodiversité du sol. Elle se définit comme la variété de la vie souterraine, des gènes et espèces aux communautés qu'ils forment, ainsi que les complexes écologiques auxquels ils contribuent et participent, des micro-habitats du sol aux paysages (FAO, 2020). Cette biodiversité

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<sup>1</sup> Il a été estimé qu'un gramme de sol contenait jusqu'à un milliard de cellules bactériennes, comprenant des dizaines de milliers de taxons, jusqu'à 200 mètres d'hyphes fongiques et un large éventail d'organismes, dont des nématodes, des vers de terre et des arthropodes (FAO, 2020).

édaphique se partage le sol à des échelles micrométriques à centimétriques (microporosité, macroporosité, agrégat, motte) et réalise de nombreuses fonctions grâce à des interactions complexes, trophiques et non trophiques, entre les composantes biotiques (i.e., organismes vivants) et abiotiques (i.e., chimiques et physiques) du sol, contribuant ainsi à la fourniture de nombreux services écosystémiques<sup>2</sup> (Groot et al., 2002; MEA, 2005; Kibblewhite et al., 2008). Par exemple, les microorganismes (bactéries et champignons) et la microfaune (e.g., protozoaires, nématodes) sont responsables de modifications chimiques qui agissent sur la dynamique et le recyclage de la matière et des nutriments. Ils participent notamment à la minéralisation des matières organiques et à la libération d'éléments minéraux qui pourront être utilisés par les plantes (Gebremikael et al., 2016; Graham et al., 2016; Nagy et al., 2017). La mésofaune (e.g., collemboles, acariens) accélère la décomposition de la litière et améliore le cycle et la disponibilité des nutriments (Rusek, 1998; Behan-Pelletier, 2011). Les invertébrés de la macrofaune (e.g., myriapodes, fourmis) participent par leurs activités de fouissement et de consommation à la structuration du sol et à l'incorporation de matière organique dans le sol (Ott et al., 2012; Cheik et al., 2019). Parmi eux, les vers terre constituent des acteurs clés de l'écosystème sol (Lavelle et al., 2006; Ojha and Devkota, 2014).

### ***1.2. Contributions des catégories écologiques lombriciennes aux fonctions des sols***

Les vers de terre, ou lombriciens en référence à la famille européenne des *Lumbricidae*, appartiennent à la sous-classe des Oligochètes et le nombre d'espèces décrites à ce jour est d'environ 3700 dans le monde (Csuzdi, 2012; Decaëns et al., 2013) et 110 en France (Bouché, 1972). Trois catégories écologiques (Figure 1) sont classiquement distinguées sur la base de traits morphologiques (taille, couleur), physiologiques (résistance à la sécheresse, durée de vie),

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<sup>2</sup> Bénéfices tirés par l'homme des écosystèmes.

et comportementaux (mobilité, milieu de vie). L'ensemble de ces caractères écologiques reflètent des stratégies écologiques et des contributions différentes aux fonctions des sols naturels ou anthropisés (Bouché, 1977; Blouin et al., 2013) :

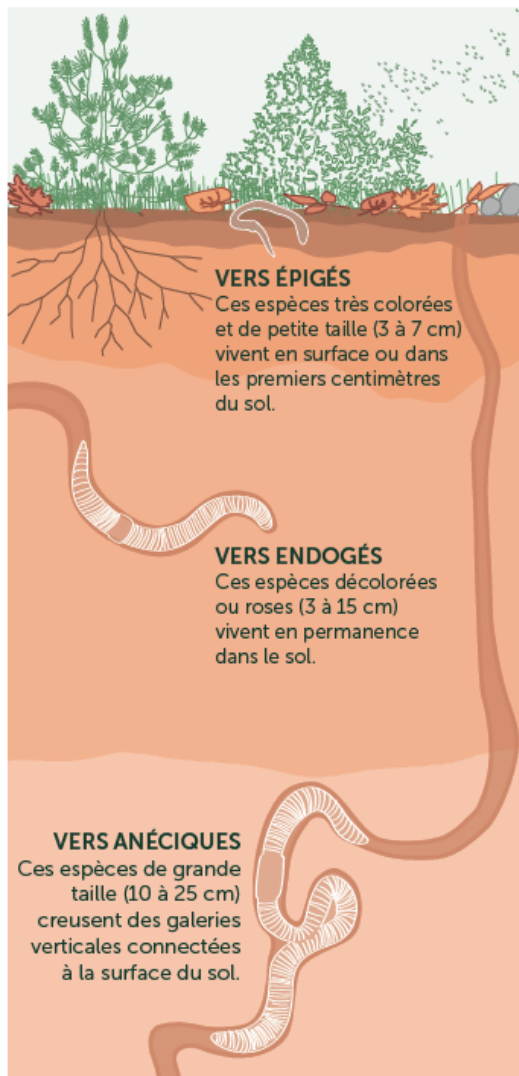


Figure 1 – Illustration des catégories écologiques lombriciennes dans le sol (source : Projet Tram'Biosol)

(1) Les vers de terre épigés sont de petite taille (3 à 7 cm), ont une pigmentation homochromique rouge brunâtre, et leur taux de reproduction est élevé (100 cocons/an). Ils sont fortement soumis aux aléas climatiques et aux prédateurs. Ils survivent au froid et à la sécheresse sous forme de cocons. Ils ne créent peu ou pas de galeries, vivent et se nourrissent de la matière organique morte ou en décomposition à la surface du sol. Ils contribuent notamment aux fonctions de recyclage de la matière organique via la décomposition des litières (Jégou et al., 1998; Manna et al., 2003), aux cycles des éléments nutritifs par la stimulation de l'activité des microorganismes (Haynes et al., 2003; Vos et al., 2019) et à la rétention en eau du sol dans leurs déjections produites à la surface du sol (Ernst et al., 2009).

(2) Les vers de terre endogés sont de petite à grande taille (3 à 15 cm), sont faiblement pigmentés et leur reproduction est limitée. Ils créent des galeries horizontales ramifiées et se nourrissent de la matière organique humifiée présente dans les horizons organo-minéraux et minéraux. Ils vivent donc constamment dans le sol et sont peu soumis aux

prédateurs et aléas climatiques. Ils peuvent entrer en quiescence lors des périodes défavorables (froid, sécheresse) pendant lesquelles ils s'enfouissent dans le sol et créent des logettes d'estivation à une humidité et température adéquates. Les endogés ont un rôle prépondérant dans la fonction de structuration des sols grâce à la production de déjections, estimée à 40-100 tonnes ha<sup>-1</sup> an<sup>-1</sup>, qui correspondent à des macroagrégats stabilisés grâce à l'incorporation de matières organiques et d'agents liants (Bossuyt et al., 2005; Sharma et al., 2017). Ils contribuent également par la création de macroporosité à l'amélioration des fonctions d'infiltration et de rétention en eau du sol (Joschko et al., 1992; Ernst et al., 2009; Hallam and Hodson, 2020). Ils participent au cycle du carbone et des éléments nutritifs, leurs déjections étant des hotspots d'activité microbienne favorisant la minéralisation du carbone et concentrant les éléments nutritifs (Amossé et al., 2015; Abail et al., 2017; Medina-Sauza et al., 2019).

- (3) Les vers de terre anéciques sont de grande taille (10 à 25 cm) et présentent un gradient de couleur antéro-postérieur de décoloration au stade adulte. Ils creusent de profondes galeries verticales tout le long du profil de sol. Les anéciques peuvent entrer en diapause ou seulement en quiescence dans des logettes d'estivation lorsque les conditions climatiques sont défavorables. Les anéciques peuvent être distingués en deux sous-catégories : épi-anéciques se nourrissent plutôt de la matière organique fraîche de surface alors que les vers de terre anéciques stricts se nourrissent préférentiellement de la matière organique âgée déjà incorporée au sol (Jégou et al., 1998; Hoeffner et al., 2019). Ainsi, les anéciques participent au stockage du carbone et au cycle des nutriments dans l'ensemble du profil grâce à l'incorporation de litière dans les horizons profonds, favorisant l'activité microbienne et la minéralisation (Brown et al., 2000; Tiunov and Scheu, 2000; Capowiez et al., 2021). Leur réseau de galeries verticales ouvertes à la

surface du sol participent à l'infiltration de l'eau et à l'aération du sol (F. Bastardie et al., 2003; Bertrand et al., 2015).

Ces différentes contributions illustrent la capacité des lombriciens à modifier les propriétés de leur habitat grâce aux bioturbations qu'ils génèrent, galeries et des agrégats (Piron et al., 2012). Ils sont ainsi considérés comme des « ingénieurs de l'écosystème sol » car leurs bioturbations modifient les ressources du sol et créent des habitats pour les autres organismes à l'échelle du profil de sol (Jones et al., 1994; Lavelle and Spain, 2001). Les vers de terre sont également qualifiés d'espèces « clés de voûte », c'est-à-dire assurant au sein des écosystèmes des fonctions essentielles qui ne peuvent être remplies par une autre espèce (Stork and Eggleton, 1991; Bond, 1994). Les catégories écologiques lombriciennes contribuent donc de façon complémentaire aux fonctions du sol, en particulier aux trois composantes physique, chimique, et biologique de la fertilité<sup>3</sup> des sols, comme synthétisé dans de nombreux ouvrages (Lee, 1985; Edwards and Bohlen, 1996; Lavelle and Spain, 2001). Ces connaissances restent néanmoins le plus souvent qualitatives (Blouin et al., 2013). Les données quantitatives disponibles sont souvent données en lien avec l'abondance totale, parfois en lien avec l'abondance des catégories écologiques, et très peu à l'échelle de l'abondance des taxons. Par exemple, les méta-analyses disponibles révèlent par exemple que la présence de vers de terre permet en moyenne d'augmenter de 23% la biomasse aérienne dans les agroécosystèmes (Van Groenigen et al., 2014) et de doubler la quantité de perte de masse de la litière (Huang et al., 2020). A l'échelle des espèces, les données sont sporadiques, souvent issues d'études en mésocosmes compte-tenu des difficultés techniques à étudier le comportement des espèces sur

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<sup>3</sup> La **fertilité** résulte d'une combinaison de composantes physique, chimique et biologique du sol qui déterminent l'approvisionnement des plantes en éléments nutritifs, les conditions de croissance et le fonctionnement des racines (Mazoyer, 2002). Elle est considérée par de nombreux auteurs comme une **fonction du sol**<sup>3</sup> au même titre que le stockage du carbone ou l'infiltration (e.g., de Groot et al., 2002; Lothodé et al., 2020).

le terrain, et par conséquent ne peuvent pas refléter la complexité des sols naturels ou anthropiques (e.g., Larink et al., 2001; Mummey et al., 2006; Deeb et al., 2016).

## **2. Sols en milieu urbain**

### ***2.1. Diversité des sols urbains***

Le milieu urbain correspond à une mosaïque de sols d'usages variés incluant des espaces résidentiels, des infrastructures commerciales, industrielles, et de transport entrecoupés d'espaces verts (Breuste et al., 2008). Selon les évolutions de la ville et de l'usage de ses espaces, les sols ont pu subir diverses perturbations telles que des excavations, nivellements, compactages, scellements, décapages, apports de matériaux terreux ou anthropiques (e.g., déblais, remblais ou restes d'activités industrielles ; Vidal-Beaudet and Rossignol, 2018a). Ainsi, les sols en milieu urbain peuvent être décrits selon un gradient d'anthropisation traduisant l'intensité des activités humaines subies, allant de sols naturels ou pseudo-naturels (i.e., ayant subi très peu de changements) à des sols fortement anthropisés (i.e., ayant subi une influence significative des activités humaines) plus ou moins fonctionnels (Pouyat et al., 2010; Morel et al., 2015; Lothodé et al., 2020). A l'extrême, la couverture pédologique peut être complètement détruite à la suite de son excavation et/ou scellement à des fins de construction d'infrastructures de transport ou de bâtis, entraînant la perte plus ou moins irréversible des fonctions écologiques assurées par les sols et leur biodiversité (Gardi et al., 2013; Geisen et al., 2019).

Sur la base du Référentiel pédologique français (Baize and Girard, 2008), les horizons et matériaux constitutifs des sols urbains peuvent être catégorisés en trois groupes selon un gradient d'anthropisation: les horizons pédologiques naturels (**N**), les horizons pédologiques naturels anthropisés (**N-**), les matériaux anthropiques (**Z-**) (Tableau 1) :



Tableau 1 – Synthèse des matériaux potentiellement identifiables dans les sols urbains sur la base du Référentiel pédologique français (Baize et Girard, 2008). Les matériaux archéo-anthropiques (Zar) sont exclus du tableau car peu représentatifs des milieux urbains. Les horizons naturels proposés dans le tableau sont des horizons de Luvisols qui correspondent au milieu de référence de notre étude en Ile-de-France (Plateau de Trappes).

	Horizons et matériaux du RP	Qualificatif	Code RP	Définition du RP
Naturel	Horizons pédologiques naturels (N)	organiques	<b>O</b>	horizons organiques constitués principalement de matières organiques directement observables
		organo-minéraux	<b>A</b>	horizons contenant en mélange des matières organiques et des matières minérales
		éluviaux	<b>E</b>	horizons organo-minéraux appauvris en fer et/ou en minéraux argileux phylliteux et/ou en aluminium, avec concentration corrélative en minéraux résistants
		argilluviaux	<b>BT</b>	horizons à structuration pédologique généralisée, non formés en surface, caractérisés par une accumulation absolue de matières (essentiellement des particules argileuses) par rapport aux autres horizons présents dans le solum
		...	<b>C</b>	Horizon de profondeur altéré, mais sans structuration pédologique
Anthropisé	Horizons pédologiques naturels anthropisés (N-)	-	<b>-z</b>	horizons peu modifiés et pouvant contenir des <b>marques d'artificialisation insuffisantes</b> pour les considérer comme des matériaux anthropiques ; volume des matériaux allochtones d'origine anthropique < 20 % du volume total de l'horizon (exemples : Az, Lz, Jz, Sz)
		-	<b>-h</b>	horizons fortement <b>enrichis en matières organiques</b>
		-	<b>-tp</b>	horizons <b>prélevés et transportés</b> (exemples : Atp, Ltp)
		-	<b>-tph</b>	horizons <b>prélevés, transportés, et enrichis en matières organiques</b> (exemples : Atp, Ltp)
		-	<b>-b</b>	horizons initiaux <b>enfouis</b> (recouverts par de nouveaux matériaux)
	Matériaux anthropiques (Z-)	-	<b>Z</b>	matériaux <b>non pédologique</b> (exemple : remblais)
		terreux	<b>Ztr</b>	matériaux d'origine pédologique ou géologique, <b>mélangés</b> , granulo <2 mm, parfois faible %EG
		technologiques	<b>Ztc</b>	matériaux issus des procédés d'extraction et de transformations par voies physiques, chimiques ou biologiques de matières premières. <b>Sous-produits</b> des activités industrielles, artisanales ou minières
		holorganiques	<b>ZO</b>	matériaux correspondant à des apports de grande quantité de matières végétales non ou peu transformées ou de <b>composts</b>

Ainsi, la succession de ces différents horizons ou matériaux permet d'identifier des solums (i.e., tranche verticale d'une couverture pédologique) de sols urbains non scellés suivant un même gradient d'anthropisation allant des sols naturels, composés d'horizons naturels non anthropisés (e.g., Luvisol), aux Anthrosoles artificiels composés de couches de matériaux anthropiques (Tableau 2) :

Tableau 2 – Synthèse des types de sols potentiellement identifiables en milieu urbain sur la base du Référentiel pédologique (RP ; Baize et Girard, 2008). Une lettre peut correspondre à un ou plusieurs horizons successifs de même nature. Les Luvisols correspondent au milieu de référence de notre étude en Ile-de-France (Plateau de Saclay). \*La définition du terme « anthropisé » utilisée dans notre étude ne correspond pas à celle du RP pour assurer la cohérence les termes précédemment définis. Les Anthroposols transformés et archéologiques sont exclus du tableau car peu présents en milieu urbain.

		Nomenclature RP	Successions d'horizons théoriques	
Sols urbains (= couverture pédologique des aires urbaines)	Sol naturel	<b>Luvisol</b> <i>Sol naturel</i>	<b>N</b>	
		<b>Luvisol anthropisé*</b> <i>Sol impacté par une action anthropique sur moins de 50 cm de profondeur</i>	<b>N- / N</b> ou <b>Z / N</b> avec <b>N-</b> et <b>Z</b> < 50 cm de profondeur	
	Sols anthropisés (= influence des activités humaines)	Non-anthroposols	<b>Anthroposol reconstitué</b> <i>Matériaux pédologiques apportés sur au moins 50 cm de profondeur pour des opérations de végétalisation urbaine</i>	<b>Ntp(h) / N</b> avec <b>Ntp(h)</b> > 50 cm de profondeur
				<b>Ntp(h)</b> > 50 cm de profondeur
		Anthroposols	<b>Anthroposol construit</b> <i>Matériaux technologiques apportés sur au moins 50 cm de profondeur pour des opérations de végétalisation urbaine</i>	<b>Ztc / N</b> avec <b>Ztc</b> > 50 cm de profondeur
				<b>Ztc</b> > 50 cm de profondeur
			<b>Anthroposol artificiel</b> <i>Matériaux anthropiques divers accumulés sur au moins 50 cm de profondeur</i>	<b>Z / N</b> avec <b>Z</b> > 50 cm de profondeur
				<b>Z</b> > 50 cm de profondeur

## 2.2. Artificialisation des sols et érosion de la biodiversité

Suite à la Loi Climat et résilience du 22 août 2021, l'artificialisation des sols est désormais définie dans l'article L.101-2-1 du code de l'urbanisme comme « l'altération durable de tout ou partie des fonctions écologiques d'un sol, en particulier de ses fonctions biologiques, hydriques et climatiques, ainsi que de son potentiel agronomique par son occupation ou son usage ». Les pertes de fonctions peuvent être générées par la perturbation des couches superficielles et

profondes des sols en lien avec les processus de construction de la ville, conduisant à des horizons compacts, hétérogènes, asphyxiants, avec peu de réserves nutritionnelles (Craul, 1985; Jim, 1998a; Li et al., 2013). Les avancées réglementaires récentes en faveur de la protection des sols s'intègrent dans un contexte d'érosion rapide et majeure de la biodiversité à l'échelle mondiale comme souligné à nouveau dans le rapport de l'IPBES<sup>4</sup> de juin 2021 (Pörtner et al., 2021). Parmi les facteurs de cette érosion, l'artificialisation des sols joue en effet un rôle essentiel et est identifiée aujourd'hui comme une des premières causes (Béchet et al., 2017). En particulier, la biodiversité du sol subit les conséquences de l'étalement urbain engendrant la réduction de leur habitat, de l'augmentation de l'hétérogénéité et de la fragmentation spatiales (FAO et al., 2020). Pour limiter l'artificialisation des sols et ses conséquences sur la biodiversité, l'objectif de zéro artificialisation nette (ZAN) a émergé du Plan biodiversité du 4 juillet 2018 et dont les concepts et leviers d'action ont été synthétisés dans le rapport de France Stratégie de juillet 2019 (Fosse et al., 2019). Ainsi, l'Etat demande explicitement aux préfets, via la circulaire interministérielle du 29 juillet 2019, des actions intervenant dans une trajectoire permettant de rendre applicable l'objectif zéro artificialisation nette des territoires en diminuant la consommation de l'espace, puis de la stopper par un usage sobre de l'espace et par des actions de type compensatoire.

La notion de compensation en faveur de la biodiversité a été introduite dès 1976 par la loi relative à la protection de la nature par le développement d'un outil réglementaire appelée séquence ERC (« Eviter, Réduire, Compenser »). Couramment appliquée pour les zones humides, la séquence ERC vise en premier lieu à sélectionner les scénarios du projet

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<sup>4</sup> La Plateforme intergouvernementale scientifique et politique sur la biodiversité et les services écosystémiques (Ipbes) est un organe intergouvernemental indépendant créé par les États membres en 2012. Il fournit aux décideurs des évaluations scientifiques objectives de l'état des connaissances sur la biodiversité de la planète, les écosystèmes et leurs bénéfices pour les individus, ainsi que les outils et les méthodes pour protéger et utiliser de manière durable ces ressources naturelles vitales (source : <https://www.fondationbiodiversite.fr/la-frb-en-action/nos-implications/ipbes/>)

d'aménagement les moins impactant pour la biodiversité via une réflexion dès les phases de conception. Les impacts ne pouvant être évités doivent être réduits pendant et après la phase travaux en adaptant par exemple les périodes d'intervention. Les impacts résiduels doivent enfin être compensés par exemple par des mesures de restauration ou de création de milieux naturels (Tarabon, 2020). Dans le contexte de l'objectif ZAN, la compensation se traduit concrètement par des opérations de renaturation de sols (ou désartificialisation), c'est-à-dire des opérations de restauration ou d'amélioration de la fonctionnalité d'un sol, ayant pour effet de transformer un sol artificialisé en un sol non artificialisé. Ainsi, le solde de l'artificialisation et de la renaturation des sols constatées sur un périmètre et sur une période donnée permet de définir une artificialisation nette<sup>5</sup>. La mesure de ce solde est aujourd'hui imparfaite car elle se base sur des fichiers fonciers<sup>6</sup> qui ne permettent pas, par exemple, de distinguer finement les degrés d'impact de l'artificialisation ou de la renaturation sur la biodiversité. Ainsi, des outils d'interprétation d'imagerie satellitaire (e.g., OCS GE de l'IGN) et des outils d'évaluation de terrain doivent poursuivre leur développement pour assurer le suivi du territoire. Le concept de renaturation implique la mise en œuvre de processus d'ingénierie pédologique.

### ***2.3. Sols issus de l'ingénierie pédologique***

En milieu urbain, les sols supports de nature en ville, illustrée par les surfaces enherbées, arbres d'alignement, forêts urbaines ou jardins urbains, constituent des éléments clés d'amélioration de la qualité des espaces urbains et du cadre de vie. Ces sols et la végétation qui y pousse apportent en effet des services complémentaires de régulation (e.g., limitation des îlots de chaleur, atténuation des inondations), de production (e.g., biomasse ornementale ou alimentaire), ou encore des services culturels (e.g., aménités paysagères, bénéfices spirituels ;

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<sup>5</sup> Article L.101-2-1 du code de l'urbanisme

<sup>6</sup> Données fiscales cadastrées, qui enregistrent les changements d'usage des sols, en particulier le passage d'un espace naturel, agricole ou forestier en terrain à bâtir.

Blanchart et al., 2018). En réponse à une attente sociétale forte de verdissement des villes croisée à une artificialisation croissante (Vidal-Beaudet and Rossignol, 2018a; Clergeau, 2020), des pratiques de construction de sols fertiles (i.e., Anthrosols reconstitués et construits) à des fins de végétalisation se développent, regroupées sous les termes d'ingénierie pédologique (Damas and Coulon, 2016). Dans le cadre d'opérations d'ingénierie pédologique (e.g., création d'une zone d'aménagement, réhabilitation de friches industrielles), et notamment de renaturation, la fertilité des sols est la fonction principale recherchée, mais des fonctions complémentaires peuvent être attendues : la portance dans le cadre de reconstitution de sol en mélange terre-pierres (i.e., association de terre fertile et d'un squelette minéral) dont les propriétés mécaniques peuvent par exemple répondre à des usages de stationnement de véhicules ; l'infiltration dans le cadre de reconstitution de sol de noues d'infiltration. Les sols fertiles sont reconstitués sur le modèle de sols naturels par superposition d'horizons favorables au développement racinaire (i.e., Anthrosols reconstitués ou construits). Chacun des horizons présente donc des propriétés physico-chimiques variables, adaptées pour répondre aux différents objectifs de sols support de végétation<sup>7</sup>. La norme NF U 44-551<sup>8</sup> (AFNOR, 2002) définit en ce sens des types de matériaux selon leur teneur en matière organique telles que la « terre végétale<sup>9</sup> » et la « terre-support<sup>10</sup> ». La fertilité des matériaux pédologiques excavés peut être améliorée notamment par des amendements organiques (e.g., compost) qui ont montré de nombreux bénéfices, entre autres, sur la stabilité des agrégats, la disponibilité des nutriments,

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<sup>7</sup> Un sol est dit fertile s'il présente une structure et une profondeur permettant aux plantes de développer leurs racines pour s'ancrer, retient suffisamment d'humidité et évacue l'eau en excès, et contient suffisamment d'éléments nutritifs (N, P, K) et d'oligo-éléments pour assurer la nutrition des plantes (Marié and Rossignol, 1997; Damas and Coulon, 2016).

<sup>8</sup> Support de culture : dénomination, spécification, marquage. Cette norme fixe les dénominations et les spécifications des supports de culture avec et sans engrais. Elle fixe également les éléments particuliers de marquage, complémentaires aux éléments de marquage définis par la réglementation.

<sup>9</sup> Terre issue d'horizons de surface humifères ou d'horizons profonds pouvant être mélangée avec des matières organiques végétales, des amendements organiques et/ou des matières minérales, avec une teneur finale en matière organique variant entre 3 et 15% (Afnor, 2002)

<sup>10</sup> Terre définie selon les mêmes termes que la terre végétale, à l'exclusion de tout mélange possible avec des matières organiques. La teneur finale en matière organique est comprise entre 1 et 5%.

l'activité microbienne et la masse volumique apparente (Sarwar et al., 2010; Chaoui et al., 2003; Diacono and Montemurro, 2010; McGrath et al., 2020).

Dans le cadre de prestations paysagères commandées par les grands aménageurs publics ou privés, les acteurs des métiers du paysage se réfèrent en France aux préconisations techniques regroupées dans deux documents de référence de la filière du Paysage : le fascicule 35<sup>11</sup> (Ministère de l'Economie des Finances et de l'Industrie, 1999; Ministère de la Transition écologique et de l'Economie, 2021) et les règles professionnelles<sup>12</sup> (Unep, 2012). Les éléments techniques relatifs à l'ingénierie pédologique à destination des aménageurs décrivent les différentes étapes de construction de sols à partir de matériaux pédologiques (i.e., Anthrosoles reconstitués) : décapage, stockage, transport de l'horizon de surface organo-minéral d'un sol ressource naturel ou agricole et remise en place d'horizons (Figure 2). Les principaux éléments techniques sont recensés dans le Tableau 3.

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<sup>11</sup> Le fascicule n°35 du cahier des clauses techniques générales (CCTG) est rédigé les acteurs de la filière du paysage, sous l'égide des Ministères de la Transition écologique et de l'Economie. Il traite de l'exécution des marchés de travaux neufs ou d'entretien des aménagements paysagers extérieurs et sert de base à l'élaboration des pièces techniques et financières de marchés de travaux. Ce dernier est précisé par le cahier des clauses techniques particulières (CCTP) spécifique à chaque marché et opération d'aménagement.

<sup>12</sup> Les règles professionnelles du paysage ont été rédigées en 2012 par des professionnels du paysage (entreprises, donneurs d'ordre, bureaux d'étude, enseignants, fournisseurs, experts) afin de compléter les prescriptions générales du fascicule 35 en décrivant les techniques mises en œuvre afin de les intégrer dans les CCTP des marchés de travaux.

Dans la version du CCTG de 1999, les sols sont identifiés comme des sources de matériaux terreux appelés « terres en place » en référence à l'ensemble du sol, ou « terre végétale » en référence aux matériaux terreux les plus organiques de surface. L'évaluation de ces matériaux se limite à une analyse physico-chimique et aucune référence à la biologie du sol n'apparaît. La reconstitution de sols consiste en l'apport d'une unique couche de terre végétale d'épaisseur variable selon le type de plantation envisagée. Les terres excédentaires sont proposées à la mise en décharge. De manière générale, le sol dans sa vision de système écologique n'est pas pris en compte.



Figure 2 – Etapes du processus d'ingénierie pédologique de reconstruction de sol dans le cadre d'un projet d'aménagement paysager

Tableau 3 - Synthèse des préconisations techniques relatives à l'ingénierie pédologique dans le cadre de projets d'aménagement (Fascicule 35 et règles professionnelles)

Phase concernée	Etapas du processus	Fascicule 35 Version 1999	Fascicule 35 Version 2021	Règles professionnelles Version 2012
Phase avant Travaux	Evaluation des matériaux ressource	<ul style="list-style-type: none"> <li>- Analyses physico-chimiques des terres en place sur le site d'aménagement ou fournies afin de vérifier que leurs caractéristiques soient adaptées à la nature des futures plantations : granulométrie, teneur en matière organique, pH, C/N, teneur en calcaire total et en calcaire actif ; teneurs en N, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O, MgO, absence de contamination par des substances phytotoxiques</li> </ul>	<ul style="list-style-type: none"> <li>- Caractérisation physico-chimique des sols en place : CEC, granulométrie cinq fractions, pH eau, pH KCl, calcaire total et actif, matière organique, N organique, rapport C/N, conductivité, quantification en N, P, K, Mg, Mn, Fe, Cu, Zn, B</li> </ul>	<ul style="list-style-type: none"> <li>- Caractérisation et analyse des terres ressources : les analyses de routine (granulométrie de la terre fine, éléments grossiers supérieurs à 2 mm, calcaire, matière organique, carbone organique, azote total, pHeau et pHKCl, conductivité, CEC, éléments nutritifs majeurs : phosphore assimilable, cations échangeables Ca<sup>2+</sup>, Mg<sup>2+</sup> et K<sup>+</sup>) et/ou analyses particulières (oligo-éléments, polluants, indice de battance, capacité de rétention et point de flétrissement, tests de germination)</li> <li>- Selon la nature des sols en place, leur caractérisation, les résultats d'analyse d'une part, et des aménagements souhaités d'autre part, les sols en place seront conservés ou non. Si les sols en place sont utilisés, des améliorations pourront leur être apportées, soit par travail du sol, soit par apport d'amendements organiques et minéraux, et/ou aussi par drainage.</li> </ul>
Phase Travaux	Décapage	<ul style="list-style-type: none"> <li>- Réalisation en conditions de sol sec ou suffisamment ressuyé pour éviter tout compactage des terres végétales existant sur l'emprise des chantiers.</li> <li>- Décapage de la terre végétale doit éviter la contamination des terres décapées par des matériaux étrangers ou de terre de sous-sol de composition physico-chimique différents.</li> <li>- Epaisseur déterminée par les résultats des analyses préalables et après sondage</li> <li>- Les caractéristiques des engins mécaniques à utiliser doivent permettre de ne pas modifier les caractéristiques des terres à décapier et celles des sols restant en place.</li> </ul>	<ul style="list-style-type: none"> <li>- Réalisation en conditions de sol sec ou suffisamment ressuyé, pour éviter tout compactage.</li> <li>- Décapage du sol fertile conduit de manière à éviter de mélanger les horizons du sol et de le contaminer par incorporation de matériaux étrangers ou de terre d'autre qualité.</li> <li>- Epaisseur déterminée par l'épaisseur du sol fertile.</li> <li>- Les caractéristiques des engins mécaniques doivent permettre de ne pas modifier les caractéristiques des terres à décapier et des sols restant en place.</li> </ul>	<ul style="list-style-type: none"> <li>- Décapage effectué lorsque le sol est suffisamment sec (humidité inférieure à la teneur en eau à la limite de plasticité).</li> <li>- Décapage effectué par couches, une couche pouvant contenir un ou plusieurs horizons.</li> <li>- La profondeur de décapage peut déterminer la teneur en matière organique du matériau décapé.</li> </ul>
	Stockage	<ul style="list-style-type: none"> <li>- Dépôt effectué sur une aire préparée et non polluée.</li> <li>- Les tas ou les cordons ne dépassant pas respectivement un ou deux mètres de hauteur.</li> <li>- Tas soigneusement « fermés » pour éviter la pénétration de l'eau.</li> <li>- Le matériau ne doit pas être compacté.</li> </ul>	<ul style="list-style-type: none"> <li>- Aucune préconisation</li> </ul>	<ul style="list-style-type: none"> <li>- Différents stockages à prévoir lorsque différents horizons pédologiques ont été identifiés</li> <li>- La forme des andains triangulaire et n'admettant pas de surface plane</li> <li>- Orientation des andains dans le sens de la pente pour empêcher la stagnation d'eau.</li> <li>- Les engins de chantier ne doivent pas rouler sur le stock, notamment lors de sa constitution.</li> <li>- Si la durée de stockage le permet, il est recommandé de semer l'andain avec une plante herbacée (phacélie, gazon...)</li> </ul>
	Réalisation des fosses de plantation	<ul style="list-style-type: none"> <li>- Sol favorable (i.e., sol aéré, non toxique et alimenté en eau, même si peu fertile ; surface du sol revêtue de terre végétale) : volume de 0.125 m<sup>3</sup> à 6 m<sup>3</sup> et profondeur de 0.30 m à 1.50 m selon les végétaux</li> <li>- Sol hostile (i.e., sites dans lesquels les plants sont directement environnés de bitume, dalle de béton ou autre revêtement imperméable à l'eau ou à l'air ; sols compactés) : volume de 0.35 m<sup>3</sup> à 12 m<sup>3</sup>, profondeur de 0.30 m à 1.50 m</li> </ul>	<ul style="list-style-type: none"> <li>- Sol favorable : sols en place décompactés sur 1-1.2 m de profondeur pour les arbres, 50-70 cm pour les arbustes et 2 à 4m<sup>2</sup> de surface, 20 cm pour les herbacées</li> <li>- Sol défavorable : creusement d'une fosse de plantation de 10 à 20 m<sup>3</sup> sur une profondeur d'environ 1 à 1.20 m</li> </ul>	<ul style="list-style-type: none"> <li>- Sol en place et encaissant favorables : fosses réalisées à partir des matériaux des sols en place sur 2 à 4 m<sup>3</sup>. Les matériaux sont déblayés puis remblayés dans l'ordre des horizons du sol initial.</li> <li>- Sol en place et encaissant défavorables : fosses reconstituées soit avec réutilisation partielle des matériaux de déblais amendés, soit avec leur remplacement total par des matériaux fertiles extérieurs au site sur 10 à 20 m<sup>3</sup>.</li> </ul>
	Reconstitution du sol	<ul style="list-style-type: none"> <li>- Mise en œuvre de terres végétales ou substrats selon les profondeurs décrites ci-dessus</li> <li>- Des amendements et engrais peuvent être mis en œuvre selon des doses établies en fonction des analyses et préconisations. Ils doivent être répartis uniformément par épandage.</li> </ul>	<ul style="list-style-type: none"> <li>- Sol favorable : réutilisation des matériaux des sols en place</li> <li>- Sol défavorable : réutilisation partielle des matériaux de déblais amendés ou remplacement total par des matériaux fertiles extérieurs au site</li> <li>- Epaisseur et nature de la terre selon le type de végétation :             <ol style="list-style-type: none"> <li>(1) Strate herbacée : 20 à 30cm</li> <li>(2) Strate arbustive : 40 à 60 cm</li> <li>(3) Strate arborée : 80 à 120 cm</li> </ol> </li> </ul>	<ul style="list-style-type: none"> <li>- Typologie de trois types de sols reconstitués à partir de matériaux types définis par la norme NFU 44-551 :             <ol style="list-style-type: none"> <li>(1) Strate herbacée : un horizon de 20 à 30 cm de terre support</li> <li>(2) Strate arbustive : un horizon de 20 à 30 cm de terre végétale sur un horizon de 20 à 40 cm de terre support</li> <li>(3) Strate arborée : un horizon de 20 à 30 cm de terre végétale sur un horizon de 40 à 100 cm de terre support, sur un horizon profond de matériau terreux de 0 à 60 cm</li> </ol> </li> <li>- Mise en place des sols reconstitués à la pelleuse par couches de faibles épaisseurs sans tassement</li> <li>- Utilisation d'engins à faible portance, chemin d'accès unique qui doit être décompacté en reculant</li> </ul>
Phase après Travaux	Gestion des terres excédentaires	<ul style="list-style-type: none"> <li>- Les terres excédentaires résultant des travaux liés à la réalisation des réseaux, y compris les ouvrages annexes s'y rapportant (regard, boîte de raccordement, chambre, ..), sont évacuées en décharge</li> </ul>	<ul style="list-style-type: none"> <li>- Si les sols en place ne peuvent être réemployés, ils doivent être valorisés (de préférence sur place)</li> </ul>	<ul style="list-style-type: none"> <li>- Valorisation des matériaux du sol en place dans le cadre de l'aménagement avec amélioration et amendements si nécessaire</li> <li>- Evacuation si nécessaire des matériaux d'origine (pollution)</li> </ul>



En lien avec l'amélioration des connaissances sur les sols et sur les prises de conscience récentes de la fragilité de ces écosystèmes, la version révisée du CCTG entrée en vigueur le 15 octobre 2021, met au cœur du fascicule la durabilité du projet et son adéquation avec le sol existant. Ainsi, trois objectifs clés sont mis en avant : (1) préserver les sols existants, (2) valoriser les matériaux en place afin de créer des sols fertiles, (3) fournir de la terre végétale extérieure au chantier, en dernier recours. Dès lors, le sol est moins considéré comme un simple matériau que comme une ressource naturelle rare non renouvelable. Par exemple, la préservation ne se porte plus sur la terre végétale uniquement qui doit être décapée et mise de côté, mais par la préservation des sols existants, considérés a priori dans leur tridimensionnalité. Cette considération écologique du sol s'illustre aussi par la volonté de ne pas mélanger les « horizons » du sol, terme qui n'apparaissait pas dans la version de 1999. Des détails sont apportés en matière de reconstitution de sol. La notion de « sols fertiles » apparaît mais aucune définition claire n'est apportée. Une confusion entre la notion de terre végétale et sol fertile semble exister : l'épaisseur du sol fertile étant indiquée comme celle à décapier. Des pièces de marché supplémentaires sont également demandées au maître d'ouvrage, à savoir (1) un plan des sols fertiles, (2) un plan des sols à évacuer, et (3) un plan des sols à reconstituer sur le site. Comme dans la version de 1999, les préconisations sont restreintes à un objectif de fertilité des sols : aucune préconisation n'est apportée concernant les organismes du sol ou les paramètres biologiques à évaluer dans les sols existants ou reconstitués. La fertilité des sols mis œuvre n'est pas vérifiée à l'issue des travaux, mais les arbres, arbustes ou vivaces plantés font l'objet d'un constat de reprise qui traduit indirectement la capacité des sols reconstitués à être support de développement de végétation. Les végétaux morts, endommagés, altérés ou en mauvais état sanitaire sont considérés comme non repris. Dans le cadre d'aménagements urbains, le taux de reprise doit être de 100% pour les arbres, et de 80% dans le cadre de boisements ou dépendances d'infrastructures autoroutières et routières non urbaines.

En 2012, les règles professionnelles préconisaient déjà par rapport au CCTG alors en vigueur « la valorisation des sols existants pour préserver les ressources naturelles, réduire les coûts d'achat de terres végétales et réduire les déplacements engendrés par le transport ». De manière générale, les règles professionnelles proposent une vision écologique des sols, définis comme un compartiment primordial non renouvelable pour l'écosystème terrestre, constitué de matières minérales et organiques, abritant de nombreux êtres vivants, organisés en horizons caractérisés par leurs constituants, l'agencement de ceux-ci et leurs propriétés physico-chimiques et biologiques. Un effort de définition de l'ensemble des termes relatifs à l'ingénierie pédologique et aux sols est réalisé (e.g., fertilité, sols en place, terres ressources, éléments de caractérisation des sols en place). Ces règles constituent à l'heure actuelle le document technique le plus abouti pour la filière du paysage en faveur de la préservation des sols et de leur fertilité. Cependant, les enjeux de biodiversité n'y sont pas identifiés. En effet, aucune préconisation n'est formulée dans un but d'évaluation ou de préservation de la biodiversité des sols dans le cadre de projets d'aménagements paysagers malgré leur rôle clé dans la réalisation des fonctions des sols et notamment leur fertilité (cf. partie 1).

### **3. Biodiversité lombricienne dans les sols urbains**

Une communauté est un assemblage d'espèces dont l'organisation (distribution, composition, structure) est régie par des contraintes externes ou filtres environnementaux (Belyea and Lancaster, 1999). Decaëns et al. (2008) décrit cinq principaux filtres régissant l'assemblage des communautés lombriciennes : (1) le filtre biogéographique, (2) le filtre des propriétés du sol et du climat (habitat), (3) le filtre d'occupation et de gestion des sols, et (4 et 5) les filtres d'interactions entre espèces. En milieu urbain, chacun des filtres est susceptible d'être particulièrement affecté par des perturbations anthropiques diverses, impactant les communautés lombriciennes, qui en tant qu'espèces « bioindicatrices » de l'état écologique des

sols, traduiront ces perturbations par des réponses en termes d'abondance, de biomasse, de richesse ou de structure de communauté (Bispo, 2008; Cluzeau et al., 2012).

### ***3.1. Réponse des communautés lombriciennes aux perturbations anthropiques***

Les études disponibles indiquent toutefois des effets des propriétés physicochimiques des sols urbains (Amossé et al., 2016; Audusseau et al., 2020), de l'usage des sols (Xie et al., 2018; Li et al., 2020; Tóth et al., 2020), et de la gestion des sols (Smetak et al., 2007; Vergnes et al., 2017). Par exemple, Smetak et al. (2007) ont observé que l'ajout d'engrais azotés dans les parcs urbains étaient associés à une augmentation de l'abondance d'une espèce anécique. Ainsi, les gammes d'abondance observées s'étendent de 0 à plus de 500 individus m<sup>-2</sup>. Enfin, les quelques études impliquant des sols issus de l'ingénierie pédologique, des Anthrosols construits (ou Technosols) en particulier, correspondant à des sols très récents (< 5 ans) de parcelles expérimentales qui reflètent des cas de figure encore rares en milieu urbain. Ils soulignent cependant l'importance des matériaux utilisés et des habitats en continuité avec ces Anthrosols, déterminants pour les processus de colonisation (Cortet et al., 2013; Burrow, 2018; Pruvost et al., 2020). De manière générale, les communautés lombriciennes ont été relativement peu étudiées en milieu urbain. Guillard et al. (2018) soulèvent des résultats d'abondance contradictoires le long de gradients urbains-ruraux notamment (e.g., Szlavecz et al., 2006 and Smetak et al., 2007), soulignant le manque de généralité des études existantes ainsi que la variabilité des communautés lombriciennes dans les sols urbains.

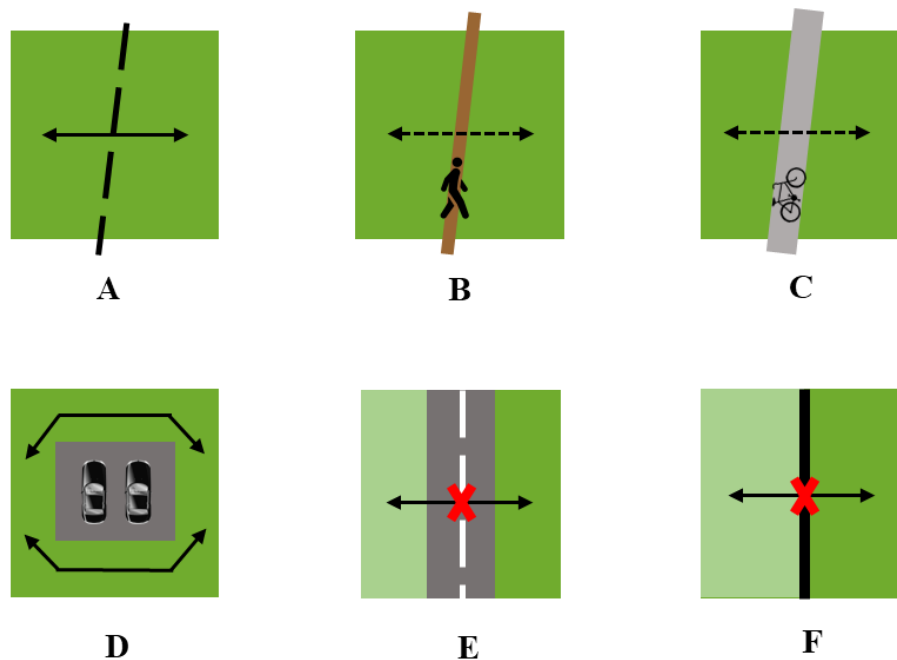
Les perturbations anthropiques peuvent constituer un facteur déclenchant de l'émigration des vers de terre pour échapper à des conditions d'habitat défavorables (e.g., propriétés du sol, faible disponibilité des ressources trophiques) et atteindre des habitats de meilleure qualité (Mathieu et al., 2010; Caro et al., 2013b). Ainsi, les capacités de dispersion des espèces lombriciennes déterminent grandement leur facilité à répondre à un changement du milieu. Des

synthèses bibliographiques relatent des capacités de dispersion pouvant aller de 1.5 à 14 m.an<sup>-1</sup> (Eijsackers, 2010, 2011). En conditions contrôlées, des études ont permis de relier des différences de comportement dispersif aux catégories écologiques lombriciennes (Caro et al., 2013a; Chatelain and Mathieu, 2017). Les connaissances sur la dispersion des vers de terre restent cependant encore peu nombreuses, notamment en milieu urbain où les trois étapes de la dispersion (i.e., départ du lieu initial, déplacement, établissement dans le lieu d'arrivée ; Ims and Yoccoz, 1997; Mathieu, 2015) peuvent être fortement impactées par une grande variété de perturbations et d'infrastructures anthropiques.

### ***3.2. Communautés lombriciennes et fragmentation du paysage urbain***

Le concept des réseaux écologiques, ou trames (Taylor et al., 1993), permet de définir deux éléments clés du paysage dans la dispersion des espèces : des réservoirs servant d'habitats aux espèces, et de corridors (linéaires ou en « pas japonais ») servant d'espaces de déplacement reliant les patches d'habitats et assurant ainsi la continuité écologique entre les habitats (Burel et Baudry, 1999). La continuité écologique du paysage correspond au degré selon lequel le paysage facilite ou altère le déplacement des individus d'un habitat à un autre (Taylor et al., 1993). En milieu urbain, les continuités écologiques peuvent être notamment altérées par les infrastructures anthropiques qui fragmentent le paysage en séparant les habitats en plusieurs éléments (Fahrig, 2003). Par exemple, l'étude de Dupont et al. (2017) suggèrent que les prairies constituent des lieux favorables à la dispersion lombricienne tandis que les routes constituent plutôt des obstacles. A l'inverse, les observations de Chuang and Chen (2008) permettent d'assimiler les routes et trottoirs à de potentiels corridors, notamment après de fortes pluies facilitant le déplacement des lombriciens sur les surfaces asphaltées. En s'inspirant de la théorie des « îles biogéographiques » (MacArthur et Wilson, 1967), les espaces verts isolés dans la ville constitueraient des espaces insulaires soumis à des modes de dispersion et d'échanges

biologiques comparables à ceux des îles océaniques isolées de la terre ferme (Saint-Laurent, 2005). Ainsi, le nombre d'espèces présentes dans une « île » (e.g., squares, ronds-points) résulterait d'un équilibre entre leur taux d'extinction et leur taux de colonisation depuis un « continent » (i.e., réservoir). De manière général, la qualification des réservoirs et des corridors est mal définie en milieu urbain pour les vers de terre, de même que l'impact des barrières anthropiques sur leur dispersion. Il existe pourtant en milieu urbain une grande diversité de barrières anthropiques, engendrant soit une perte d'habitat soit une coupure, dont le degré de perméabilité au passage des vers de terre n'est pas quantifié à l'heure actuelle (Figure 3).



*Figure 3 – Schémas simplifiés des types de barrières anthropiques (modifié d'après Sordello et al., 2014). Hypothèses de perméabilité associées : A = clôture perméable, n'engendrant ni fragmentation ni perte d'habitat ; B = chemin de terre gravillonné semi-perméable engendrant une faible fragmentation et une dégradation partielle de l'habitat ; C = trottoir ou piste cyclable asphaltée engendrant une fragmentation partielle et une disparition d'habitat ; D = parking asphalté engendrant une destruction d'habitat, pas de fragmentation des habitats restant ; E = route asphaltée engendrant une fragmentation totale et une destruction d'habitat ; F = mur avec fondations profondes engendrant une fragmentation totale avec perte d'habitat.*

Une meilleure compréhension des communautés lombriciennes au sein des paysages urbains permettrait de discuter d'une organisation en méta-communautés, c'est-à-dire ensemble des communautés locales interconnectées par la dispersion de multiples espèces potentiellement en

interaction (Gilpin et al., 1991; Wilson, 1992). Ce concept, qui découle de la théorie biogéographique, met en avant une organisation des communautés sous l'influence à la fois d'interactions locales mais aussi de processus régionaux. Concernant les vers de terre, ce fonctionnement en méta-communautés se justifierait au sein d'un territoire d'aménagement d'un part comme la conséquence directe de la fragmentation et l'isolement des habitats lombriciens par les ouvrages anthropiques (e.g., bâti, infrastructures de transport) impactant la dispersion active des lombriciens (i.e., par leurs propres moyens) ; d'autre part comme la conséquence de mouvements de terre (e.g., terres décapées, mottes de plantation) engendrant des phénomènes de dispersion passive (i.e., via un agent de transport ; James and Hendrix, 2004; Eijsackers, 2010, 2011).

Le concept de Trame Brune est encore mal identifié dans les paysages urbains et la littérature scientifique internationale (Pouyat et al., 2010; Pickett et al., 2011; Pouyat and Trammell, 2019), et aucune définition n'existe dans le droit. Actuellement décrite comme l'équivalent pour les sols de la Trame Verte pour la végétation urbaine dans la fourniture de services écosystémiques (Pouyat et al., 2010; Pouyat and Trammell, 2019), son rôle écologique reste à définir. En tant que réseau écologique, la Trame Brune serait constituée de réservoirs et de corridors pédologiques ; ces derniers assureraient la continuité écologique entre tous les types de sols (naturels ou anthropisés) à l'échelle de la ville ou du quartier. Elle devrait permettre les déplacements d'espèces vivant toutes leurs étapes de vie dans le sol (i.e., faune géobionte telle que les vers de terre ; Wallwork, 1970) et ayant des capacités de dispersion limitées (i.e., appendices arthropodiques absents ou très réduits et absence d'ailes au stade adulte ; Knop et al., 2011). En milieu urbain, la Trame Brune serait donc soumise à la fois à des contraintes d'anthropisation et d'isolement des sols.

#### **4. Modalités de l'étude**

Les modalités de sol étudiées au cours de cette thèse ont été sélectionnées à partir de modalités préexistantes sur le terrain. Plusieurs critères de sélection ont été considérés afin d'assurer la robustesse de l'étude tels que l'homogénéité du contexte pédoclimatique, la proximité des sites entre eux, la connaissance de l'historique des sols disponibles, la longueur des ouvrages permettant la réalisation de répliqués vrais, mais également les autorisations d'accès par les villes concernées. Trois sites d'étude répondant à ces critères ont été ciblés répartis entre un boulevard du quartier de l'école polytechnique de Palaiseau au sein de la communauté d'agglomération de Paris-Saclay et deux avenues de Guyancourt au sein de la communauté d'agglomération de Saint-Quentin-en-Yvelines (Figure 4 et Figure 5Figure 2). Chacun des sites présente des modalités de sol étudiées selon (1) un gradient d'anthropisation et (2) un gradient d'isolement spatial.



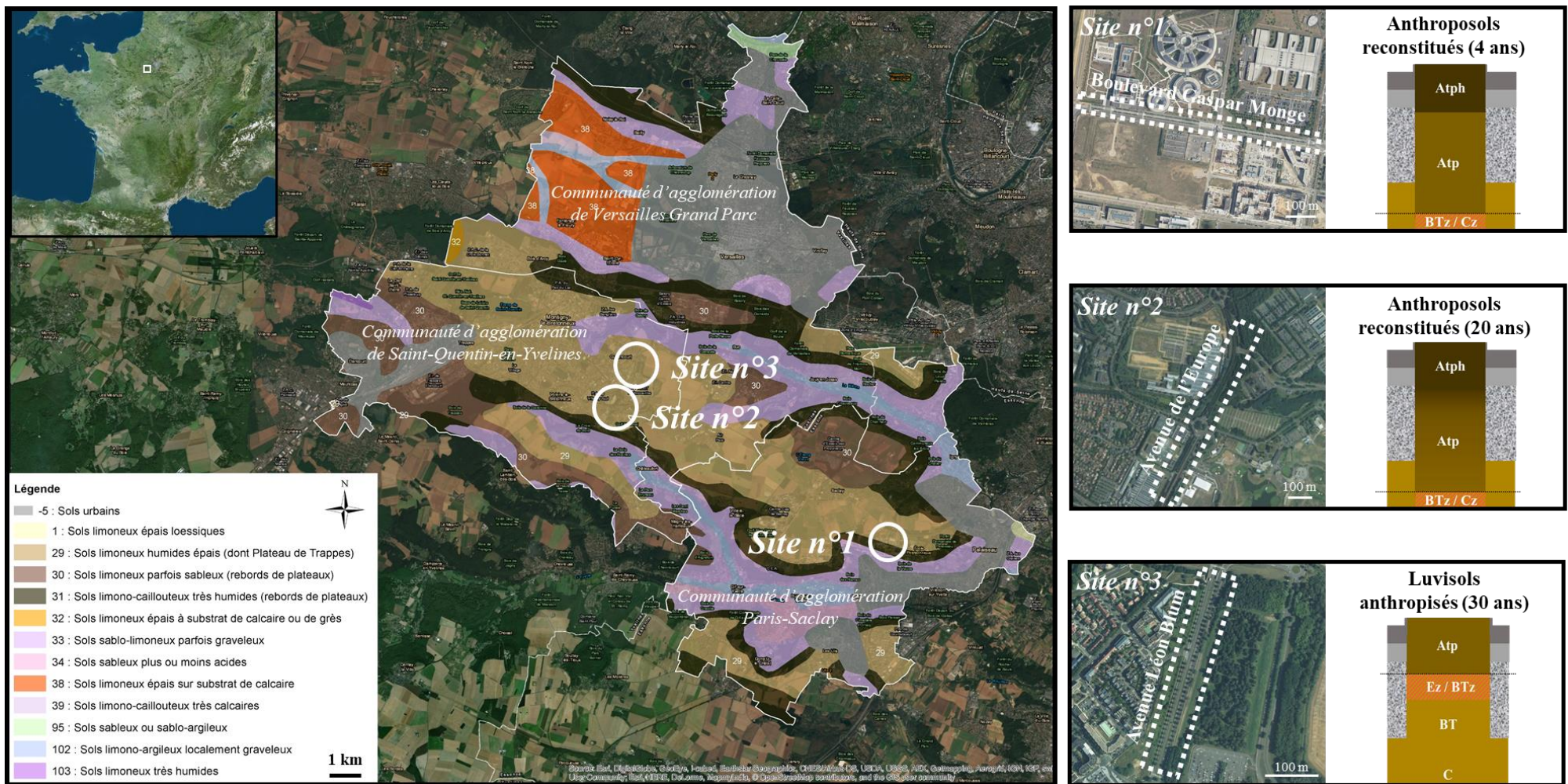


Figure 4 - Localisation des sites d'étude sur la carte pédologique des communautés d'agglomération de Versailles Grand Parc, Saint-Quentin-en-Yvelines et Paris-Saclay (d'après la carte du Référentiel Régional Pédologique de la région Ile-de-France au 1/250000 ; Roque, 2003). Source photos aériennes : Géoportail.





**Site n°1** – Anthrosols reconstitués (4 ans)  
*Boulevard Gaspar Monge, Palaiseau*



**Site n°2** – Anthrosols reconstitués (20 ans)  
*Avenue de l'Europe, Guyancourt*



**Site n°3** – Luvisols anthropisés (30 ans)  
*Avenue Léon Blum, Guyancourt*

*Figure 5 - Linéaires d'accompagnement de voirie étudiés dans le cadre de la thèse (Photos : J. Maréchal)*

#### ***4.1. Modalités du gradient d'anthropisation***

Trois modalités du gradient d'anthropisation décrit en 2.1 ont été sélectionnées pour notre étude et correspondent aux trois premiers types de sol synthétisés dans le Tableau 2 : (1) Luvisol, (2) Luvisol anthropisé, (3) Anthroposol reconstitué. Les Luvisols<sup>13</sup> correspondent aux sols représentés sur le Plateau de Trappes<sup>14</sup> où sont localisés nos sites d'étude (Roque, 2003). Ils seront considérés par la suite comme sols témoins. Les Luvisols anthropisés (30 ans) sont définis comme des Luvisols tronqués sur environ 30 cm de profondeur pour les besoins de la construction de la route, suivi par la réinstallation d'une épaisseur similaire de terre végétale décapée pour permettre la mise en place d'un couvert herbacé. Ils sont situés avenue Léon Blum à Guyancourt (Figure 4). Ce processus sera par la suite qualifié de « basique » car peu d'étapes sont impliquées et aucune modification des matériaux n'est apportée. Les Anthroposols reconstitués correspondent à des sols entièrement reconstruits sur 1.50 m de profondeur dont un horizon profond d'1.20 m de terre végétale décapée, au-dessus un horizon de mélange terre-compost de 30 cm d'épaisseur puis un mulch organique en surface (broyat de déchets verts pour les sols de 4 ans et écorces de pin pour les sols de 20 ans). Les sols de 4 ans sont situés boulevard Gaspar Monge à Palaiseau, les sols de 20 ans sont situés avenue de l'Europe à Guyancourt (Figure 4). Ce processus sera qualifié d'« élaboré » car il implique plusieurs étapes de mise en

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<sup>13</sup> Sol limoneux épais, hydromorphe, reposant sur argile. Roche-mère de type limon éolien.

<sup>14</sup> Plateau limoneux humide sur argile à meulière de Montmorency.

œuvre d'horizons avec une incorporation de compost. Les coupes schématiques de sol sont présentées en Figure 6.

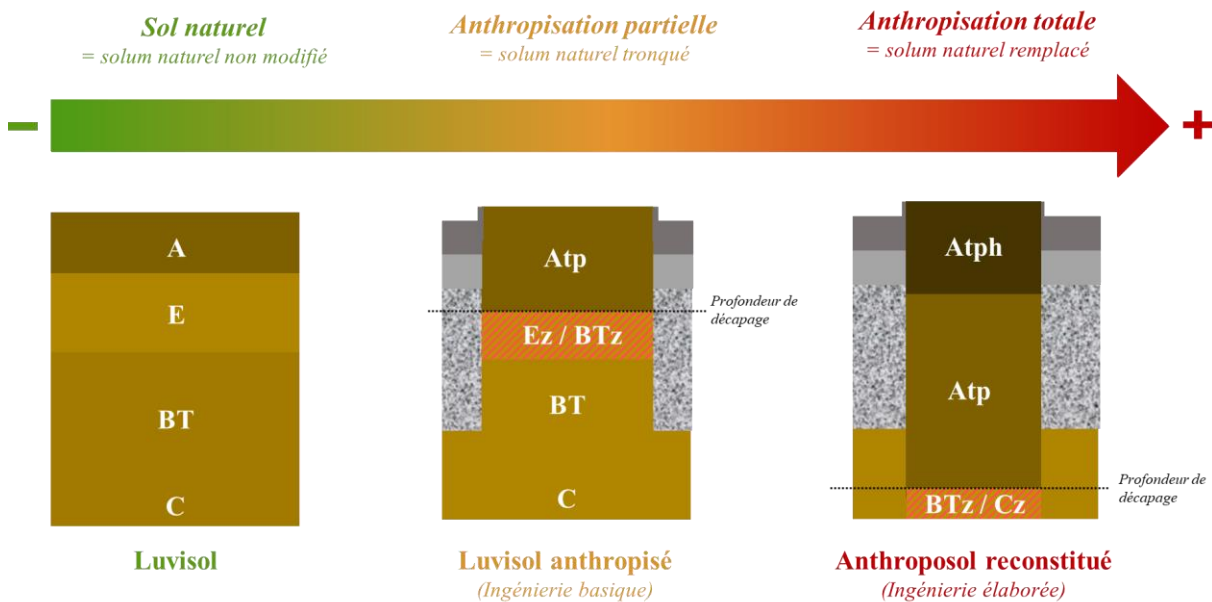


Figure 6 - Modalités d'étude présentées selon un gradient d'anthropisation (voir Tableau 1 pour les légendes des horizons ; les éléments gris correspondent à des couches constitutives de la route)

#### 4.2. Modalités du gradient d'isolement

Pour chacune des modalités de sols anthropisés (Luvisols anthropisés et Anthroposols reconstitués), deux modalités d'isolement ont été définies : (1) isolement partiel et (2) isolement total (Figure 7). Dans notre étude, l'isolement partiel d'un sol définit une séparation avec un potentiel sol réservoir par une surface asphaltée de faible largeur ( $\leq 3$  m), sans bordure, et assurant le passage de piétons ou de vélos, permettant *a priori* le passage des vers de terre par la surface. L'isolement total d'un sol définit une séparation de part et d'autre avec un potentiel sol réservoir par une surface asphaltée de largeur importante ( $\geq 6$  m) assurant la circulation de voitures, avec une bordure de voirie de 20 cm de hauteur, ne permettant pas *a priori* le franchissement par les vers de terre pour rejoindre le sol en milieu de route. Le sol sous les voiries est également considéré infranchissable du fait du remplacement du solum pédologique, sur 30 à 50 cm d'épaisseur, par des graves constituant l'assise des bétons bitumineux de

revêtement de chaussée. De plus sous les assises, le sol support est fortement compacté pour assurer une bonne stabilité mécanique à l'ensemble. Les sols de types réservoirs sont caractérisés au contraire par une connectivité totale en surface et en profondeur. Une coupe transversale schématique des modalités d'isolement est proposée en Figure 8.



Figure 7 - Modalités d'étude présentées selon un gradient d'isolement



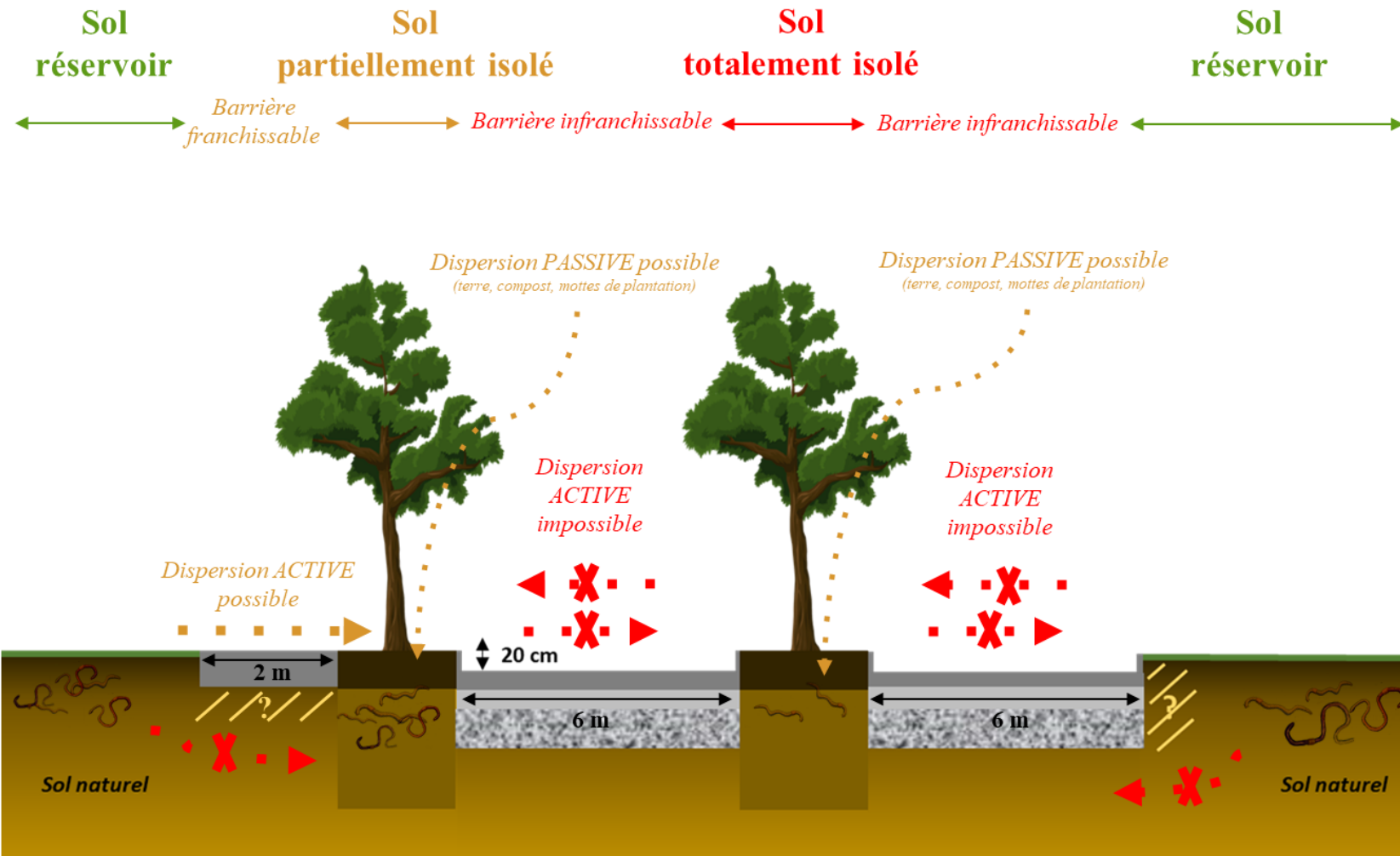


Figure 8 - Coupe schématique des modalités d'isolement de l'étude et hypothèses de dispersion lombricienne associées (exemple de coupe transversale d'une voirie avec des Anthrosoles reconstitués)

## 5. Objectifs de la thèse

L'ingénierie pédologique se développe pour assurer le développement de la végétation en ville malgré les contraintes inhérentes au milieu urbain (Li et al., 2013; Deeb et al., 2020). Des solutions plus ou moins élaborées sont mises en place selon les contextes des projets d'aménagement pour principalement assurer la fonction de fertilité des sols support de végétation (Marié and Rossignol, 1997; Strohmayer, 1999; Vidal-Beaudet and Rossignol, 2018a). Les pratiques d'ingénierie pédologique sont de plus relativement récentes et souvent empiriques (Vidal-Beaudet and Rossignol, 2018a), si bien que peu de suivis scientifiques relatent de la fertilité physique, chimique et biologique de sols issus de l'ingénierie pédologique plusieurs années après leur mise en œuvre. Parmi la biodiversité du sol, les vers de terre constituent des acteurs majeurs des écosystèmes sols dont ils contribuent largement à la fertilité et aux services écosystémiques (Blouin et al., 2013; Sharma et al., 2017). Cependant, les études scientifiques sont encore peu nombreuses en milieu urbain en lien avec les contraintes d'étude des communautés lombriciennes dans les sols urbains, particulièrement hétérogènes et sous l'influence de multiples facteurs naturels et anthropiques (e.g., types de sols, physico-chimie, historique, usage, gestion ; Xie et al., 2018 ; Tóth et al., 2020). Malgré la volonté grandissante de lutter contre l'artificialisation des sols, la perte de biodiversité et de fonctionnalité dans les sols, le manque de connaissances scientifiques ne permet pas à l'heure actuelle de proposer des préconisations techniques en faveur de la faune du sol dans le cadre de projets d'aménagement. De plus, les espèces à faible capacité de dispersion telles que les vers de terre sont *a priori* impactés à la fragmentation du paysage urbain par des barrières anthropiques plus ou moins franchissables (chemins, routes, trottoirs...) qui isolent les habitats les uns des autres. Cependant, les impacts l'isolement des sols par rapport à de potentiels réservoirs sur les communautés lombriciennes ne sont pas bien définis (Dupont et al., 2017).

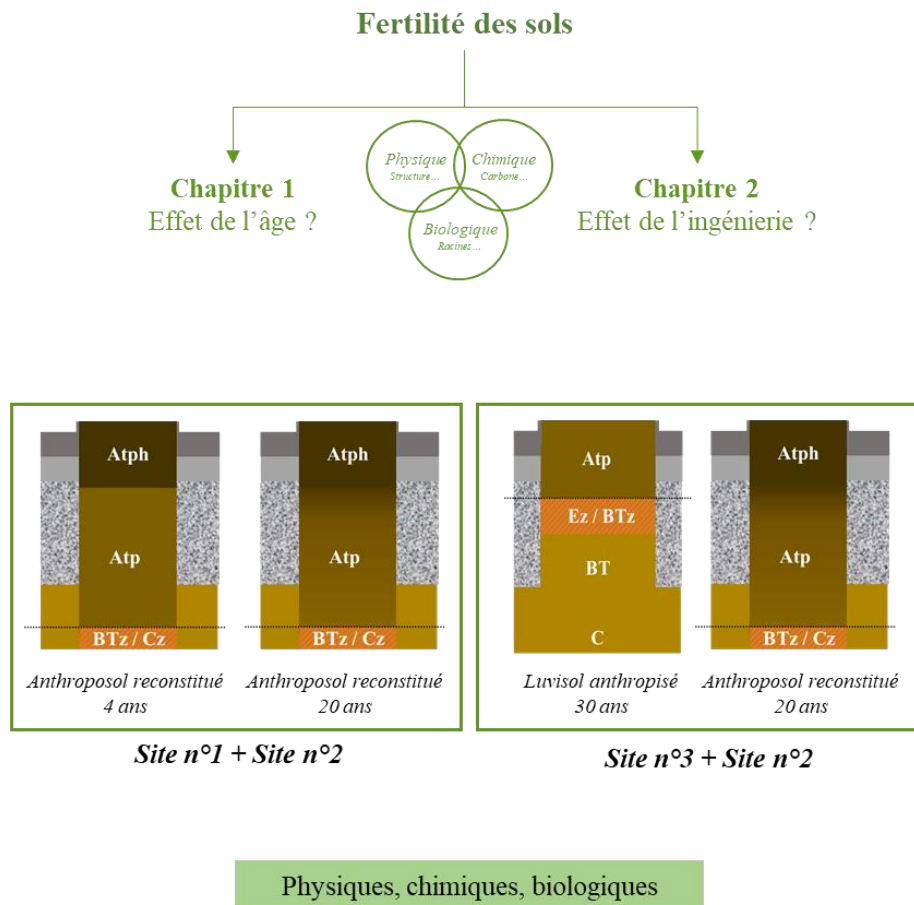
Le premier objectif de la thèse a été de caractériser les composantes physiques, chimiques et biologiques de la fertilité de sols issus de l'ingénierie pédologique. Pour ce faire, nous avons mesuré dans chaque horizon des paramètres physico-chimiques classiquement préconisés dans le cadre d'opérations d'aménagement pour la caractérisation de matériaux pédologiques (Tableau 3), auxquels nous avons ajouté la mesure de paramètres biologiques (biomasse et activité microbiennes, indices nématofauniques, pourcentages de faciès de bioturbation et de racines dans le profil). Dans un premier temps, nous avons caractérisé la fertilité d'Anthrosols reconstitués suivant le même processus d'ingénierie pédologique. Nous avons sélectionné des Anthrosols de deux âges différents, respectivement 4 ans et 20 ans, afin d'appréhender l'effet de la temporalité sur leurs propriétés (Chapitre 1). Dans un second temps, nous avons caractérisé la fertilité de sols d'âges similaires mais issus de deux processus d'ingénierie pédologique distincts, menant à des sols de degrés d'anthropisation différents (Tableau 2). Ainsi, nous avons caractérisé des Luvisols anthropisés de 30 ans et des Anthrosols reconstitués de 20 ans (Chapitre 2). Le deuxième objectif de la thèse a été de caractériser les communautés lombriciennes dans ces mêmes sols issus de l'ingénierie pédologique et également d'appréhender les impacts de leur isolement par rapport à de potentiels réservoirs. Nous avons d'abord comparé l'effet de deux degrés d'isolement contrastés sur les communautés lombriciennes dans des Anthrosols reconstitués de 4 et 20 ans (Chapitre 3). Puis, nous avons étudié l'effet croisé de ces deux mêmes degrés d'isolement à deux degrés d'anthropisation des sols (i.e., Luvisols anthropisés et Anthrosols reconstitués ; Chapitre 4). Des Luvisols naturels témoins à proximité des sites d'étude ont également été échantillonnés dans le cadre de ces deux derniers chapitres. Un schéma conceptuel de la thèse est présenté en Figure 9.

Axes de recherche

Chapitres

Modalités étudiées

Propriétés étudiées

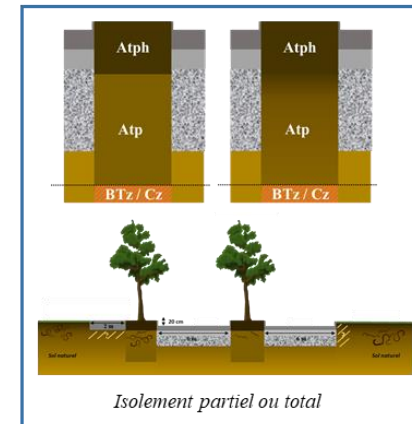


**Communautés lombriciennes**

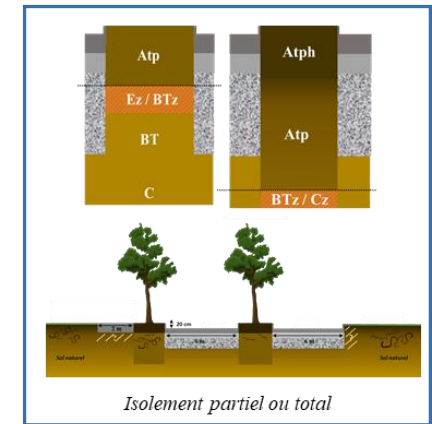
**Chapitre 3**  
Effet de l'âge ?  
X  
Effet de l'isolement ?



**Chapitre 4**  
Effet de l'ingénierie ?  
X  
Effet de l'isolement ?



**Site n°1 + Site n°2**



**Site n°3 + Site n°2**

Abondance et biomasse totales et fonctionnelles,  
richesse spécifique, occurrence

Figure 9 – Schéma conceptuel de la thèse



**CHAPITRE 1 - Caractérisation de la fertilité physique, chimique  
et biologique d'Anthroposols reconstitués de 4 et 20 ans**

## **Objectifs de l'étude**

Le but de cette étude était de caractériser la fertilité physique, chimique et biologique de sols reconstitués à deux périodes différentes selon un processus similaire d'ingénierie élaborée, impliquant une reconstitution de l'ensemble du profil de sol, dans un même contexte pédoclimatique et urbain.

## **Méthodologie**

Nous avons travaillé sur des Anthrosols reconstitués d'accompagnement de voirie de 4 ans et de 20 ans. Chacun des sols était constitué d'un horizon profond et épais de 1.20 m de terre végétale décapée (horizon organo-minéral), d'un horizon de surface de 30 cm de mélange terre-compost, et d'un mulch organique en surface (broyat de déchets verts pour le sol de 4 ans, écorces de pin pour le sol de 20 ans). Neuf fosses pédologiques ont été observées par type de sol afin de décrire les différents horizons et de quantifier les pourcentages de racines et de biostructures lombriciennes. Pour trois fosses de chacun des types de sol, une quarantaine de paramètres physiques, chimiques et biologiques ont été mesurés sur chacun des horizons pour caractériser les différentes composantes de la fertilité du sol.

## **Principaux résultats**

La fertilité était très contrastée entre les horizons d'un même type de sol et entre les horizons équivalents de types de sols différents. L'horizon de mélange terre-compost des sols de 4 ans présentait les teneurs les plus élevées en éléments nutritifs pour les plantes et les organismes du sol, ainsi que les pourcentages de biostructures lombriciennes les plus élevés. L'horizon sous-jacent non amendé présentait une transition nette avec le mélange terre-compost, une densité apparente plus élevée, deux fois moins de biostructures et une fertilité chimique nettement inférieure. En ce

qui concerne les anthroposols de 20 ans, un léger contraste entre ces deux horizons était encore visible en termes de fertilité chimique. Une litière épaisse s'était développée en surface ainsi qu'un réseau racinaire à hauteur de 40% dans la litière et le mélange terre-compost. La richesse nématofaunique et le pourcentage de biostructures du mélange terre-compost de 20 ans étaient en moyenne 1.8 fois inférieurs à celui de 4 ans, ainsi qu'un C/N 1.7 fois plus élevé traduisant une mauvaise dégradation de la matière organique.

**Title: Characterisation of the physical, chemical, and biological fertility of 4-year-old and 20-year-old reconstructed Anthrosoils in urban areas**

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## **Abstract**

The fertility of soils in urban areas is mostly degraded by anthropogenic activities which can highly constraint the growth of urban trees. To deal with these constraints, one solution developed by soil engineering is to reconstruct deep soils by superposing several layers of stripped topsoil with compost incorporation in the surface horizon. They are called reconstructed Anthrosoils. However, this process is rarely used and little knowledge is available about the physical, chemical, and biological components of Anthrosoils' fertility several years after their implementation in urban areas. To fill this gap, the objective of our study was to characterise the fertility of two similar Anthrosoils contrasted in age. Thus, we selected 4- and 20-year-old linear roadside Anthrosoils reconstructed following the same engineering process in the Paris region. Almost 40 soil properties were measured from samples analyses and soil profile observations (percentages of roots and earthworm biostructures in particular). We concluded a very significant and positive effect of compost incorporation on soil chemical fertility, particularly in the 4-year-old Anthrosoil. It provided a direct source of nutrients for plants but also trophic for earthworms favouring their bioturbation activity and the structuration of the soil. However, the underlying non-amended horizon presented higher apparent density and two times fewer earthworm biostructures. Regarding 20-year-old Anthrosoils, a light contrast between these two horizons was still visible in terms of fertility. Besides, the mulch of pine barks added on the surface and the development of planted ivy cover seemed to negatively impact biological fertility. Overall, soil fertility in its three components is a dynamic function that evolves at the scale of the soil profile and highly depends on engineering processes choices.

**Keywords: soil structure, urban soils, pedological engineering, aggregation, bioturbation, earthworm casts**

## 1. Introduction

City dwellers' well-being partly depends on elements of nature included in urban environments which provide numerous benefits such as mitigation of the urban heat island effect, improvement of air quality notably allowed by the presence of trees (Livesley et al., 2016; Bodnaruk et al., 2017). However, urban trees may be subject to severe growth constraints due to poor physical, chemical, and biological soil fertility (Watson et al., 2014; Morel et al., 2017). Indeed, soil fertility can be altered notably by severe compaction or poor nutrient availability due to anthropic activities (e.g., construction, repeated trampling, Craul, 1985; Lehmann and Stahr, 2007). To cope with these constraints, soil engineering allows constructing deep fertile soils on the model of natural soils (i.e., Anthrosoils; Rossignol et al., 2008) by superposing different types of horizons made of soil material meeting physicochemical fertility requirements (Damas and Coulon, 2016; Séré et al., 2017). For example, the amount of organic matter in the surface horizon is recommended to be superior to 3% to optimise soil fertility and root development (AFNOR, 2002; Unep, 2012). Thus, several physicochemical properties (e.g., pH, nitrogen, phosphorus, exchangeable cations) of materials intended to support vegetation are characterised prior to the soil construction and are amended if necessary (AFNOR, 2002; Vidal-Beaudet et al., 2018). However, biological properties of soil materials are rarely characterised in the context of urban projects whereas they provide complementary information about soil functioning and fertility (Chaussod, 1996; Abbott and Murphy, 2007). For instance, different indexes have been developed from the analysis of nematofauna and allow to characterize food webs or the dominant pathways of organic matter mineralisation (Bongers and Bongers, 1998; Ferris et al., 2001).

Soil organisms, earthworms in particular, are key actors of soil fertility because of their widely-known contributions to the physical, chemical, and biological components of soil fertility (Lee, 1985; Edwards and Bohlen, 1996; Lavelle and Spain, 2001). Thanks to their bioturbation activity

(i.e., biological reworking of soils; Meysman et al., 2006), they create casts and galleries which improve, among others, soil structure, water retention, and nutrient availability (Görres et al., 2001; Bastardie et al., 2005; Singh et al., 2016). These biostructures can be observed on clods or soil profiles which allows to visually assess soil functioning and to identify potential constraining areas for root growth (Roger-Estrade et al., 2004; Piron et al., 2012, 2017). These methodologies were established in the first intention for agricultural soils, so that in newly implemented engineered soils in urban areas, little is known about structuration by earthworms with regard to the disturbances they undergo during the soil construction (Jangorzo et al., 2013; Maréchal et al., 2021). Besides, investigating urban soils can be very constraining due to their wide diversity (difficulty in finding replicas and historical knowledge), the presence of numerous buried networks, road traffic, or administrative requests to get the authorisation to open pedological pits.

The objective of this study was to characterise soil fertility of two reconstructed Anthrosoils similarly engineered respectively 4 and 20 years ago with a soil-compost horizon on the surface, superposing a deep non-amended horizon of topsoil. We hypothesised significant beneficial effects of compost-amendment on soil physical, chemical and biological fertility of both types of Anthrosoils. Soil fertility was assessed from the analyses of almost 40 physical, chemical, and biological parameters, including the quantification of earthworm biostructures on soil profiles.

## **2. Materials and methods**

### ***2.1. Study sites***

The study sites corresponded to linear roadside soils located in the Paris region (France) and were divided between Palaiseau (48° 71' 42'' N; 2° 19' 64'' E) and Saint-Quentin-en-Yvelines (48° 75' 75'' N; 2° 07' 25'' E) cities on the Plateau of Saclay. The substratum corresponds to thick

loess deposits (> 3m) (Mégnyien, 1989). The climate is temperate, with a mean annual precipitation of 563 mm and a mean annual temperature of 11.2°C, a monthly minimum of -2.3°C and a monthly maximum of 26.4 °C (data from Météo France, 2008-2018 Trappes station).

We selected (i) a 4-year-old reconstructed Anthrosoil (4RA) along the Gaspar Monge Boulevard in Palaiseau, and (ii) a 20-year-old reconstructed Anthrosoil (20RA) along the Europe Avenue in Saint-Quentin-en-Yvelines. Briefly, the cumulated length of 4RA and 20RA sections were 970 m and 790 m, respectively. 4RA were 1.8-metre-wide, planted with 3 main tree species every 4 metres (*Quercus robur*, *Carpinus betulus*, *Fagus sylvatica*,). 20RA were also 1.8-metre-wide and only one tree species was planted in 20RA every 7 metres (*Tilia pallida*). More details are available on Table S1. Each Anthrosoil was reconstructed following a similar engineering process prescribed by the same company. Firstly, trenches of 150 cm deep were excavated in urban soils present before the reconstruction. Secondly, 125 cm of stripped and stockpiled silty topsoil (organo-mineral material) was added in the trenches as a horizon of topsoil (HT). In both 4RA and 20RA, the topsoil used for the soil reconstruction was the result of stripping in dry conditions of 30 cm of soil in the right-of-way of the development project. Stripped topsoil was stockpiled for 2 years in stocks over 2.5 metres high. Thirdly, a 25 cm-thick surface horizon of topsoil mixed with green-waste compost was implemented (HTC). Finally, an additional layer of 5 cm of organic mulch (respectively green waste shreds for 4RA, and pine barks for 20RA) and a permanent plant cover were implemented on the surface as a litter horizon (HL). The succession of soil horizons was confirmed by the observation of nine pedological pits (100 cm x 150 cm x 110 cm, length × width × depth) were dug 4RA and nine in 20RA. Each pit was located in the middle of an inter-tree. Due to constraints of opening pedological pits in urban areas (e.g., location of buried networks, authorisation requests), 4RA pits were observed in July 2019 and 20RA pits in September 2019.



Soil horizons were described and identified according to the French pedological reference system (Table 1; Baize and Girard, 2008) and associated to a type of horizon previously described.

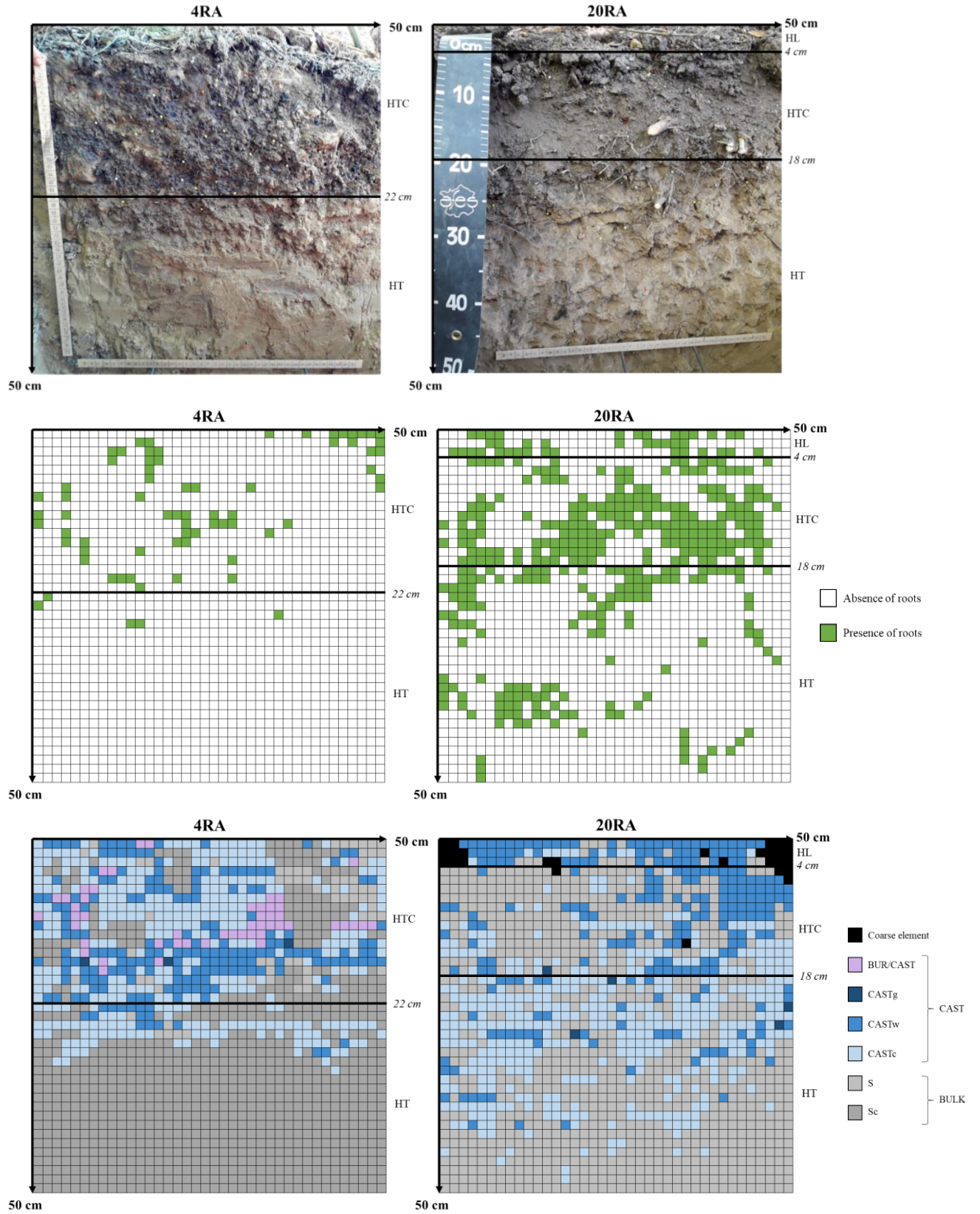
**Table 4** Description of soil horizons from 0 to 50 cm deep per type of Anthrosoil based on the observation of nine 4-year-old reconstructed Anthrosoils and nine 20-year-old reconstructed Anthrosoils pedological pits.

4RA (n = 9)						20RA (n = 9)					
Horizon	Code	Mean depth (cm ± SD)	Colour (Munsell colour chart)	Type of horizon (Baize & Girard, 2008)	Main characteristics	Horizon	Code	Mean depth (cm ± SD)	Colour (Munsell colour chart)	Type of horizon (Baize & Girard, 2008)	Main characteristics
-	-	-	-	-	-	<b>HL</b> Litter	20RA-HL	0 - 8 (± 3.3)	10YR 3/1: Very dark grey	<b>O:</b> Very organic litter with mulch of pine barks	Crumbly structure Loamy Very organic Weakly compact
<b>HTC</b> Topsoil + Compost	4RA-HTC	0 - 20 (± 4.8)	10YR 4/3: Brown	<b>AtpH:</b> Stripped, transported and compost-amended topsoil	Crumbly structure Loamy Organo-mineral Compact	<b>HTC</b> Topsoil + Compost	20RA-HTC	8 - 22 (± 4.8)	10YR 4/2: Dark greyish brown	<b>AtpH:</b> Stripped, transported and compost-amended topsoil	Crumbly to polyhedral structure Loamy Organo-mineral Weakly compact
<b>HT</b> Topsoil	4RA-HT	20 - 47 (± 4.8)	10YR 6/4: Light yellowish brown	<b>Atp:</b> Stripped and transported topsoil	Polyhedral structure Loamy Organo-mineral Very compact	<b>HT</b> Topsoil	20RA-HT	22 - 49 (± 6.6)	10YR 5/6: Yellowish brown	<b>Atp:</b> Stripped and transported topsoil	Polyhedral to massive structure Loamy Mineral Compact

## 2.2. Mapping of soil-structure patterns and roots on soil profiles

In that study, analyses were focused on the 50 first cm of the soil profile because it concentrates the main part of organic carbon, mineral nutrients, and biological activity such as roots and earthworm bioturbation which are visible on a soil profile (Henin et al., 1969). Thus, mapping of

roots and earthworm biostructures (casts and burrows) were achieved following protocols adapted respectively from Tardieu and Manichon (1987) and Piron et al. (2012). Thus, soil profiles were carefully prepared with a knife to reveal the soil aggregates in their original aspect. Then, a grid frame (1.2 cm x 1.2 cm) was attached against the soil profiles (Fig.1) and pictures of the grid frame were taken to allow image analysis and give a precise transcription of observed roots and earthworm biostructures. Then, a digital mapping was achieved by associating each cell of the grid (1.44 cm<sup>2</sup>) to the presence or absence of a root, and to the major earthworm biostructure (i.e., representing more than 50% of the total surface of a cell) following a typology adapted from Piron et al. (2012) which distinguishes different soil-structure patterns: earthworm burrows (BUR), fresh casts (CASTg), aged casts (CASTw), degraded casts (CASTC), compacted bulk soil (S) or not (Sc). The typology used in that study is presented in Table S2. In total, 1482 cells were filled for each pit, covering 2134 cm<sup>2</sup> of soil profile. Examples of root maps and soil-structure pattern map are given in Fig.1. Thereafter, % of soil-structure patterns were calculated (i) on the whole 50 cm x 50 cm square described; (ii) per type of horizon defined on the field; (iii) every 5 cm regardless of soil horizons.



**Fig. 1.** Examples of (a) soil profiles and associated (b) root maps and (c) soil-structure patterns maps (on the left = 4RA; on the right = 20RA). HL = horizon of litter, HTC = horizon of topsoil and compost, HT = horizon of topsoil.

### *2.3. Soil sampling and laboratory analyses*

Apparent density ( $D_a$ ;  $\text{g}\cdot\text{cm}^{-3}$ ) was measured in each horizon of each pit according to the NF X31-501 method (AFNOR, 1992): one calibrated cylinder of 100  $\text{cm}^3$  of soil was sampled at each site in the soil profile. The sampling was then dried at  $105^\circ\text{C}$  and weighed ( $\pm 10$  cg).

Three pits among the nine 4RA pits and three pits among the nine 20RA pits were randomly selected to achieve physical, chemical, and biological laboratory analyses. Soil samples were separately taken from each horizon between 0 and 50 cm-deep of each selected pit. For each horizon, the sample was taken from a pool of 10 sub-samples. As 2 horizons were observed in 4RA and 3 were observed in 20RA, 5 types of samples were analysed: (i) 4RA-HTC ( $n = 3$ ), (ii) 4RA-HT ( $n = 3$ ), (iii) 20RA-HL ( $n = 3$ ), (iv) 20RA-HTC ( $n = 3$ ), and (v) 20RA-HT ( $n = 3$ ).

Regarding physical and chemical properties, each sample of the 5 types detailed above was analysed for particle-size distribution (clay  $<2\ \mu\text{m}$ ; silt  $2<\mu\text{m}<50$ ; sand  $50<\mu\text{m}<200$ , %; NFX 31-107 without decarbonatation; AFNOR, 2003), electrical conductivity (EC;  $\text{mS}\cdot\text{cm}^{-1}$ ; NF EN 13038; AFNOR, 2012), pH (pH; NF ISO 10390; AFNOR, 2005), organic carbon ( $C_{\text{org}}$ ; %; NF ISO 14235; AFNOR, 1998a), total nitrogen ( $N_{\text{tot}}$ ; %; NF ISO 13878; AFNOR, 1998b), phosphorus ( $\text{P}_2\text{O}_5$ ;  $\text{mg}\cdot\text{kg}^{-1}$ ; NF ISO 11263; AFNOR, 1995), cation exchange capacity (CEC;  $\text{meq}\cdot 100\text{g}^{-1}$ ; NF X 31.130; AFNOR, 1999), exchangeable cations ( $\text{CaO}$ ,  $\text{K}_2\text{O}$ ,  $\text{MgO}$ ,  $\text{Na}_2\text{O}$ ;  $\text{mg}\cdot\text{kg}^{-1}$ ; NF X 31.108; AFNOR, 2002), and the calcium saturation rate was calculated ( $\text{Ca}/\text{CEC}$ ). Metal trace elements (Cd, Cr, Cu, Hg, Ni, Pb, Zn; ppm) have been analysed in every horizon by portable X-ray fluorescence method.

In detail, measurements have been carried out on each sample of the 5 types sieved to 2 mm through a Bruker portable XRF. In the rest of the study, Cd and Hg will not be detailed as they were not detected in any sample.

Complementary analyses were achieved in surface horizons to precise their characterisation. In addition, for 4RA-HTC, 20RA-HL, and 20RA-HTC, fractions between 0 and 50  $\mu\text{m}$  and between 50 and 2000  $\mu\text{m}$  were measured for Corg ( $C_{0:50}$  and  $C_{50:2000}$ ; % of Corg; NF ISO 14235; AFNOR, 1998a), and Ntot ( $N_{0:50}$  and  $N_{50:2000}$ ; % of Ntot; NF ISO 13878; AFNOR, 1998b). Regarding biological properties, samples were analysed for microbial biomass (MB;  $\text{mg}\cdot\text{kg}^{-1}$ ; NF ISO 14240-2; AFNOR, 2011), and for substrate-induced respiration (SIR;  $\mu\text{g}\cdot\text{C}\cdot\text{CO}_2\text{ g}^{-1}\text{ h}^{-1}$ ), denitrification rate (DEA;  $\mu\text{g}\cdot\text{N}\cdot\text{N}_2\text{O g}^{-1}\text{ h}^{-1}$ ), and nitrification rate (NEA;  $\mu\text{g N}\cdot\text{NO}_2+\text{NO}_3\text{ g}^{-1}\text{ h}^{-1}$ ). For SIR, DEA, and NEA analyses, soil samples were sieved to 2 mm and stored at  $-18^\circ\text{C}$  until microbial analyses were achieved. SIR was measured as  $\text{CO}_2$  production (Anderson and Domsch, 1978). Soil samples (5 g) were placed in an airtight flask in 21%  $\text{O}_2$  atmosphere, containing 0.5 ml of a nutritive solution including glucose (1.2 mg of C-glucose  $\text{g}^{-1}$  of dried soil) then incubated at  $28^\circ\text{C}$ .  $\text{CO}_2$  concentrations were measured each hour for 5 h and the slope of the linear regression was used to estimate aerobic respiration as the  $\text{CO}_2$  produced ( $\text{g}^{-1}\text{ h}^{-1}$ ). DEA was measured in soil using a method adapted from Dassonville et al. (2011). First 5 g of soil sample was placed in 150-ml airtight plasma-flasks sealed with rubber stoppers. In each flask, air was removed and replaced with a He/ $\text{C}_2\text{H}_2$  mixture (90: 10, v/v) to create anoxic conditions and inhibit  $\text{N}_2\text{O}$ -reductase. A nutritive solution (0.5 ml) containing glucose (0.5 mg of C-glucose  $\text{g}^{-1}$  of dried soil), glutamic acid (0.5 mg of C-glutamic acid  $\text{g}^{-1}$  of dried soil) and potassium nitrate (50  $\mu\text{g}$  of N- $\text{KNO}_3\text{ g}^{-1}$  of dried soil) was added to the soil. The amount of  $\text{N}_2\text{O}$  during incubation at  $28^\circ\text{C}$  was measured each hour for 6 to 8 h. The slope of the linear regression was used to estimate anaerobic respiration (denitrification) as the  $\text{N}_2\text{O}$  produced ( $\text{g}^{-1}\text{ h}^{-1}$ ). NEA was determined according to a modified

method described by Dassonville et al. (2011). Soil samples were incubated under aerobic conditions for 10h with 30 mL of  $(\text{NH}_4)_2\text{SO}_4$  ( $1.25 \text{ mg N.L}^{-1}$ ) using a continuous shaking (140 rpm,  $28^\circ\text{C}$ ).  $\text{NO}_2^-$  and  $\text{NO}_3^-$  production rates were measured after 2, 4, 8 and 10 hr by ionic chromatography (DX120; Dionex, Salt Lake City, UT, USA). Nematofauna analyses were carried out on 4RA-HTC ( $n = 3$ ) and 20RA-HTC ( $n = 3$ ) and achieved by ELISOL Environment company to compare nematode communities in a soil-compost horizon of two different sites (NF EN ISO 23611-4; AFNOR, 2011b). Nematofauna was characterised by the abundances of the different functional groups (opportunistic bacterial-feeders, bacterivores, other bacterial-feeders, fungal-feeders, facultative plant feeders, obligate plant-feeders, omnivores-predators), taxonomic richness: nematode family number (N) and indices: Enrichment Index (EI), Structure Index (SI), Nematode Channel Ratio (NCR; Bongers and Bongers, 1998; Ferris et al., 2001), and Shannon diversity ( $H'$ ). EI varies from 0 to 100 and increases with the availability of nutrients. SI varies from 0 to 100 and increases with the complexity of the food web and the stability of the environment. NCR indicates the dominant pathways of mineralization of organic matter (bacterial or fungal). The balance between the two is sought.  $H'$  increases with the diversity of organisms in the sample.

#### ***2.4. Statistical analyses***

A Shapiro-Wilk normality test was performed to verify the normality of all the data prior further analyses. All statistical analyses were performed on R software (R Core Team, 2019). Significance was evaluated in all cases at  $P < 0.05$ .

A principal component analysis (PCA) was performed on data collected from soil profile mapping to assess the contrasts of apparent density, root and soil-structure patterns percentages between soil types and soil horizons. Four different PCAs were performed on data from samples

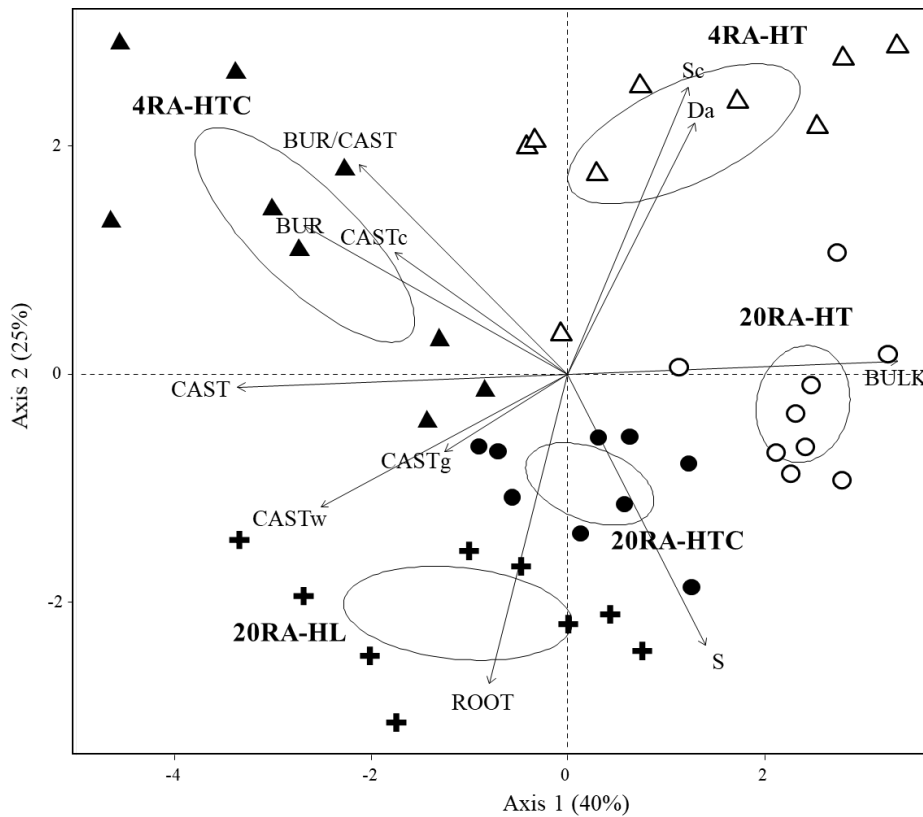
analyses to compare soil properties between (1) 4RA-HTC and 4RA-HT, (2) 20RA-HL, 20RA-HTC, and 20RA-HT (i.e., comparisons of horizons of a same soil type), (3) 4RA-HTC and 20RA-HTC, (4) 4RA-HT and 20RA-HT (i.e., comparisons of equivalent horizons of two different soil types). In addition, another PCA was performed on abundances of the different functional groups of nematodes in HTC. The variables used for each PCA corresponded to variables common to each type of horizon or each type of soil. The variables associated with soil texture (i.e., clay, silt, sand) were excluded from the analyses because the soil material used for soil construction was similar in both types of soil, and soil engineering do not impact this parameter.

### 3. Results

Regarding 4RA soil profiles, CAST decreased between HTC and HT (respectively  $68.7 \pm 11.0\%$  and  $36.1 \pm 15.2\%$ ; Fig. 2, Fig S1, Table S3). Indeed, HTC was characterised by CASTc, BUR/CAST, and BUR patterns whereas HT showed higher values of Sc pattern associated with higher Da values (Fig. 2, Table S3). HTC was also characterised by higher values of CEC,  $P_2O_5$ , Corg, Ntot, C:N,  $K_2O$ ,  $Na_2O$ , Pb, and Zn than HT (Fig. 3a, Table S4). Regarding 20RA soil profiles, CAST decreased along soil profiles (respectively  $56.7 \pm 15.8\%$ ,  $46.8 \pm 10.4\%$ ,  $23.4 \pm 7.7\%$ , in HL, HTC and HT; Fig. 2, Fig S1, Table S3). HL showed high percentages of ROOT and CASTw whereas HTC and HT were respectively characterised by S and BULK patterns (Fig.2, Table S3). HL presented higher values of CEC,  $P_2O_5$ , Corg, Ntot,  $K_2O$ , MgO, Zn, and Cu which decreased along soil profiles (Fig. 3b, Table S4).

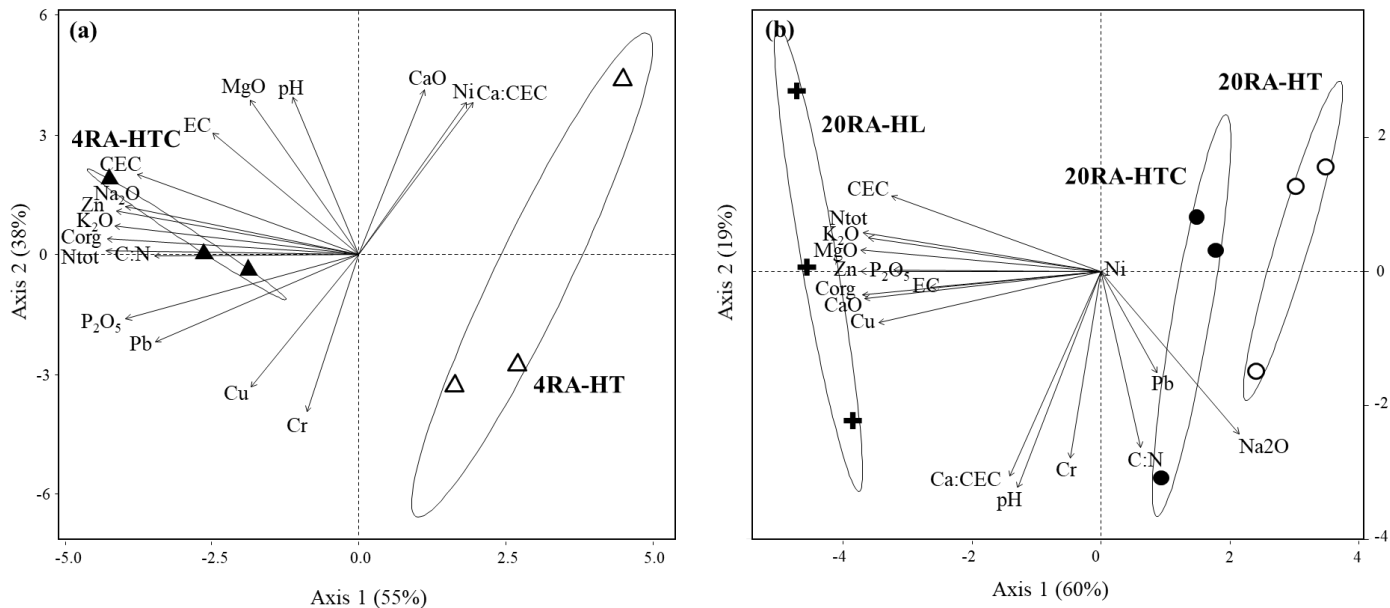
Regarding HTC horizons, 4RA showed higher values of pH, CEC,  $P_2O_5$ ,  $C_{0:50}$ , Ntot,  $N_{50:2000}$ , CaO,  $K_2O$ , MgO, H', N, and abundance of bacterivore-other nematodes (i.e., bacterial-feeder nematodes) than 20RA (Fig. 4a, Fig. S2). By contrast, 20RA was characterised by higher values of  $C_{50:2000}$ ,  $N_{0:50}$ , C:N,  $Na_2O$ , Pb, Cu, MB, SIR, DEA, abundances of phytoparasitic and facultative-

phytophagous nematodes (i.e., plant-feeders nematodes; Fig. 4a, Fig.S2). Regarding HT horizons, 20RA showed higher values of Corg, Ntot, C:N, Pb, Zn, and Cr (Fig.4b).

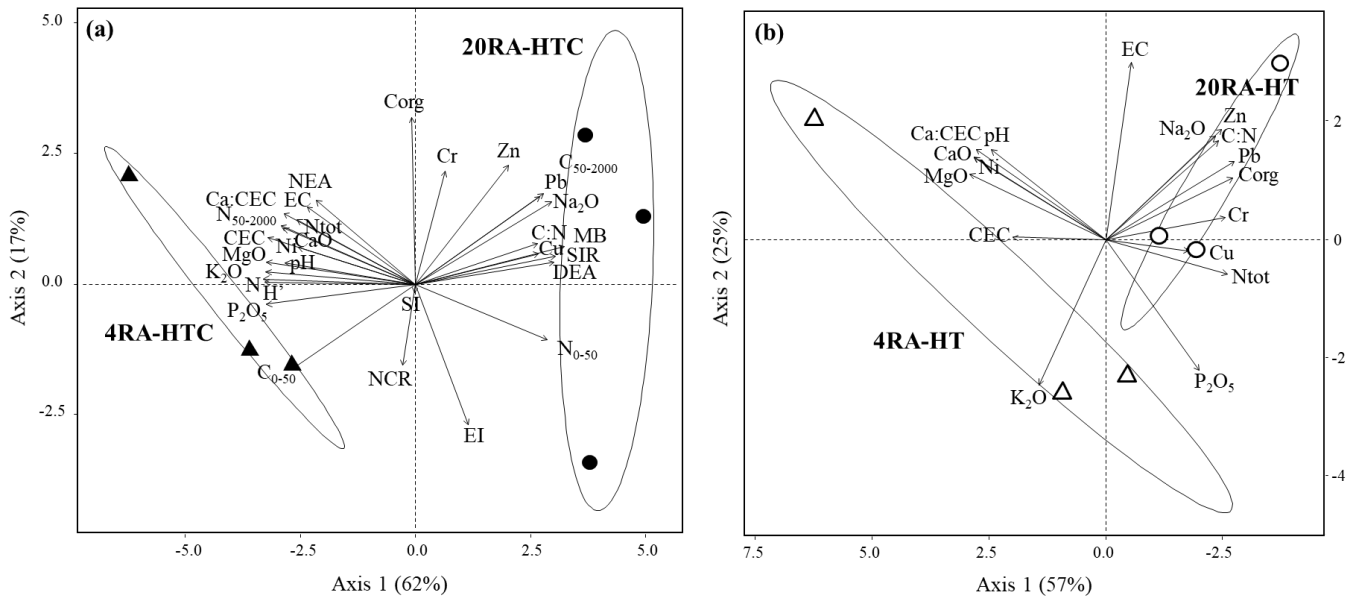


**Fig. 2** Principal component analysis using values of apparent density (Da), percentages of roots (ROOT) and soil-structure patterns measured on nine 4RA soil profiles, and nine 20RA soil profiles (see Table S2 for legend). Arrows correspond to variables associated to the first two axes. ▲ = horizon HTC of a 4RA pit; △ = horizon HT of a 4RA pit; + = horizon HL of a 20RA pit; ● = horizon HTC a 20RA pit; ○ = horizon HT of a 20RA pit. 95% confidence ellipses correspond to soil horizons per soil type. See Table 1 and Table S2 for detailed legends.





**Fig. 3** Principal component analysis using physical, chemical, and biological properties measured on samples from (a) three 4RA pits and (b) three 20RA pits. Arrows correspond to variables associated to the first two axes. ▲ = horizon HTC of a 4RA pit; △ = horizon HT of a 4RA pit; + = horizon HL of a 20RA pit; ● = horizon HTC a 20RA pit; ○ = horizon HT of a 20RA pit (see Table 1 for detailed legend). 95% confidence ellipses correspond to soil horizons per soil type. EC = electrical conductivity ( $\text{mS}\cdot\text{cm}^{-1}$ ); pH (-); CEC = cation exchangeable capacity ( $\text{meq}\cdot 100\text{g}^{-1}$ ); Ca:CEC =  $\text{Ca}^{2+}$  saturation rate;  $\text{P}_2\text{O}_5$ : phosphorus ( $\text{mg}\cdot\text{kg}^{-1}$ ); Corg = organic carbon (%); Ntot = total nitrogen; C:N = carbon on nitrogen ratio; CaO,  $\text{K}_2\text{O}$ , MgO,  $\text{Na}_2\text{O}$  = exchangeable cations ( $\text{mg}\cdot\text{kg}^{-1}$ ); Cr, Cu, Ni, Pb, Zn = metal trace elements (ppm).



**Fig. 4.** Principal component analysis using physical, chemical, and biological properties measured on samples of (a) HTC and (b) HT from three 4RA and three 20RA pits. Arrows correspond to variables associated to the first two axes. ▲ = horizon HTC of a 4RA pit; ● = horizon HT of a 4RA pit; △ = horizon HTC a 20RA pit; ○ = horizon HT of a 20RA pit. 95% confidence ellipses correspond to soil horizons per soil type. EC = electrical conductivity ( $\text{mS}\cdot\text{cm}^{-1}$ ); pH (-); CEC = cation exchangeable capacity ( $\text{meq}\cdot 100\text{g}^{-1}$ );  $\text{P}_2\text{O}_5$ : phosphorus ( $\text{mg}\cdot\text{kg}^{-1}$ ); Corg = organic carbon (%);  $\text{C}_{0:50}$  = 0-50  $\mu\text{m}$  carbon fraction (% of Corg);  $\text{C}_{50:2000}$  = 50-2000  $\mu\text{m}$  carbon fraction (% of Corg); Ntot = total nitrogen;  $\text{N}_{0:50}$  = 0-50  $\mu\text{m}$  nitrogen fraction (% of Ntot);  $\text{N}_{50:2000}$  = 50-2000  $\mu\text{m}$  nitrogen fraction (% of Ntot); C:N = carbon on nitrogen ratio; CaO,  $\text{K}_2\text{O}$ , MgO,  $\text{Na}_2\text{O}$  = exchangeable cations ( $\text{mg}\cdot\text{kg}^{-1}$ ); Cr, Cu, Ni, Pb, Zn = metal trace elements (ppm); MB = microbial biomass ( $\text{mg}\cdot\text{kg}^{-1}$ ); SIR = substrate-induced respiration ( $\mu\text{g}\cdot\text{C}\cdot\text{CO}_2\text{ h}^{-1}\text{ g}^{-1}$ ); DEA = denitrification rate ( $\mu\text{g}\cdot\text{N}\cdot\text{N}_2\text{O h}^{-1}\text{ g}^{-1}$ ); NEA = nitrification rate ( $\mu\text{g}\cdot\text{N}\cdot\text{NO}_2+\text{NO}_3\text{ h}^{-1}\text{ g}^{-1}$ ); EI = Enrichment Index (-); SI = Structure Index (-); NCR = Nematode Channel Ratio (-);  $H'$  = Shannon diversity index (-); N = nematode family number (-).

#### 4. Discussion

The chemical fertility of the compost-amended horizon in 4-year-old Anthrosoils is particularly enhanced compared to the non-amended horizon located deeper, and also compared to the compost-amended horizon in 20-year-old Anthrosoils. This was notably illustrated by higher trophic resources for plants and soil organisms (e.g.,  $P_2O_5$ , Corg, Ntot,  $K_2O$ ,  $Na_2O$ ). For example, Ntot was 2.7 times higher than in the 4-year-old non-amended horizon, and 1.5 times higher than in the 20-year-old amended horizon. The pH was also higher which is commonly associated with a compost incorporation in the short-term (Walker et al., 2004; Sarwar et al., 2010), but the alkalisation can also be explained as the dissolution of calcareous materials in the cement and concrete nearby Anthrosoils (Jim, 1998a; Park et al., 2010). Regarding 20-year-old Anthrosoils, the chemical fertility was also still enhanced in the amended horizon compared to the non-amended horizon in terms of Corg, Ntot,  $P_2O_5$ , CEC, and exchangeable cations. These results are consistent with numerous studies highlighting the beneficial effects of compost incorporation for soil chemical fertility (Chaoui et al., 2003; Vidal-Beaudet et al., 2018; Kranz et al., 2020). Besides, values of CEC,  $P_2O_5$ , Corg, and Ntot in 4- and 20-year old amended horizons were significantly higher than values identified in non-amended roadside soils, highlighting the efficiency of this engineering process for chemical fertility (Jim, 1998b, 1998a; Jimenez et al., 2013). The 4-year-old amended horizon also presented the highest proportion of earthworm casts and burrows (respectively 68.7% and 15.6%), exceeding values observed in a pasture soil with a similar approach (i.e., 22 to 42%, casts and burrows combined; Piron et al., 2006). This could be explained by abundant trophic resources which enhance earthworm bioturbation activities (Hughes et al., 1996; Pérès et al., 1998). Besides, organic matter from compost highly improves the stability of aggregates and protect them against disaggregation (Pagliai et al., 2004; Bronick and Lal, 2005).

By contrast, the total percentage of casts was almost divided per two in the subjacent non-amended horizon, with a clear transition between the two horizons. Indeed, trophic resources are less abundant (e.g., amount of Corg divided per 2), and apparent density is higher which could limit earthworm bioturbation activities in depth (Söchtig and Larink, 1992; Buck et al., 2000). Overall, the incorporation of compost into stripped topsoil is highly favourable for the fertility of engineered soils as it provides high amounts of nutrients for plants (i.e., higher chemical fertility) and trophic resources for earthworms which highly improve soil physical and chemical fertility through their bioturbation activities (Shipitalo and Le Bayon, 2004; Clause et al., 2015; Vos et al., 2019).

Regarding the 20-year-old amended horizon, earthworm biostructures were on average 1.5 times less abundant than in the 4-year-old amended horizon whereas bulk soil patterns were 1.7 times more abundant. Earthworm activity may be negatively impacted by the poor quality of the new resources entering the system, mainly from ivy cover and mulch of pine barks which can be unpalatable for earthworms (i.e., enriched in polyphenol; Hendriksen, 1990; Liebeke et al., 2015), explaining the low degradation of the litter and the lower number of biostructures. Besides, the high C:N ratio observed in the amended horizon (18.6 against 10.8 in the 4-year-old amended horizon) indicated a poor biological degradation of organic matter (Recous and Machet, 1999; Moritsuka et al., 2004) despite higher microbial biomass and activity by at least 1.9 times, but the reason remains unknown. Regarding nematofauna parameters, the Shannon index and the taxonomic richness were also lower in the 20-year-old amended horizon. This low diversity of nematodes is explained by a clear predominance of phytophagous nematodes, probably associated with by the presence of well-developed trees (*Tilia pallida*) and creepy ivy cover providing 1.5 times more roots, providing significant food resources for phytophagous nematodes (Wasilewska, 1989). However, enrichment and structure indexes in both Anthroposoils (respectively, EI = 63.0 and 70.7, SI = 48.5 and 52.0) indicated moderate nutrient availability and habitat stability (Ferris

et al., 2001), consistently with values observed in urban areas (Li et al., 2011). The nematode channel ratio was also similar in both sites (respectively, IVD = 68.3 and 58.7) and indicated a good balance between the fungal and the bacterial pathways of mineralisation (Ruess, 2003). Thus, the evolution of soil cover with time should be considered in the design of engineered soils as it can greatly impact biological communities (Byrne et al., 2008; Frouz, 2021). Regarding metal trace elements, a contrast was also observed between studied Anthro soils as Pb and Zn contents were higher in 20- than in 4-year-old Anthro soils (respectively +36% and +14%). This could be explained either by an accumulation with time due to road traffic as engineered soils studied are roadside soils, or by a natural higher geochemical background (Khalil et al., 2013). However, these values do not exceed limit values given by the standard for soil engineering (AFNOR, 2012).

## **5. Conclusion**

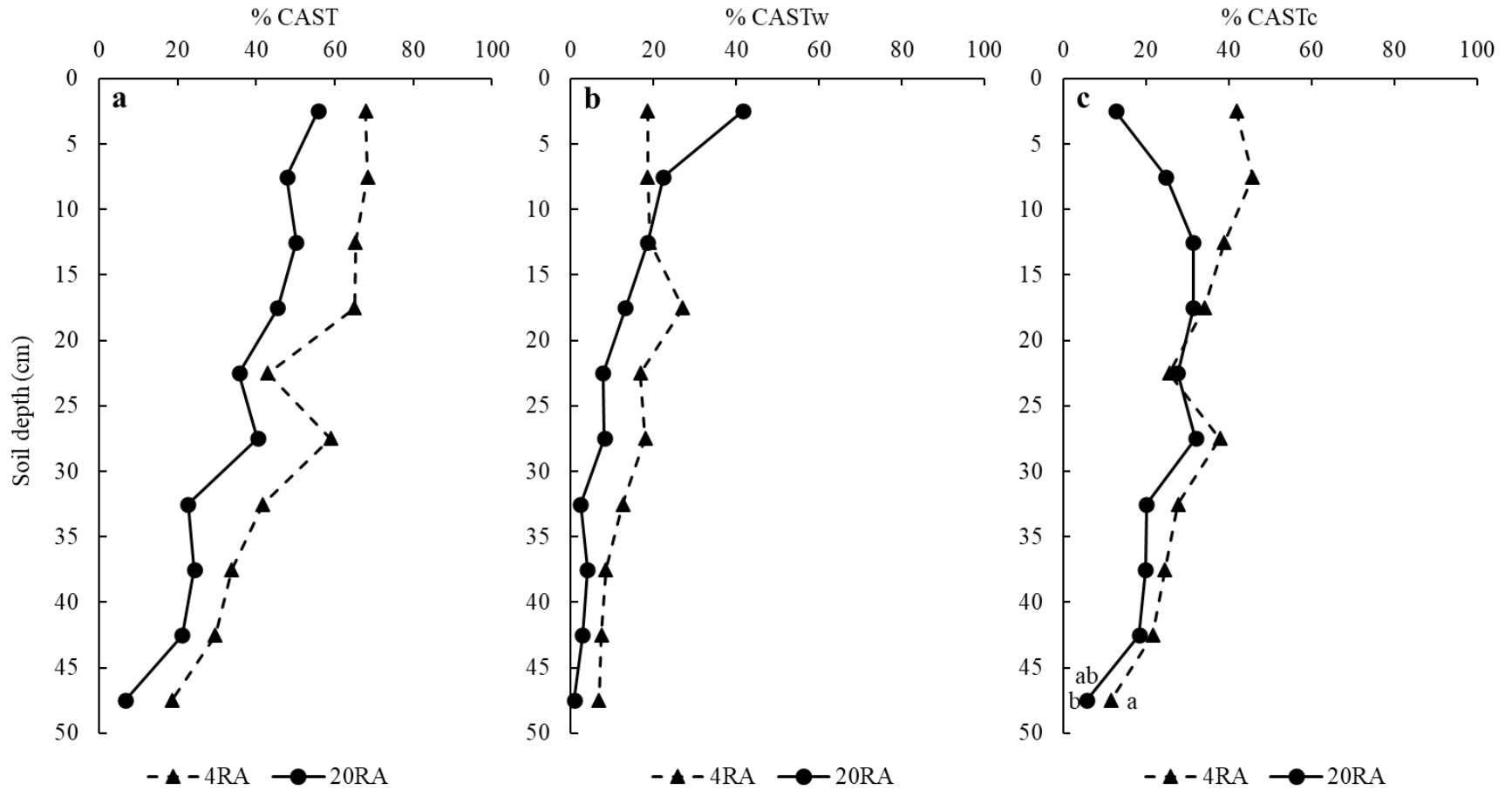
The fertility of reconstructed Anthro soils is particularly enhanced in recently reconstructed soils by compost incorporation in the surface horizon. It favours the availability of plant nutrients and trophic resources for earthworms, whose bioturbation activities also contribute to soil physical, chemical, and biological fertility. The positive effects of compost on chemical fertility are less contrasted but still visible in 20-year-old Anthro soils. The biological fertility is also dependent of the evolution of the vegetal cover and the type of mulch implemented which drive the quality of the new organic resources, and thus, the micro- and macrofauna development and activity. To better understand and prevent these changes, soil monitoring should be carried out with some chemical, physical, biological parameters, as it is recommended for an agricultural soil, and to potentially advise corrective measures such as re-amendment or soil work.

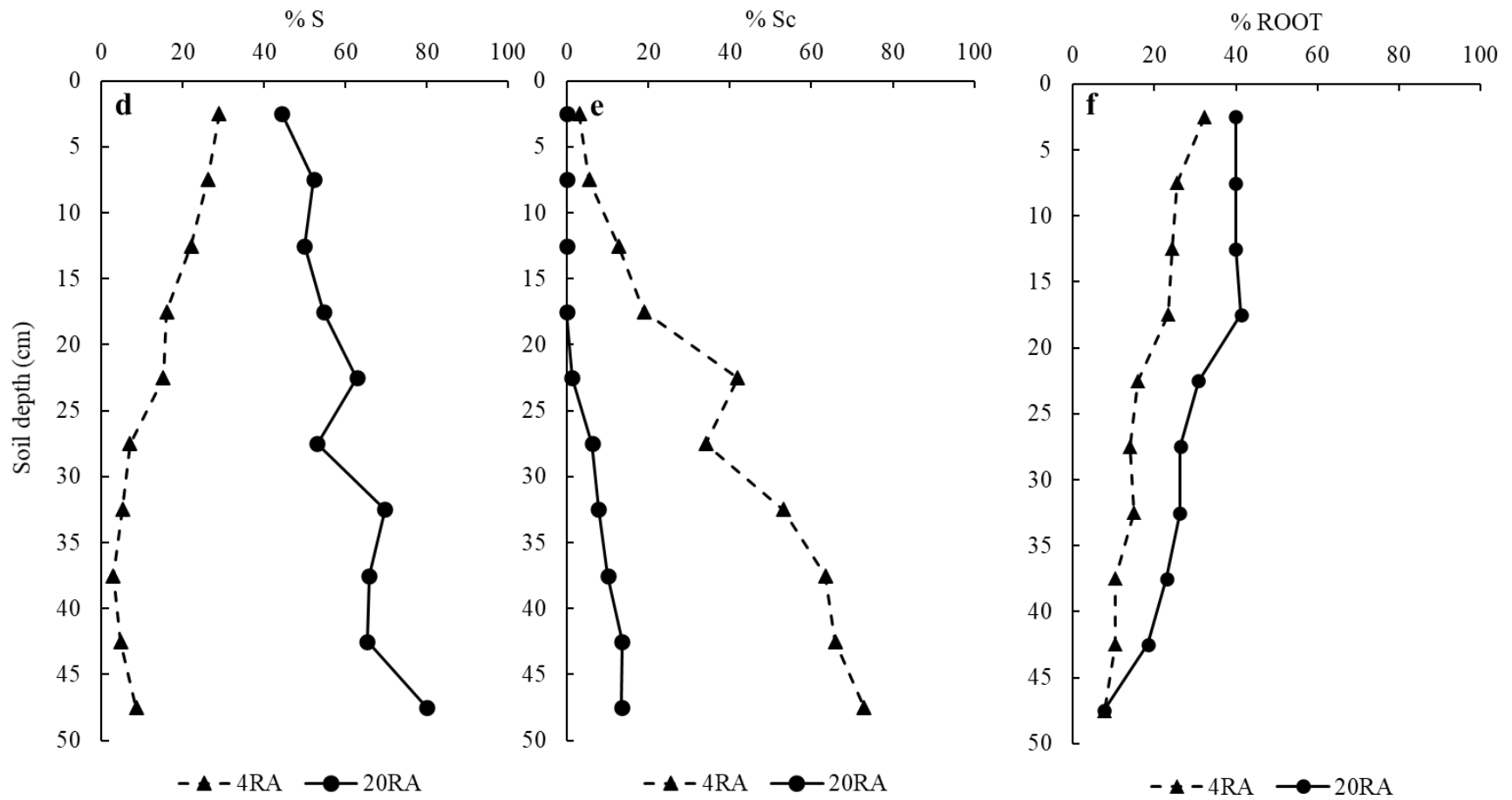
## **Acknowledgments**

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## SUPPORTING INFORMATION

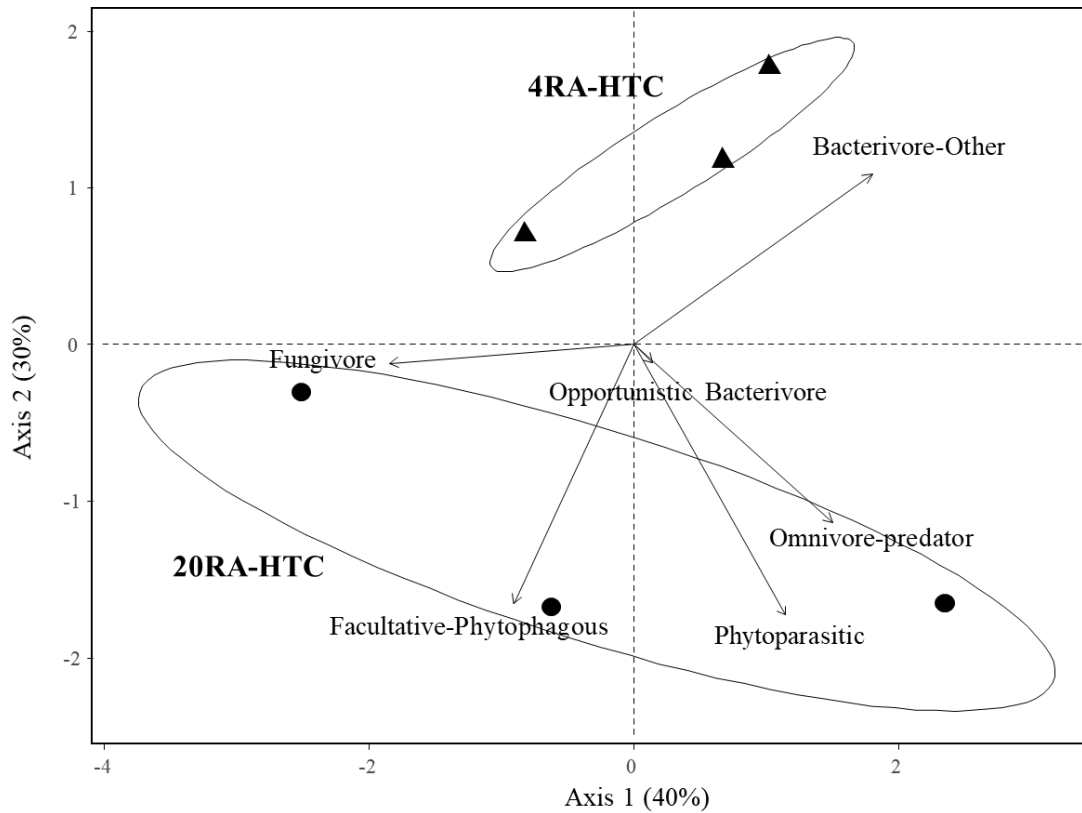
**Fig.S1** Relationships between soil depth (between 0 and 50 cm) and percentages of (a) root, (b) CAST, and (c) CASTw impacted by soil horizon in AL and RA. AL = Anthropised Luvisol, RA = Reconstructed Anthroposoil.







**Fig. S2.** Principal component analysis using abundances of the different functional groups of nematodes in HTC from three 4RA pits and three 20RA pits. Arrows correspond to variables associated to the first two axes; “▲” corresponds to a 4RA pit and “●” corresponds to a 20RA pit on the factorial plane defined by the two axes. 95% confidence ellipses correspond to soil types.



**Table S1** Description of study sites.

Code	Soil type	Soil name (IUSS, 2007)	Land-use	Location	Initial stripping depth (cm)	Age (years)	Number of sections	Cumulated length (m)	Width (m)	Inter-tree (m)	Cover	Trees
AL	Anthropised Luvisol	Terric Luvisol (Transportic, Siltic)	Roadside	Leon Blum Avenue	30	30	3	800	2.45	10	Grass	<i>Acer platanoides</i>
RA	Reconstructed Anthroposoil	Hortic Anthrosol (Organotransportic, Siltic)	Roadside	Europe Avenue	150	20	3	570	1.8	7	Creeping ivy	<i>Tilia pallida</i>

**Table S2** Summary table of soil-structure patterns observed in the studied soil profiles (adapted from Piron et al., 2012). \*Sc replaces

Ac pattern and includes both natural and anthropogenic soil compaction processes.

Soil-structure pattern	Morphological characteristics	Code (Piron et al., 2012)
CAST =	Packing of cast aggregates of homogeneous shape and size, individual and not welded. High roughness aspect and high porosity.	<b>CASTg</b>
CASTg + CASTw + CASTc + BUR/CAST	Packing of welded cast aggregate. High roughness aspect and average porosity.	<b>CASTw</b>
	Packing of strongly welded casts. Smooth aspect.	<b>CASTc</b>
	Packing of casts intersected by burrows	<b>BUR/CAST</b>
<b>BUR</b>	Earthworm burrow	<b>BUR</b>
<b>BULK =</b>	Structure without an aggregative aspect. Porosity is still visible but not composed of large macropores. Average roughness aspect.	<b>S</b>
<b>S + Sc</b>	Compacted area without discernible porosity. Low roughness.	<b>Sc*</b>

**Table S3** Percentages of roots, cast and bulk soil patterns, and apparent density ( $\text{g.cm}^{-3}$ ) per soil type and soil horizon from nine pits of 4RA and 9 pits of 20RA. Values are mean  $\pm$  standard deviation.

<i>Properties</i>		<b>4RA-HTC</b> (n = 9)	<b>4RA-HT</b> (n = 9)	<b>20RA-HL</b> (n = 9)	<b>20RA-HTC</b> (n = 9)	<b>20RA-HT</b> (n = 9)
<b>Roots</b>	<b>ROOT</b>	26.7 $\pm$ 15.0	11.7 $\pm$ 11.4	40.9 $\pm$ 13.7	38.3 $\pm$ 8.5	20.9 $\pm$ 7.4
<b>Burrows</b>	<b>BUR</b>	15.6 $\pm$ 5.9	3.3 $\pm$ 2.8	4.1 $\pm$ 5.0	2.0 $\pm$ 2.1	0.5 $\pm$ 0.5
<b>Cast patterns</b>	<b>CAST</b>	68.7 $\pm$ 11.0	36.1 $\pm$ 15.2	56.7 $\pm$ 15.8	46.8 $\pm$ 10.4	23.4 $\pm$ 7.7
	<b>BUR/CAST</b>	4.8 $\pm$ 3.6	0.9 $\pm$ 0.9	0.0 $\pm$ 0.0	0.0 $\pm$ 0.0	0.0 $\pm$ 0.0
	<b>CASTg</b>	0.8 $\pm$ 1.6	0.1 $\pm$ 0.2	1.1 $\pm$ 2.4	0.5 $\pm$ 1.0	0.2 $\pm$ 0.3
	<b>CASTw</b>	22.7 $\pm$ 12.6	10.0 $\pm$ 8.1	37.0 $\pm$ 18.2	16.1 $\pm$ 6.6	3.7 $\pm$ 1.5
	<b>CASTc</b>	40.5 $\pm$ 12.1	25.1 $\pm$ 8.9	18.7 $\pm$ 4.5	30.3 $\pm$ 9.3	19.6 $\pm$ 6.5
<b>Bulk soil patterns</b>	<b>BULK</b>	31.3 $\pm$ 11.0	63.9 $\pm$ 15.2	43.3 $\pm$ 15.8	53.2 $\pm$ 10.4	76.6 $\pm$ 7.7
	<b>S</b>	23.5 $\pm$ 15.5	6.2 $\pm$ 7.6	43.3 $\pm$ 15.8	52.7 $\pm$ 10.3	67.5 $\pm$ 17.4
	<b>Sc</b>	7.8 $\pm$ 7.5	57.6 $\pm$ 19.7	0.0 $\pm$ 0.0	0.5 $\pm$ 1.4	9.1 $\pm$ 19.7
<b>Apparent density</b>	<b>Da</b>	1.2 $\pm$ 0.1	1.4 $\pm$ 0.1	0.5 $\pm$ 0.2	1.2 $\pm$ 0.1	1.4 $\pm$ 0.1

**Table S4** Values of physical, chemical, and biological properties per soil type and soil horizon from samples of three 4RA pits and three 20 RA pits. Abundances of nematodes are expressed in individuals g<sup>-1</sup> of soil (Bac.O = Bacterivore-Other; Opp.Bac = Opportunistic Bacterivore; Phy.Par = Phyt parasitic; Fac.Phy = Facultative Phytophagous; Fung. = Fungivore; Omn.Pred = Omnivore-predator). See Fig.4 for complete legend.

<i>Properties</i>		<b>4RA-HTC</b> (n = 3)	<b>4RA-HT</b> (n = 3)	<b>20RA-HL</b> (n = 3)	<b>20RA-HTC</b> (n = 3)	<b>20RA-HT</b> (n = 3)
<i>Physical</i>	<b>Clay</b>	20.1 ± 1.0	19.2 ± 3.1	15.2 ± 1.2	18.8 ± 0.4	16.5 ± 1.6
	<b>Silt</b>	68.8 ± 2.5	72.9 ± 5.1	63.6 ± 1.4	68.3 ± 3.1	71.5 ± 5.9
	<b>Sand</b>	7.4 ± 0.9	6.9 ± 2.3	12.7 ± 1.0	9.1 ± 2.1	8.1 ± 3.2
	<b>EC</b>	0.17 ± 0.06	0.12 ± 0.04	0.20 ± 0.01	0.13 ± 0.00	0.16 ± 0.05
<i>Chemical</i>	<b>pH</b>	8.1 ± 0.06	7.6 ± 0.7	7.3 ± 0.3	7.3 ± 0.5	7.0 ± 0.4
	<b>Corg</b>	2.0 ± 0.2	0.5 ± 0.3	5.4 ± 0.7	2.0 ± 0.3	0.9 ± 0.4
	<b>C 0:50</b>	80.7 ± 0.5	-	74.7 ± 11.3	77.4 ± 2.3	-
	<b>C 50:2000</b>	19.4 ± 0.5	-	25.3 ± 11.3	22.6 ± 2.3	-
	<b>Ntot</b>	0.19 ± 0.03	0.07 ± 0.03	0.47 ± 0.08	0.13 ± 0.03	0.08 ± 0.02
	<b>N 0:50</b>	78.4 ± 2.3	-	66.5 ± 7.4	81.7 ± 1.0	-
	<b>N 50:2000</b>	21.6 ± 2.3	-	33.5 ± 7.4a	18.3 ± 1.0	-
	<b>C:N</b>	10.8 ± 0.5	7.9 ± 1.8	11.8 ± 2.7	18.6 ± 4.1	11.2 ± 1.5
	<b>P<sub>2</sub>O<sub>5</sub></b>	107.7 ± 11.3	56.2 ± 35.7	105.9 ± 13.5	67.9 ± 7.3	60.7 ± 15.2
	<b>CEC</b>	13.6 ± 0.7	10.3 ± 0.9	21.0 ± 7.8	11.8 ± 0.5	7.7 ± 1.3
	<b>CaO</b>	6361.7 ± 511.7	6590.0 ± 6334.2	7867.0 ± 715.3	4351.0 ± 1129.3	2507.7 ± 511.2
	<b>K<sub>2</sub>O</b>	485.0 ± 113.1	190.7 ± 0.2	326.7 ± 36.9	139.7 ± 12.5	21.0 ± 36.4
	<b>MgO</b>	269.7 ± 34.4	175.7 ± 1.0	504.0 ± 72.2	156.8 ± 35.3	3.5 ± 6.1
	<b>Na<sub>2</sub>O</b>	95.3 ± 5.1	41.7 ± 0.1	199.0 ± 44.2	270.7 ± 71.0	27.5 ± 47.6
	<b>Ca/CEC</b>	166.8 ± 5.3	219.0 ± 195.5	144.3 ± 41.7	131.4 ± 28.3	116.5 ± 12.0
<i>Chemical (TME)</i>	<b>Cr</b>	62.6 ± 4.9	61.2 ± 11.0	80.6 ± 46.9	65.5 ± 16.6	75.1 ± 16.1
	<b>Cu</b>	17.3 ± 1.3	16.0 ± 3.6	26.0 ± 1.8	19.6 ± 1.4	16.0 ± 3.8
	<b>Ni</b>	3.3 ± 3.1	6.8 ± 11.8	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	<b>Pb</b>	23.1 ± 1.7	18.5 ± 4.8	29.7 ± 1.4	31.4 ± 2.9	33.2 ± 8.5
	<b>Zn</b>	45.7 ± 4.8	32.1 ± 1.3	89.4 ± 5.0	53.5 ± 3.3	36.5 ± 2.0
<i>Biological (Microorganisms)</i>	<b>MB</b>	259.4 ± 10.6	-	1721.5 ± 528.1	489.5 ± 35.4	-
	<b>SIR</b>	6.8 ± 1.0	-	43.7 ± 29.4	12.4 ± 0.7	-
	<b>DEA</b>	0.43 ± 0.15	-	4.6 ± 1.6	1.23 ± 0.12	-
	<b>NEA</b>	0.17 ± 0.06	-	0.27 ± 0.21	0.07 ± 0.12	-
<i>Biological (Nematofauna)</i>	<b>EI</b>	63.0 ± 14.6	-	-	70.7 ± 17.2	-
	<b>SI</b>	48.5 ± 14.5	-	-	52.0 ± 46.4	-
	<b>IVD</b>	68.3 ± 33.0	-	-	58.7 ± 35.4	-
	<b>H'</b>	1.7 ± 0.3	-	-	0.8 ± 0.1	-
	<b>N</b>	13.8 ± 1.3	-	-	6.7 ± 1.2	-
	<b>Bac.O</b>	108.2 ± 52.9	-	-	46.0 ± 57.2	-
	<b>Opp.Bac</b>	80.2 ± 104.4	-	-	95.7 ± 110.4	-
	<b>Phy.Par</b>	286.8 ± 176.2	-	-	2355.0 ± 1923.2	-

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<b>Fac.Phy</b>	411.7 ± 249.1	-	-	3327.7 ± 1394.1	-
<b>Fung.</b>	77.8 ± 83.7	-	-	92.3 ± 69.9	-
<b>Omn.Pred</b>	41.0 ± 20.7	-	-	55.3 ± 55.0	-

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**CHAPITRE 2 - Caractérisation de la fertilité physique, chimique,  
et biologique de sols urbains issus de l'ingénierie pédologique**

## **Objectifs de l'étude**

Le but de cette étude était de caractériser la fertilité physique, chimique et biologique de deux types de sols issus de l'ingénierie pédologique reconstitués à des périodes similaires, dans un même contexte pédoclimatique et urbain.

## **Méthodologie**

Nous avons travaillé sur deux linéaires d'accompagnement de voirie distants de moins de 2 km l'un de l'autre : (i) des Luvisols anthropisés de 30 ans issus d'une ingénierie basique (i.e., reconstitution partielle sans amendement organique), (ii) des Anthrosols reconstitués de 20 ans (i.e., reconstitution totale avec amendement organique). Six fosses pédologiques ont été observées dans les Luvisols anthropisés, et neuf dans les Anthrosols reconstitués afin de décrire les différents horizons et de quantifier les pourcentages de racines et de biostructures lombriciennes. Pour trois fosses de chacun des types de sol, une vingtaine de paramètres physiques, chimiques et biologiques ont été mesurés sur chacun des horizons pour caractériser les différentes composantes de la fertilité du sol.

## **Principaux résultats**

Pour les deux types de sol, les composantes physiques, chimiques et biologiques de la fertilité sont clairement distinctes entre horizons d'un même sol et entre horizons équivalents de deux types de sol différents. De manière générale, la quantité de racines, de biostructures lombriciennes, de ressources trophiques pour les plantes et les organismes du sol décroissait avec la profondeur. L'horizon de surface des Luvisols anthropisés concentrait nettement l'activité racinaire et les ressources trophiques tandis que leur répartition était plus homogène dans les deux premiers horizons des Anthrosols reconstitués (i.e., litière et mélange terre-compost). A horizons



équivalents, la fertilité physique et chimique était nettement favorisée dans les Anthrosoles reconstitués par rapport aux Luvisols anthropisés.

**Title: Characterisation of the physical, chemical, and biological fertility of engineered soils in urban areas**

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## **Abstract**

The construction of cities is mostly associated with damages on urban soils' physical, chemical, and biological fertility. At the same time, there is a growing demand for including more nature in cities. To face this challenge, different soil engineering strategies exist to promote soil fertility. Basic strategies follow a systematic stripping of the topsoil for the needs of the construction (e.g., road) and involve the re-implementation of a stripped topsoil layer after the construction to allow vegetation growth. Elaborate strategies, more rarely used, involve the excavation of the whole existing soil (>1.50m) followed by the re-implementation of a deep horizon made of topsoil, a horizon of compost-amended topsoil over it, and finally a thin layer of organic mulch. However, little is known about the fertility of such soils several years after their implementation in urban areas. The objective of this study was to characterise the physical, chemical, and biological components of fertility in such engineered soils. Thus, we selected linear roadside soils implemented 30 years ago according to the basic process (called anthropised Luvisols in our study), and according to the elaborate process 20 years ago (called reconstructed Anthroposoils in our study). We conclude positive effects of compost amendment on physical and chemical fertility, still visible 20 years after Anthroposoils implementation. Besides, for both types of engineered soils, soil fertility was clearly distinguished at the scale of soil horizons and decreased with depth. Regarding anthropised Luvisols, there were significant negative effects of soil compaction which constrained the development of roots and earthworm activity beyond the second horizon. Overall, the elaborate process appeared more beneficial to soil fertility than the basic one.

**Keywords: soil structure, urban soils, pedological engineering, aggregation, bioturbation, earthworm casts**

## **1. Introduction**

Urban environments are a major issue in the context of environmental pollution, demographic growth, and climate change (McDonald et al., 2011; Filho et al., 2020). As a result, there is a growing interest for city dwellers in urban nature through green spaces (Lee and Maheswaran, 2011; Jennings et al., 2016) mostly made up of trees that provide shade, limit extreme temperatures, improve air quality, and increase well-being for city dwellers (Livesley et al., 2016; Bodnaruk et al., 2017). The fertility of urban soils plays a key role in the sustainability of trees in cities and corresponds to the capacity of soils to provide physical, chemical, and biological needs for the growth of plants (Abbott and Murphy, 2007). However, numerous studies related the detrimental consequences of the construction cities processes and urban environments on soil fertility (e.g., Chi Yung Jim, 1998; Li et al., 2013). For example, the physical structure of urban soils can be damaged by more or less irreversible mechanical disturbances (e.g., stripping of topsoil, manipulation by construction machines; Craul, 1985; Cogger, 2005; Vidal-Beaudet and Rossignol, 2018), leading to soil compaction which restricts root growth, limits aeration and water movement (Zisa et al., 1980; Craul and Patterson, 1989; Mullins, 1991). Biological properties of urban soils, such as microbial biomass and activity, can be particularly altered in highly or recently disturbed areas due to anthropogenic particles which can reduce their contribution to nutrient cycles (Lorenz and Kandeler, 2006; Wang et al., 2011). Thus, various strategies are developed by soil engineering to provide fertile soils favourable to tree growth and counter the constraints of the urban environment (Muller et al., 2012; Vidal-Beaudet and Rossignol, 2018; Maréchal et al., 2021). For example, anthropogenic materials (e.g., urban wastes) or natural material (e.g., stripped topsoil) can be used, amended or not, to provide shallow or deep soils depending on their future use (Cannavo et al., 2015; Deeb et al., 2020). However, it is difficult to access information about the implementation of engineered soils because urban projects involve many actors, and project specifications can be

confidential. Consequently, we still have little insight on the long-term consequences of contrasted soil engineering processes on soil fertility and root growth several decades after their implementation in urban areas (Jangorzo et al., 2018; Kranz et al., 2020).

In terms of soil fertility, soil engineering processes mostly involve the implementation of one or several horizons with different levels of fertility depending on the future land use and vegetation (Unep, 2012; Damas and Coulon, 2016). For example, deep engineered soils mostly provide high nutrient content in the surface horizon to favour root growth whereas sub-surface horizons do not necessarily require as many nutrients (Marié and Rossignol, 1997; Deeb et al., 2020). In that context, the physicochemical properties of soil material are characterised before their implementation by laboratory analyses in order to meet fertility requirements (e.g., pH, nutrients, water content; AFNOR, 2009). After soil construction, complementary information on fertility can be collected through the observation of soil profiles to observe soil structure and identify potential obstacles to root penetration and earthworm activity (Tardieu and Manichon, 1986; Piron et al., 2012). For example, visual assessment has been widely used in agricultural soils to describe and interpret soil structure dynamics depending on tillage intensity (Peigné et al., 2013; Boizard et al., 2016). However, urban soils are more difficult to investigate given the different constraints of digging pedological pits and finding replicates (e.g., access to historical knowledge, buried networks, authorisation requests, rehabilitation of soil surface after digging). Despite these constraints, these methods could be adapted to engineered soils to assess their fertility more precisely, validate or improve processes of soil construction and optimise the provision of ecosystem services (Blanchart et al., 2018).

The objective of this study was to assess soil fertility of two types of engineered soils from contrasted processes applied respectively 30 and 20 years ago: (i) basic soil engineering process involving the implementation of a single shallow horizon of non-amended topsoil on a truncated

natural soil (i.e., Anthropised Luvisols) and (ii) advanced soil engineering process involving the implementation of three horizons and the incorporation of compost in the surface horizon (i.e., Reconstructed Anthroposoils). We hypothesised higher fertility in Reconstructed Anthroposoils than in Anthropised Luvisols due to more favourable conditions implemented for root growth at the scale of the profile and soil horizons. Soil fertility was assessed from a physical, chemical, and biological laboratory analyses coupled with a fine-scale mapping of root and soil-structure patterns.

## **2. Materials and methods**

### ***2.1. Study sites***

The study area is located around the city of Paris (France), in the commune of Guyancourt (48° 76' 18'' N; 2° 07' 23'' E) which belongs to the urban community of Saint-Quentin-en-Yvelines, built as a “new city” in the 1970s to avoid urban concentration in Paris. The city expands on large agricultural parcels where the substrate is characterized by fine loess deposits more than 3 metres deep (Mégnyen, 1989). The climate is temperate, with a mean annual precipitation of 563 mm and a mean annual temperature of 11.2°C, a monthly minimum of -2.3°C and a monthly maximum of 26.4 °C (data from Météo France, 2008-2018 Trappes station).

Two types of engineered soils were studied made by following distinct engineering processes: (i) Anthropised Luvisols (AL), and (ii) Reconstructed Anthroposoils (RA). AL are the result of a basic soil engineering process that is commonly used in the urban context after civil engineering activities such as road construction (Strohmayr, 1999). It involved a systematic shallow stripping of 30 cm of the existing soil (i.e., topsoil removal), as it cannot be used for road base due to too low density (Strohmayr, 1999). After the road construction, about 30 cm of stripped and transported silty topsoil (organo-mineral material) was added to the surface of the truncated existing soil (i.e., Luvisol in our study) to favour the replanting of grass and trees planted

in unitary pits of topsoil. In our study, they were implemented at the end of the 1980s. RA are the result of an advanced soil engineering process that is more rarely used in the urban context. It involved a deep soil stripping of 150 cm of the existing soil and the reconstruction of an entire soil profile to optimise the root development and the growth of planted trees. Several layers were superposed from bottom to top: 125 cm of silty topsoil (organo-mineral material), 25 cm of a mixture of silty topsoil and compost from green waste (2 volumes of topsoil for 1 volume of compost), and finally a thin layer of pine bark mulch on which a permanent cover of creeping ivy was planted. The green waste compost complied with the NFU-44-551 standard and was screened at 20 mm (AFNOR, 2002). In our study, RA were implemented at the end of the 1980s. Both types of engineered soils corresponded to linear roadside soils (respectively 800 and 570-metre-long) bordered by asphalt surfaces (a 6-metre-wide road on one side and a 3-metre-wide sidewalk on the other side). Soil horizons were described in July 2020 and identified according to the French pedological reference system (Table 1; Baize and Girard, 2008) and associated to a type of horizon previously described. In the study, soil horizons are defined from top to bottom as H1, H2, H3 (Table 1).

Briefly, AL were 2.45-meter-wide, covered by grass, and planted with *Acer platanoides* every 10 m and RA were 1.8-metre-wide, covered by creeping ivy, and planted with *Tilia pallida* every 7 m, more details are available on Table S1. Given the difficulty to study soils in urban areas (e.g., availability of historical knowledge, authorisation of access, replicability), the two types of engineered soils located in Guyancourt were the more relevant to select after field investigations, the study of historical aerial photographs, exchanges with local managers and soil engineering experts. Thus, AL and RA were selected for the detailed historical knowledge available, their proximity (less than 2 km apart), their great age allowing to study their state decades after their

implementation (30 and 20 years old respectively), and the contrasting soil engineering processes followed for their construction (Table S1).

**Table 5** Description of soil horizons from 0 to 50 cm deep per type of engineered soil based on the observation of 6 Anthropised Luvisols and 9 Reconstructed Anthroposoils pedological pits.

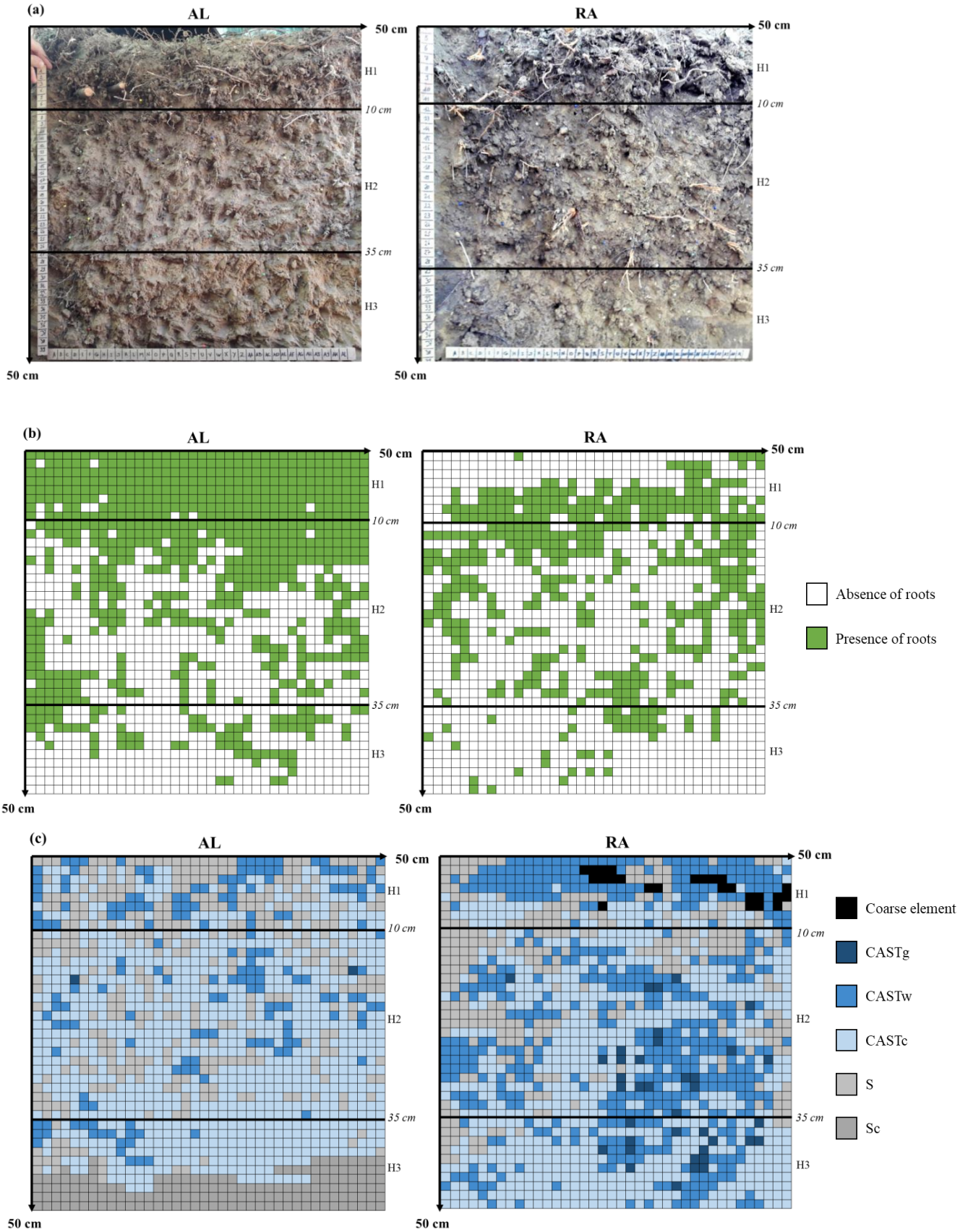
Anthropised Luvisol (n= 6)					Reconstructed Anthroposoil (n = 9)				
Horizon Code	Mean depth (cm ± SD)	Colour (Munsell colour chart)	Type of horizon (Baize & Girard, 2008)	Main characteristics	Horizon Code	Mean depth (cm ± SD)	Colour (Munsell colour chart)	Type of horizon (Baize & Girard, 2008)	Main characteristics
H1 AL-H1	0 – 13 (± 2.5)	7.5YR 5/2: Brown	(O) + Atp: Stripped and transported topsoil	Crumbly structure Loamy Organo-mineral Weakly compact	H1 RA-H1	0 - 7 (± 2.8)	10YR 3/1: Very dark grey	O: Very organic litter with mulch of pine barks	Crumbly structure Loamy Very organic Weakly compact
H2 AL-H2	13 – 29 (± 5.3)	10YR 5/6: Yellowish brown	Atp: Stripped and transported topsoil	Polyhedral structure Loamy Organo-mineral Compact	H2 RA-H2	7 – 32 (± 6.4)	10YR 4/2: Dark greyish brown	Atp: Stripped, transported and compost-amended topsoil	Crumbly to polyhedral structure Loamy Organo-mineral Weakly compact
H3 AL-H3	29 – 48 (± 5.2)	10YR 5/4: Light yellowish brown	E: Horizon E truncated	Polyhedral structure Loamy Mineral Very compact	H3 RA-H3	32 – 47 (± 8.3)	10YR 5/6: Yellowish brown	Atp: Stripped and transported topsoil	Polyhedral to massive structure Loamy Mineral Compact

## 2.2. Mapping of soil-structure patterns and roots on soil profiles

Pedological pits (100 cm x 150 cm x 110 cm, length × width × depth) were dug in the two types of engineered roadside soils, respectively 6 pits in AL and 9 in RA. Each pit was located in the middle of an inter-tree. Locations were selected in order to cover the entire length of the two avenues and to avoid areas with underground networks (e.g., electricity cables, pipes). Prior to



mapping, soil horizons were described and identified according to the French pedological reference system (Table 1; Baize and Girard, 2008). In that study, analyses were focused on the 50 first cm of the soil profile because it concentrates the main part of earthworms, roots, organic carbon and mineral nutrients (Henin et al., 1969). Mapping of roots and soil-structure patterns produced by earthworms were achieved following protocols adapted respectively from Tardieu and Manichon (1987) and Piron et al. (2012). Thus, soil profiles were carefully prepared with a knife to reveal the soil aggregates in their original aspect. Then, a grid frame (1.2 cm x 1.2 cm) was attached against the soil profiles (Fig.1) and pictures of the grid frame were taken to allow image analysis and give a precise transcription of observed roots and soil structure patterns. Then, digital mapping was achieved by associating each cell of the grid (1.44 cm<sup>2</sup>) to the presence or absence of a root, and to the major soil-structure pattern (i.e., representing more than 50% of the total surface of a cell) following a typology adapted from Piron et al. (2012) which distinguishes earthworm burrows (BUR), casts (fresh to very degraded casts: CASTg, CASTw, CASTC), and bulk soil (compacted or not: S, Sc). The typology used in that study is presented in Table S2. In total, 1482 cells were filled for each pit, covering 2134 cm<sup>2</sup> of the soil profile. Examples of root maps and soil-structure pattern maps are given in Fig.1. Thereafter, % of soil-structure patterns were calculated (i) on the whole 50 cm x 50 cm square described; (ii) per type of horizon defined on the field; (iii) every 5 cm regardless of soil horizons.



**Fig. 1.** Examples of (a) soil profiles and associated (b) root maps and (c) soil-structure patterns maps (on the left = AL; on the right = RA). H1 = surface horizon, H2 = sub-surface horizon, H3 = deep horizon.

### *2.3. Soil sampling and laboratory analyses*

Three pits among the six AL pits and three pits among the nine RA pits were randomly selected to achieve physical, chemical, and biological laboratory analyses. Soil samples were separately taken from each horizon between 0 and 50 cm-deep of each selected pit. For each horizon, the sample was taken from a pool of 10 sub-samples. Thus, 6 types of samples were analysed (i) AL-H1 (n = 3), (ii) RA-H1 (n = 3), (iii) AL-H2 (n = 3), (iv) RA-H2 (n = 3), (v) AL-H3 (n = 3), and (vi) RA-H3 (n = 3).

Regarding physical and chemical properties, each sample was analysed for particle-size distribution (clay  $<2 \mu\text{m}$ ; silt  $2 < \mu\text{m} < 50$ ; sand  $50 < \mu\text{m} < 200$ , %; NFX 31-107 without decarbonatation; AFNOR, 2003), electrical conductivity (EC;  $\text{mS}\cdot\text{cm}^{-1}$ ; NF EN 13038; AFNOR, 2012), pH (water; NF ISO 10390; AFNOR, 2005), organic carbon ( $C_{\text{org}}$ ; %; NF ISO 14235; AFNOR, 1998), total nitrogen ( $N_{\text{tot}}$ ; %; NF ISO 13878; AFNOR, 1998a), phosphorus ( $P_2O_5$ ;  $\text{mg}\cdot\text{kg}^{-1}$ ; NF ISO 11263; AFNOR, 1995), cation exchange capacity (CEC;  $\text{meq}\cdot 100\text{g}^{-1}$ ; NF X 31.130; AFNOR, 1999), exchangeable cations (CaO,  $K_2O$ , MgO,  $Na_2O$ ;  $\text{mg}\cdot\text{kg}^{-1}$ ; NF X 31.108; AFNOR, 2002), and the calcium saturation rate was calculated (Ca/CEC). In addition, water retention was studied on undisturbed soil cylinders of  $100 \text{ cm}^3$  sampled in H2 from six distinct pits of AL (n = 6) and RA (n = 6) to compare their soil retention curves following the ISO 11274 standard (AFNOR, 2019). Thus, soil cylinders were saturated and brought to the pressure heads of pF 2, 2.3, 2.5, 2.8, 3, 3.3, 3.5, 4 and 4.2 using a pressure cell. At each pressure head, the wet weight of soil cylinders was taken. Finally, soil cylinders were oven-dried at  $105^\circ\text{C}$  during 48 hours and

their dry weight was determined. Water mass contents ( $W_{pF}$ ) were calculated as follows:

$$W_{pFx}(g/g) = \frac{\text{wet weight at } pFx - \text{dry weight}}{\text{dry weight}}$$

Besides, apparent density ( $Da$ ;  $g.cm^{-3}$ ), water reserve

( $WR$ ;  $mm/cm$ ) and Dexter index ( $S_{dexter}$ ) were calculated from this data.  $Da$  was calculated as follows:  $Da (g.cm^{-3}) = \frac{\text{dry weight}}{\text{cylinder volume}}$ .  $WR$  was calculated as follows:  $WR (mm/cm) = Da * (W_{pF2.5} - W_{pF4.2}) * 10$  (Baize, 2018).  $S_{dexter}$  is an index of soil physical quality defined by Dexter

(2004) and equal to the slope of the soil water retention curve fitted with the van Genuchten equation (1980) at its inflection point.

Regarding biological properties, only AL-H1 ( $n = 3$ ), RA-H1 ( $n = 3$ ), AL-H2 ( $n = 3$ ), and RA-H2 ( $n = 3$ ) were sampled and then analysed in terms of microbial biomass ( $MB$ ;  $mg.kg^{-1}$ ; NF ISO 14240-2; AFNOR, 2011), and for substrate-induced respiration ( $SIR$ ;  $\mu g.C-CO_2 g^{-1} h^{-1}$ ), denitrification rate ( $DEA$ ;  $\mu g.N-N_2O g^{-1} h^{-1}$ ), and nitrification rate ( $NEA$ ;  $\mu g N-NO_2+NO_3 g^{-1} h^{-1}$ ) as microbial activities are concentrated in surface and sub-surface horizons (Taylor et al., 2002; Lorenz and Kandeler, 2006). For  $SIR$ ,  $DEA$ , and  $NEA$  analyses, soil samples were sieved to 2 mm and stored at  $-18^\circ C$  until microbial analyses were achieved.  $SIR$  was measured as  $CO_2$  production (Anderson and Domsch, 1978). Soil samples (5 g) were placed in an airtight flask in 21%  $O_2$  atmosphere, containing 0.5 ml of a nutritive solution including glucose (1.2 mg of C-glucose  $g^{-1}$  of dried soil) then incubated at  $28^\circ C$ .  $CO_2$  concentrations were measured each hour for 5 h and the slope of the linear regression was used to estimate aerobic respiration as the  $CO_2$  produced ( $g^{-1} h^{-1}$ ).  $DEA$  was measured in soil using a method adapted from Dassonville et al. (2011). First 5, g of soil sample was placed in 150-ml airtight plasma-flasks sealed with rubber stoppers. In each flask, air was removed and replaced with a  $He/C_2H_2$  mixture (90: 10, v/v) to create anoxic conditions and inhibit  $N_2O$ -reductase. A nutritive solution (0.5 ml) containing glucose (0.5 mg of C-glucose  $g^{-1}$  of dried soil), glutamic acid (0.5 mg of C-glutamic acid  $g^{-1}$  of dried soil) and

potassium nitrate ( $50 \mu\text{g}$  of  $\text{N-KNO}_3 \text{ g}^{-1}$  of dried soil) was added to the soil. The amount of  $\text{N}_2\text{O}$  during incubation at  $28^\circ\text{C}$  was measured each hour for 6 to 8 h. The slope of the linear regression was used to estimate anaerobic respiration (denitrification) as the  $\text{N}_2\text{O}$  produced ( $\text{g}^{-1} \text{ h}^{-1}$ ). NEA was determined according to a modified method described by Dassonville et al. (2011). Soil samples were incubated under aerobic conditions for 10h with 30 mL of  $(\text{NH}_4)_2\text{SO}_4$  ( $1.25 \text{ mg N.L}^{-1}$ ) using a continuous shaking (140 rpm,  $28^\circ\text{C}$ ).  $\text{NO}_2^-$  and  $\text{NO}_3^-$  production rates were measured after 2, 4, 8 and 10 hr by ionic chromatography (DX120; Dionex, Salt Lake City, UT, USA).

#### ***2.4. Statistical analyses***

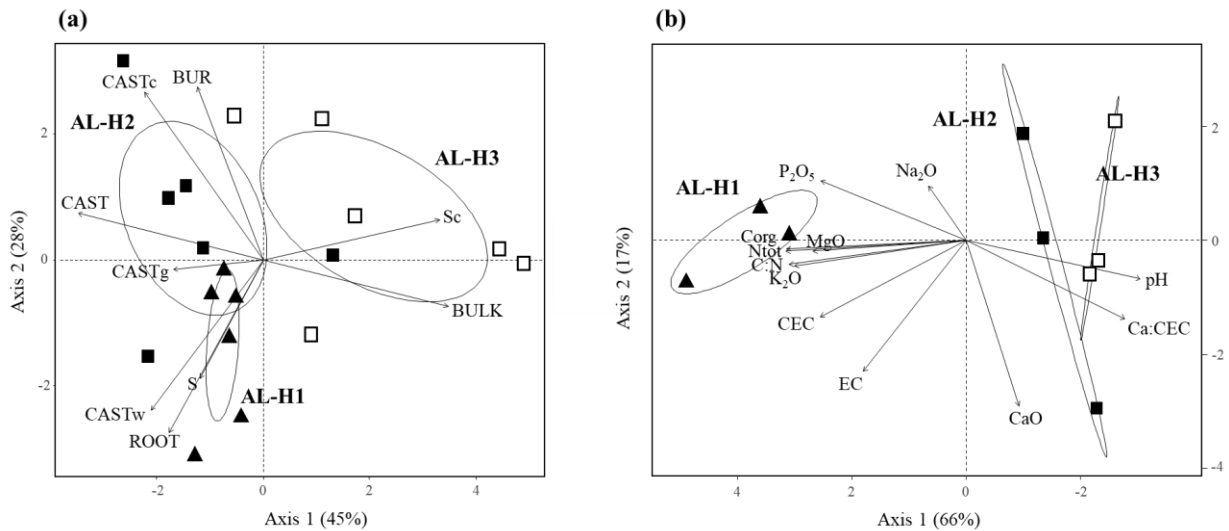
A Shapiro-Wilk normality test was performed to verify the normality of all the data prior further analyses. All statistical analyses were performed on R software (R Core Team, 2019). Significance was evaluated in all cases at  $P < 0.05$ .

In order to analyse physical, chemical, and biological properties along soil profiles, principal component analyses (PCAs) were performed on data collected (i) from AL-H1, AL-H2, AL-H3 and (ii) from RA-H1, RA-H2, RA-H3. PCAs were also performed to analyse soil properties of equivalent horizon of the two types of engineered soils: (i) AL-H1 and RA-H1, (ii) AL-H2 and RA-H2, (iii) AL-H3 and RA-H3. For analyses, data from soil profiles were treated separately to data from laboratory analyses because the number of replicates was different. The variables used for each PCA corresponded to variables common to each type of horizon or each type of soil. The variables associated with soil texture (i.e., clay, silt, sand) were excluded from the analyses because the soil material used for soil construction was similar in both types of soil, and soil engineering do not impact this parameter.

### 3. Results

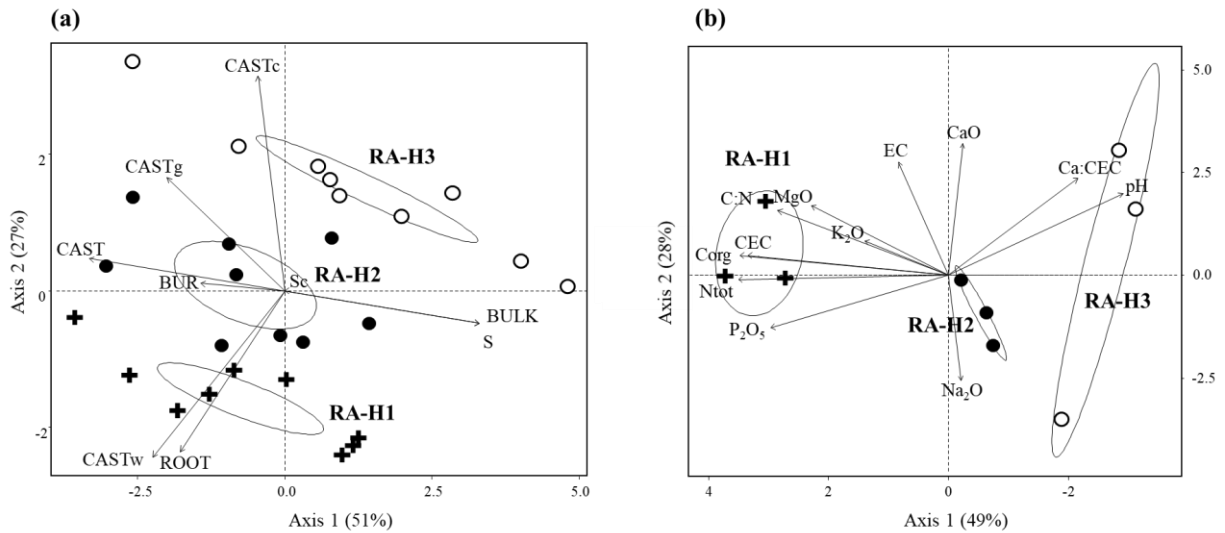
#### 3.1. Soil properties along engineered soil profiles

Regarding AL soil profiles, H1 concentrated the highest percentage of ROOT (72.4% ± 16.5; Table S3) which was clearly visible on root maps (e.g., Fig.1b). Chemical properties of H1 were also clearly differentiated from H2 and H3 by higher values of Corg, Ntot, C:N, K<sub>2</sub>O, and MgO (Fig. 2b; Table S4). H2 and H3 were characterised by higher pH and Ca:CEC ratio. In terms of soil-structure patterns, percentages of CAST were the highest in H1 and H2 (Fig. 2a), mostly represented by CASTc patterns (respectively 41.1% ± 14.2 and 59.9% ± 17.1; Table S3). By contrast, H3 presented higher percentages of BULK patterns, Sc in particular (39.0% ± 34.5; Fig. 2b; Table S3). Regarding RA soil profiles, each horizon had very distinct properties (Fig. 3). From surface to depth, decreases in ROOT, CASTw, Corg, Ntot, C:N, P<sub>2</sub>O<sub>5</sub>, and CEC were observed whereas CASTc and pH increased with depth (Fig. 3; Fig. S1).



**Fig. 2.** Principal component analysis using physical, chemical, and biological properties of AL horizons from (a) soil profile data and (b) laboratory data. Arrows correspond to variables associated to the first two axes; ▲ = horizon H1 of an AL pit; ■ = horizon H2 of an AL pit; □ =

horizon H3 of an AL pit. EC = electrical conductivity ( $\text{mS}\cdot\text{cm}^{-1}$ ); CEC = cation exchangeable capacity ( $\text{meq}\cdot 100\text{g}^{-1}$ );  $\text{P}_2\text{O}_5$  = available phosphorus ( $\text{mg}\cdot\text{kg}^{-1}$ ); Corg = organic carbon (%); Ntot = total nitrogen (%); C:N = carbon on nitrogen ratio (-); CaO,  $\text{K}_2\text{O}$ , MgO,  $\text{Na}_2\text{O}$  = exchangeable cations ( $\text{mg}\cdot\text{kg}^{-1}$ ); Ca:CEC =  $\text{Ca}^{2+}$  saturation rate. 95% confidence ellipses correspond to soil horizons. See Table S2 for soil-structure pattern legend.



**Fig. 3.** Principal component analysis using physical, chemical, and biological properties of RA horizons from (a) soil profile data and (b) laboratory data. Arrows correspond to variables associated to the first two axes;  $\oplus$  = horizon H1 of a RA pit;  $\bullet$  = horizon H2 of a RA pit;  $\circ$  = horizon H3 of a RA pit. EC = electrical conductivity ( $\text{mS}\cdot\text{cm}^{-1}$ ); CEC = cation exchangeable capacity ( $\text{meq}\cdot 100\text{g}^{-1}$ );  $\text{P}_2\text{O}_5$  = available phosphorus ( $\text{mg}\cdot\text{kg}^{-1}$ ); Corg = organic carbon (%); Ntot = total nitrogen (%); C:N = carbon on nitrogen ratio (-); CaO,  $\text{K}_2\text{O}$ , MgO,  $\text{Na}_2\text{O}$  = exchangeable cations ( $\text{mg}\cdot\text{kg}^{-1}$ ); Ca:CEC =  $\text{Ca}^{2+}$  saturation rate. 95% confidence ellipses correspond to soil horizons. See Table S2 for soil-structure pattern legend.

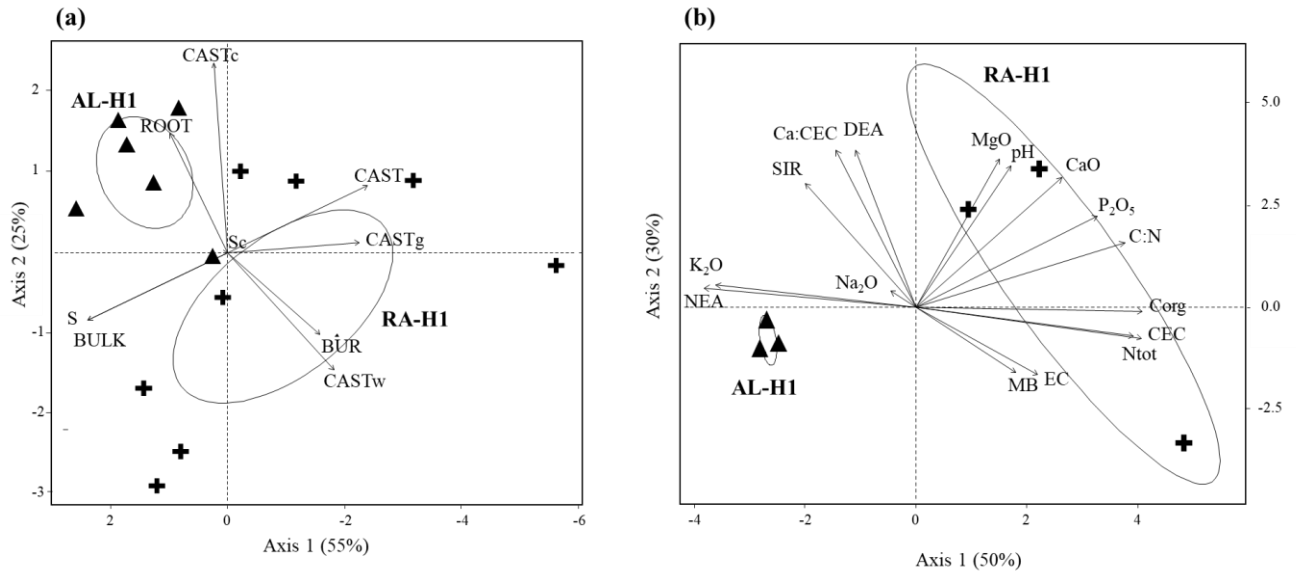
### 3.2. Comparison of equivalent horizons of engineered soils

Regarding the H1 horizon, it was characterised in AL by a higher percentage of ROOT than in RA (respectively  $72.4\% \pm 16.5$  and  $52.7\% \pm 15.0$ ; Fig. 4a; Table S3) and almost two times more CASTc patterns (respectively  $41.1\% \pm 14.2$  and  $21.2\% \pm 15.0$ ; Fig. 4a; Table S3). On the contrary, H1 presented notably in RA a higher percentage of CASTw ( $52.7\% \pm 8.1$  against  $23.5\% \pm 15.4$ ; Fig. 4a; Table S3). Overall, the total percentage of casts was more heterogenous in RA ( $74.1\% \pm 15.0$ ; Fig. 4a; Table S3) than in AL ( $64.6\% \pm 5.8$ ; Fig. 4a; Table S3). In terms of chemical and biological properties, RA was characterised by higher values of Corg, Ntot, C:N, P<sub>2</sub>O<sub>5</sub>, and CEC whereas AL was characterised by higher values of K<sub>2</sub>O and NEA (Fig. 4b; Table S4).

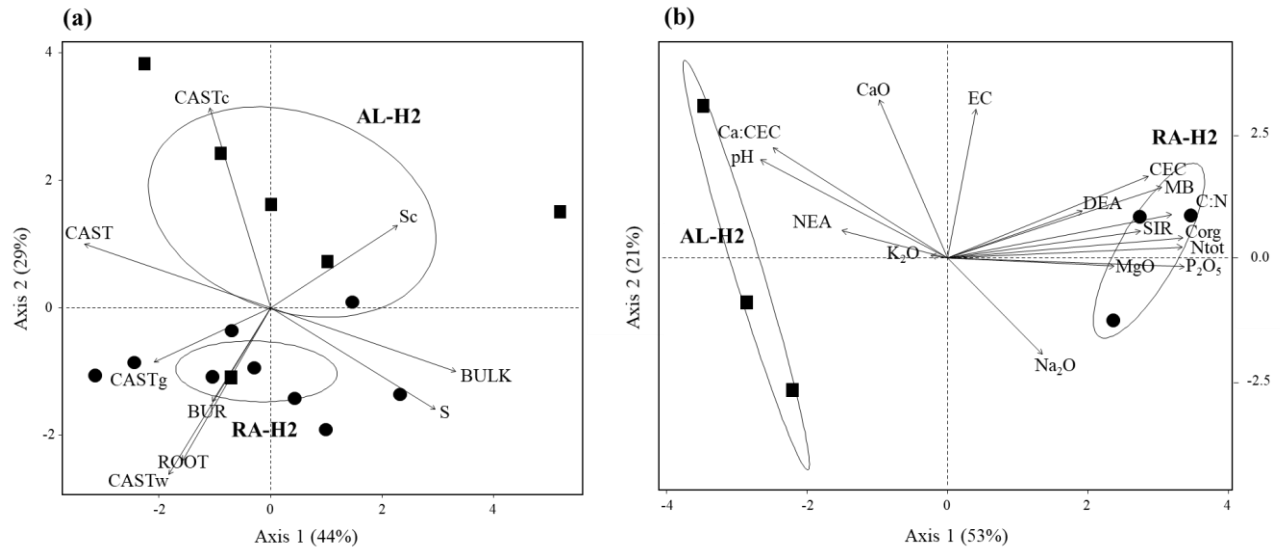
Regarding the H2 horizon, RA presented higher ROOT values than AL contrary to H1 (respectively  $45.2\% \pm 11.8$  and  $33.5\% \pm 19.9$ ; Fig. 5a; Table S4). As for H1, CASTw patterns were more numerous in RA whereas CASTc were more numerous in AL (Fig. 5a; Table S3). The H2 horizon was also contrasted in terms of chemical and biological properties as RA showed higher values of Corg, Ntot, C:N, P<sub>2</sub>O<sub>5</sub>, CEC, MB, and SIR whereas AL showed higher values of pH and Ca:CEC (Fig. 5b; Table S4). On the contrary, water retention, apparent density, useful reserve, and Dexter index were similar between AL and RA (Fig. S2; Table S5).

Regarding the H3 horizon, the percentage of ROOT was similar in AL and RA (respectively  $16.3\% \pm 8.7$  and  $15.5\% \pm 6.0$ ; Table S3). Sc was only represented in AL, which particularly differentiated the two types of soil (Fig. 6a), and was as abundant as CASTc pattern (respectively  $39.0\% \pm 35.5$  and  $39.1\% \pm 17.1$ ; Table S3). CASTc was also the major pattern in RA, reaching  $50.6\% \pm 13.4$ , followed by S pattern ( $37.3\% \pm 19.8$ ; Table S3). Overall, the total percentage of CAST was higher in RA than in AL (respectively  $62.7\% \pm 19.8$  and  $42.9\% \pm 18.6$ ; Table S3; Fig.S1) AL and RA were more contrasted in terms of chemical properties as RA showed higher values of Corg, Ntot, P<sub>2</sub>O<sub>5</sub>, and K<sub>2</sub>O (Fig. 6b; Table S4).

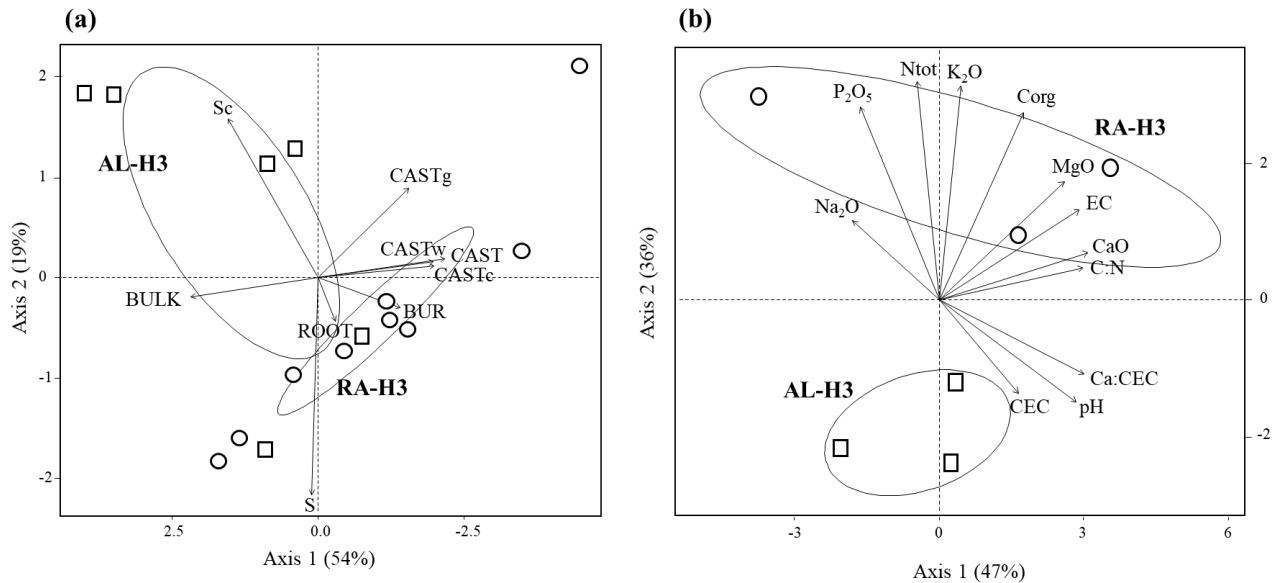




**Fig. 4.** Principal component analysis using physical, chemical, and biological properties of AL-H1 and RA-H1 from (a) soil profile data and (b) laboratory data. Arrows correspond to variables associated to the first two axes. ▲ = horizon H1 of an AL pit; + = horizon H1 of a RA pit. EC = electrical conductivity ( $\text{mS}\cdot\text{cm}^{-1}$ ); CEC = cation exchangeable capacity ( $\text{meq}\cdot 100\text{g}^{-1}$ );  $\text{P}_2\text{O}_5$  = available phosphorus ( $\text{mg}\cdot\text{kg}^{-1}$ ); Corg = organic carbon (%); Ntot = total nitrogen (%); C:N = carbon on nitrogen ratio (-); CaO,  $\text{K}_2\text{O}$ , MgO,  $\text{Na}_2\text{O}$  = exchangeable cations ( $\text{mg}\cdot\text{kg}^{-1}$ ); MB = microbial biomass ( $\text{mg}\cdot\text{kg}^{-1}$ ); SIR = substrate-induced respiration ( $\mu\text{g}\cdot\text{C}\cdot\text{CO}_2\text{ h}^{-1}\text{ g}^{-1}$ ); DEA = denitrification rate ( $\mu\text{g}\cdot\text{N}\cdot\text{N}_2\text{O h}^{-1}\text{ g}^{-1}$ ); NEA = nitrification rate ( $\mu\text{g}\text{ N}\cdot\text{NO}_2+\text{NO}_3\text{ h}^{-1}\text{ g}^{-1}$ ). See Table S2 for soil-structure pattern legend. 95% confidence ellipses correspond to soil types.



**Fig. 5.** Principal component analysis using physical, chemical, and biological properties of AL-H2 and RA-H2 from (a) soil profile data and (b) laboratory data. Arrows correspond to variables associated to the first two axes. ■ = horizon H2 of an AL pit; ● = horizon H2 of a RA pit. EC = electrical conductivity ( $\text{mS}\cdot\text{cm}^{-1}$ ); CEC = cation exchangeable capacity ( $\text{meq}\cdot 100\text{g}^{-1}$ );  $\text{P}_2\text{O}_5$  = available phosphorus ( $\text{mg}\cdot\text{kg}^{-1}$ );  $\text{C}_{\text{org}}$  = organic carbon (%);  $\text{N}_{\text{tot}}$  = total nitrogen (%); C:N = carbon on nitrogen ratio (-); CaO,  $\text{K}_2\text{O}$ , MgO,  $\text{Na}_2\text{O}$  = exchangeable cations ( $\text{mg}\cdot\text{kg}^{-1}$ ); MB = microbial biomass ( $\text{mg}\cdot\text{kg}^{-1}$ ); SIR = substrate-induced respiration ( $\mu\text{g}\cdot\text{C}\cdot\text{CO}_2\text{ h}^{-1}\text{ g}^{-1}$ ); DEA = denitrification rate ( $\mu\text{g}\cdot\text{N}\cdot\text{N}_2\text{O h}^{-1}\text{ g}^{-1}$ ); NEA = nitrification rate ( $\mu\text{g}\text{ N}\cdot\text{NO}_2+\text{NO}_3\text{ h}^{-1}\text{ g}^{-1}$ ). See Table S2 for soil-structure pattern legend. 95% confidence ellipses correspond to soil types.



**Fig. 6.** Principal component analysis using physical, chemical, and biological properties of AL-H3 and RA-H3 from (a) soil profile data and (b) laboratory data. Arrows correspond to variables associated to the first two axes.  $\square$  = horizon H3 of an AL pit;  $\circ$  = horizon H3 of a RA pit. EC = electrical conductivity ( $\text{mS}\cdot\text{cm}^{-1}$ ); CEC = cation exchangeable capacity ( $\text{meq}\cdot 100\text{g}^{-1}$ );  $\text{P}_2\text{O}_5$  = available phosphorus ( $\text{mg}\cdot\text{kg}^{-1}$ ); Corg = organic carbon (%); Ntot = total nitrogen (%); C:N = carbon on nitrogen ratio (-); CaO,  $\text{K}_2\text{O}$ , MgO,  $\text{Na}_2\text{O}$  = exchangeable cations ( $\text{mg}\cdot\text{kg}^{-1}$ ). See Table S2 for soil-structure pattern legend. 95% confidence ellipses correspond to soil types.

## 4. Discussion

### 4.1. Physical fertility of engineered soils

CAST represented on average 65% of the total surface studied which is 1.5 to 6.5 times superior to values observed in agricultural soils with a similar approach (i.e., 10 to 42%; Piron et al., 2006; Piron, 2008). This could be explained by the absence of recent structural deterioration caused by mechanical disturbances (e.g., tillage, stripping) in engineered soils contrary to agricultural soils (VandenBygaart et al., 2000; Bottinelli et al., 2010). Thus, root growth may be enhanced in

engineered soils thanks to the contribution of earthworm casts, among others, to nutrient availability for plants (Chaoui et al., 2003; Van Groenigen et al., 2019). However, soil engineering significantly impacted detailed cast patterns. CASTw (i.e., the first level of degradation of fresh casts) was 2.2 times higher in RA than in AL whereas CASTc (i.e., the last level of degradation of fresh casts) was 1.2 times higher in AL than in RA. This difference could be a direct consequence of the higher organic matter content in RA, due to the thick organic litter in H1 and to the compost amendment in H2, which stimulates earthworm activity (Hughes et al., 1996; Pérès et al., 1998) and may favour the production of fresh casts. Besides, organic matter content positively impacts cast stability thanks to binding agents between organic and mineral particles (Tisdall and Oades, 1982; Guggenberger et al., 1996; Scullion and Malik, 2000). Thus, CASTw may degrade slower in RA allowing their observation probably several weeks after their production. This higher proportion of CASTw in RA could also be beneficial for global soil fertility as nutrient supply and retention are enhanced in fresh casts (Aira and Domínguez, 2014; Singh et al., 2016; Van Groenigen et al., 2019).

Regarding the distribution of cast patterns and roots among horizons of engineered soils, CAST, CASTw and ROOT decreased along soil profiles whereas BULK and Sc were the highest in the deepest horizon. This is partly explained by the highest biological activity on the surface compared to deep horizons (Henin et al., 1969). However, AL profiles differed from RA profiles by the concentration of ROOT in the surface horizon H1 (until 13 cm-deep on average) in comparison to H2 and H3, whereas in RA profiles, ROOT were concentrated in both H1 and H2 (until 32 cm-deep on average). Besides, at similar depths, the deepest horizon AL-H3 was composed at 57% of bulk soil patterns, including 39% of compacted bulk soil (i.e., Sc), whereas RA-H3 was composed at 63% of cast patterns. These observations can be linked to higher compaction from H2 observed in AL compared to RA which highly constrains root growth and

earthworm activity into the sub-surface and deep horizons (Unger and Kaspar, 1994; Buck et al., 2000; Stovold et al., 2004). Compaction in AL may be explained either by natural or anthropogenic compaction during the construction phase (e.g., repeated passage of heavy machinery on stripped soil; Jim, 1998; Strohmayer, 1999). In RA, this phenomenon is avoided by the excavation of surface and deep horizons which may be compacted, followed by the implementation of soil material manipulated in dry conditions to preserve soil structure (i.e., water content below the plasticity limit; Marié and Rossignol, 1997; Muller et al., 2012). Besides, the use of organo-mineral material for deep horizons and the incorporation of compost in the surface horizon provide RA high content of organic matter which is known to effectively reduce soil bulk density (Chaudhari et al., 2013; McGrath et al., 2020). Thus, the visual assessment of soil physical fertility of engineered soils highlighted more favourable conditions in RA than in AL for earthworm activity, root growth and anchorage in sub-surface and deep horizons. In addition, the Dexter index indicated for both types of engineered soil a good structural quality (on average 0.066 in AL-H2 and 0.055 in RA-H2), and, in particular, a well-defined microstructural porosity (Dexter, 2004). Besides, apparent densities of  $1.3 \text{ g.cm}^{-3}$  in both types of soils associated with water reserve between 1.4 and 1.5 mm/cm correspond to ranges beneficial to root growth (Craul, 1985; Bruand et al., 1996).

#### ***4.2. Chemical and biological fertility of engineered soils***

As we hypothesised, chemical properties clearly differed between the two types of engineered soils, at the profile scale (between 0 and 50 cm deep) and also at the scale of soil horizons. Chemical properties associated with nutrient supply and retention such as Corg, Ntot, CEC, and  $\text{P}_2\text{O}_5$  were on average 1.3 to 3 times superior in RA than in AL, and globally higher in RA than values found in urban soils (Jim, 1998a, 1998b; Jimenez et al., 2013). For example, the large amount of available

phosphorus in RA ( $79.8 \pm 26.1$  mg/kg) illustrated an improvement in nutrient supply compared to low values ( $\sim 10$  mg/kg) usually observed in urban soils (Jim, 1998b; Park et al., 2010; Yue et al., 2017). These results could be mainly explained by the presence in RA of a thick litter on the surface and compost-amendment in sub-surface which ensure the provision of nutrients as showed by Cogger (2005) and Kranz et al. (2020) in urban soils. Kranz et al. (2020) also underlined the lack of knowledge regarding the effects of compost incorporation in the long term. Some studies reported an effect 6 years after incorporation but our results showed remaining significant effects 20 years after incorporation. Surprisingly, biological properties were similar between AL and RA with values of microbial biomass (MB) for example lower than in non-amended engineered roadside soils of New York, but higher microbial activity (Deeb et al., 2018; Joyner et al., 2019). Nonetheless, nitrification rate (NEA) was 1.9 times lower in RA than in AL which can be linked to the stabilisation of nitrification due to the protection of organic matter in dry casts (Decaëns et al., 1999).

By looking in detail at soil properties from surface to depth, chemical and biological fertility clearly decreased along soil profiles, regardless of soil engineering. Indeed, almost all the properties associated with nutrient supply and retention were the highest in H1 (excepted pH, CaO, Na<sub>2</sub>O, and NEA). For example, Corg and P<sub>2</sub>O<sub>5</sub> were respectively 7 times and 2 times higher in H1 than in H3 which is consistent with numerous studies highlighting a negative effect of soil depth on chemical fertility mostly due to the natural accumulation of fresh organic matter at the soil surface (e.g., Sharma et al., 2009; Mengistu et al., 2017), associated with the implementation of organic surface horizons (i.e., organo-mineral topsoil in AL, organo-mineral amended topsoil in RA). A difference can be noticed in the repartition of nutrients in soil profiles between AL and RA. Indeed, regarding AL, Corg and K<sub>2</sub>O which were concentrated in H1 (respectively 4 and 2.4 times higher than in H2) whereas fewer contrasts were observed between RA-H1 and RA-H2

(respectively 2 and 1.6 times higher in H1 than in H2). Thus, the concentration of nutrients in the surface horizon of AL could be less favourable to root development and anchorage in deeper horizons than a more homogenous provision of nutrients among surface and sub-surface horizons of RA. Similarly, biological properties were enhanced in the surface horizon (excepted NEA) by at least 60% compared with the sub-surface horizon in relation to higher trophic resources at lower depths (Taylor et al., 2002; Lorenz and Kandeler, 2006). Overall, the laboratory assessment of engineered soil fertility highlighted similar physical and biological properties but clear beneficial effects of engineering processes leading to RA in terms of chemical fertility at the profile and horizon scales.

## **5. Conclusion**

The combined approaches of soil profile mapping (soil-structure patterns and roots) and laboratory analyses offered complementary results for the assessment of engineered soils' fertility. Overall, our study highlighted a higher physical and chemical fertility of reconstructed Anthrosoils from advanced engineering processes, particularly enhanced by compost incorporation in H2, whereas biological fertility was not distinguished between anthropised Luvisols and Reconstructed Anthrosoils. Fine-scale soil profiles maps allowed to precisely describe the distribution of cast aggregates and roots among soil horizons, showing their concentration in the surface horizon of anthropised Luvisols due to compaction of deeper horizons.

## **Acknowledgments**

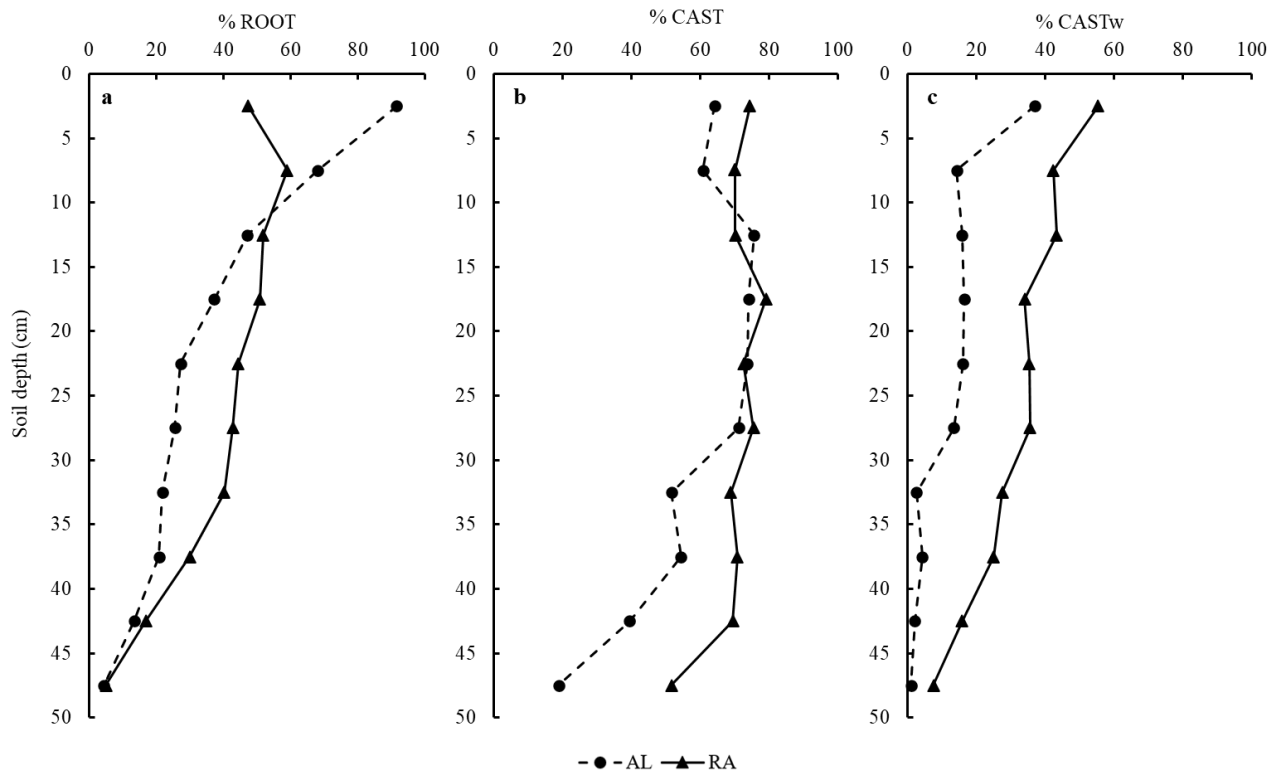
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## SUPPORTING INFORMATION

**Fig.S1.** Relationships between soil depth (between 0 and 50 cm) and percentages of (a) ROOT, (b) CAST, and (c) CASTw impacted by soil horizon in AL and RA. AL = Anthropised Luvisol, RA = Reconstructed Anthroposoil.



**Fig. S2.** Principal component analysis using values of water mass contents ( $W_{pF}$ ), Dexter indexes ( $S_{Dexter}$ ), apparent density ( $Da$ ), and useful reserve ( $UR$ ). Arrows correspond to variables associated to the first two axes. ■ = horizon H2 of an AL pit; ● = horizon H2 of a RA pit.

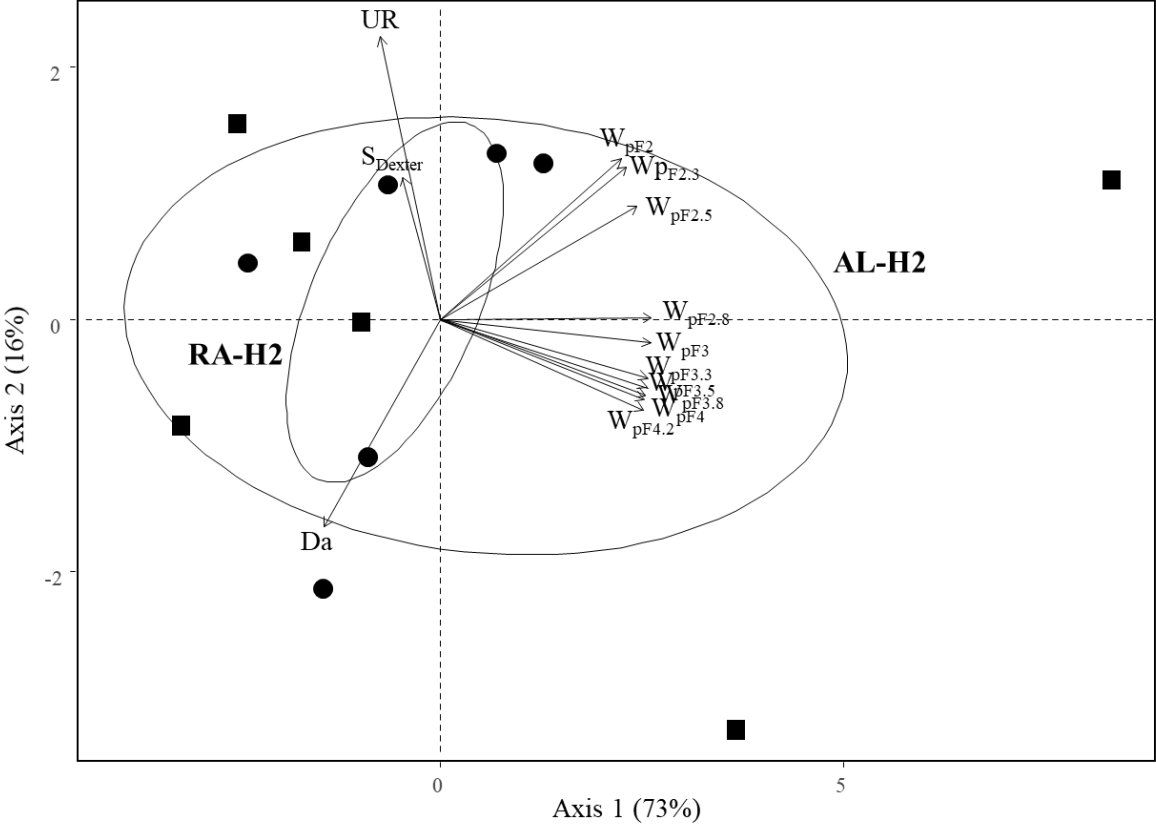
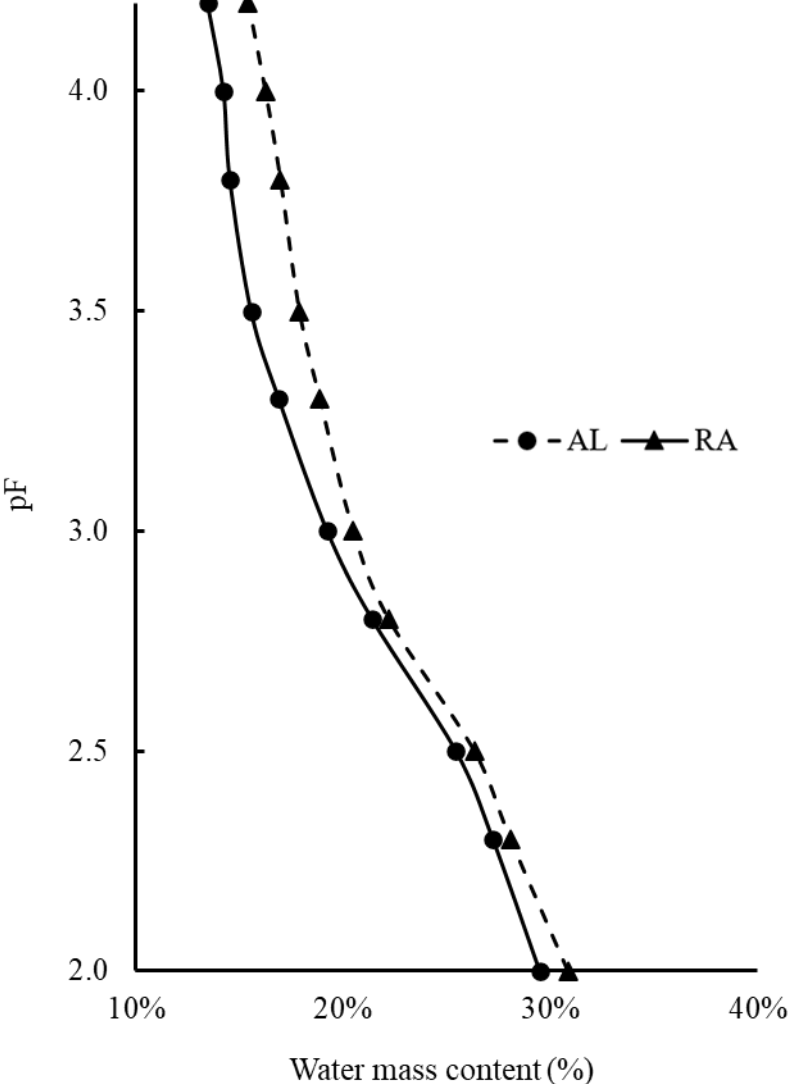


Fig. S3. Water retention curves of AL-H2 and RA-H2.



**Table S1** Description of study sites

Code	Soil type	Soil name (IUSS, 2007)	Land-use	Location	Initial stripping depth (cm)	Age (years)	Number of sections	Cumulated length (m)	Width (m)	Inter- tree (m)	Cover	Trees
AL	Anthropised Luvisol	Terric Luvisol (Transportic, Siltic)	Roadside	Leon Blum Avenue	30	30	3	800	2.45	10	Grass	<i>Acer platanoides</i>
RA	Reconstructed Anthroposoil	Hortic Anthrosol (Organotransportic, Siltic)	Roadside	Europe Avenue	150	20	3	570	1.8	7	Creeping ivy	<i>Tilia pallida</i>

**Table S2** Summary table of soil-structure patterns observed in the studied soil profiles (adapted from Piron et al., 2012). \*Sc replaces

Ac pattern and includes both natural and anthropogenic soil compaction processes.

Soil-structure pattern	Morphological characteristics	Code (Piron et al., 2012)
<b>CAST =</b>	Packing of cast aggregates of homogeneous shape and size, individual and not welded. High roughness aspect and high porosity.	<b>CASTg</b>
CASTg + CASTw + CASTc	Packing of welded cast aggregate. High roughness aspect and average porosity.	<b>CASTw</b>
	Packing of strongly welded casts. Smooth aspect.	<b>CASTc</b>
<b>BUR</b>	Earthworm burrow	<b>BUR</b>
<b>BULK =</b>	Structure without an aggregative aspect. Porosity is still visible but not composed of large macropores. Average roughness aspect.	<b>S</b>
S + Sc	Compacted area without discernible porosity. Low roughness.	<b>Sc*</b>

**Table S3** Percentages of roots, cast and bulk soil patterns per soil type and soil horizon from six pits of AL and 9 pits of RA. Values are mean  $\pm$  standard deviation.

Properties		Anthropised Luvisols			Reconstructed Anthroposoils		
		AL-H1 (n = 6)	AL-H2 (n = 6)	AL-H3 (n = 6)	RA-H1 (n = 9)	RA-H2 (n = 9)	RA-H3 (n = 9)
<i>Roots</i>	ROOT	72.4 $\pm$ 16.5	33.5 $\pm$ 19.9	16.3 $\pm$ 8.7	52.7 $\pm$ 16.1	45.2 $\pm$ 11.8	15.5 $\pm$ 6.0
<i>Burrows</i>	BUR	0.4 $\pm$ 0.5	1.0 $\pm$ 0.9	0.9 $\pm$ 1.5	4.1 $\pm$ 4.7	6.8 $\pm$ 4.6	2.4 $\pm$ 2.0
<i>Cast patterns</i>	<u>CAST</u>	64.6 $\pm$ 5.8	74.8 $\pm$ 16.1	42.9 $\pm$ 18.6	74.1 $\pm$ 15.0	73.3 $\pm$ 10.0	62.7 $\pm$ 19.8
	CASTg	0.0 $\pm$ 0.1	0.7 $\pm$ 0.7	0.0 $\pm$ 0.1	0.3 $\pm$ 0.5	1.4 $\pm$ 1.9	0.8 $\pm$ 1.5
	CASTw	23.5 $\pm$ 15.4	17.1 $\pm$ 10.4	3.8 $\pm$ 2.9	52.7 $\pm$ 8.1	33.2 $\pm$ 8.3	11.3 $\pm$ 6.8
	CASTc	41.1 $\pm$ 14.2	56.9 $\pm$ 17.1	39.1 $\pm$ 17.1	21.2 $\pm$ 15.0	38.7 $\pm$ 5.3	50.6 $\pm$ 13.4
<i>Bulk soil patterns</i>	<u>BULK</u>	35.4 $\pm$ 5.8	25.2 $\pm$ 16.1	57.1 $\pm$ 18.6	25.9 $\pm$ 15.0	26.7 $\pm$ 10.0	37.3 $\pm$ 19.8
	S	35.4 $\pm$ 5.8	21.8 $\pm$ 12.2	18.2 $\pm$ 23.5	25.9 $\pm$ 15.0	26.7 $\pm$ 10.0	37.3 $\pm$ 19.8
	Sc	0.0 $\pm$ 0.0	3.4 $\pm$ 5.9	39.0 $\pm$ 34.5	0.0 $\pm$ 0.0	0.0 $\pm$ 0.0	0.0 $\pm$ 0.0

**Table S4** Values of physical, chemical, and biological properties per soil type and soil horizon from samples of three AL pits and three RA pits.

<i>Properties</i>		<b>Anthropised Luvisols</b>			<b>Reconstructed Anthroposoils</b>		
		<b>AL-H1 (n = 3)</b>	<b>AL-H2 (n = 3)</b>	<b>AL-H3 (n = 3)</b>	<b>RA-H1 (n = 3)</b>	<b>RA-H2 (n = 3)</b>	<b>RA-H3 (n = 3)</b>
<i>Physical</i>	Clay	17.8 ± 1.1	22.1 ± 3.7	25.2 ± 3.1	15.3 ± 1.0	18.0 ± 0.8	19.6 ± 3.1
	Silt	65.5 ± 2.8	65.5 ± 7.7	66.8 ± 12.0	63.5 ± 1.6	70.5 ± 1.2	70.8 ± 7.2
	Sand	11.0 ± 3.7	11.0 ± 7.6	7.5 ± 9.4	12.6 ± 2.2	7.6 ± 0.9	8.0 ± 3.7
	EC	0.12 ± 0.03	0.11 ± 0.03	0.08 ± 0.03	0.14 ± 0.03	0.11 ± 0.01	0.12 ± 0.05
<i>Chemical</i>	pH	6.7 ± 0.2	7.9 ± 0.5	8.1 ± 0.2	6.9 ± 0.2	7.3 ± 0.1	8.0 ± 0.7
	Corg	3.3 ± 0.2	0.8 ± 0.1	0.3 ± 0.1	5.0 ± 0.6	2.3 ± 0.2	0.9 ± 0.2
	Ntot	0.23 ± 0.02	0.09 ± 0.01	0.04 ± 0.01	0.31 ± 0.04	0.18 ± 0.02	0.11 ± 0.02
	C:N	14.1 ± 0.3	9.7 ± 0.5	7.6 ± 0.5	16.3 ± 0.6	12.8 ± 0.21	8.8 ± 3.6
	P <sub>2</sub> O <sub>5</sub>	48.1 ± 13.8	26.1 ± 9.9	19.4 ± 7.0	103.1 ± 11.8	84.5 ± 3.4	51.9 ± 23.9
	CEC	15.2 ± 0.8	11.2 ± 1.2	11.4 ± 0.2	23.2 ± 4.4	14.8 ± 1.2	10.4 ± 0.9
	CaO	4088.7 ± 44.0	5316.7 ± 2662.7	4406.3 ± 1221.3	6036.7 ± 1066.1	4697.7 ± 652.6	6209.0 ± 4235.2
	K <sub>2</sub> O	301.0 ± 54.6	124.3 ± 7.0	112.7 ± 26.3	203.3 ± 55.6	125.3 ± 18.2	168.0 ± 18.2
	MgO	215.0 ± 35.2	101.1 ± 20.0	141.0 ± 30.5	316.0 ± 97.5	145.0 ± 33.8	179.7 ± 31.9
	Na <sub>2</sub> O	90.0 ± 31.7	82.0 ± 45.3	84.3 ± 49.7	98.3 ± 66.4	121.3 ± 75.0	107.0 ± 86.9
	Ca:CEC	96.5 ± 5.1	133.2 ± 16.8	135.7 ± 15.8	96.4 ± 29.8	113.1 ± 6.8	128.3 ± 37.6
	<i>Biological</i>	MB	935.7 ± 230.5	140.7 ± 80.8	-	1016.0 ± 185.0	415.3 ± 54.0
SIR		19.6 ± 5.7	3.6 ± 0.7	-	17.3 ± 9.5	9.5 ± 4.5	-
DEA		1.16 ± 0.19	0.38 ± 0.23	-	1.22 ± 0.54	0.59 ± 0.18	-
NEA		0.30 ± 0.08	0.39 ± 0.16	-	0.11 ± 0.10	0.25 ± 0.13	-

**Table S5** Values of water mass content at different pF values ( $W_{pF}$ ; %), apparent density (Da; g.cm-3), water reserve (WR; mm/cm), and Dexter index ( $S_{Dexter}$ ). Values are mean  $\pm$  standard deviation.

	<b>AL-H2</b> <b>(n = 6)</b>	<b>RA-H2</b> <b>(n = 6)</b>
$W_{pF2}$	0.31 $\pm$ 0.06	0.29 $\pm$ 0.03
$W_{pF2.3}$	0.28 $\pm$ 0.06	0.27 $\pm$ 0.03
$W_{pF2.5}$	0.26 $\pm$ 0.06	0.26 $\pm$ 0.02
$W_{pF2.8}$	0.22 $\pm$ 0.06	0.21 $\pm$ 0.02
$W_{pF3}$	0.21 $\pm$ 0.06	0.19 $\pm$ 0.02
$W_{pF3.3}$	0.19 $\pm$ 0.06	0.17 $\pm$ 0.01
$W_{pF3.5}$	0.18 $\pm$ 0.06	0.16 $\pm$ 0.02
$W_{pF3.8}$	0.17 $\pm$ 0.06	0.15 $\pm$ 0.02
$W_{pF4}$	0.17 $\pm$ 0.06	0.14 $\pm$ 0.01
$W_{pF4.2}$	0.16 $\pm$ 0.06	0.14 $\pm$ 0.01
Da	1.31 $\pm$ 0.07	1.30 $\pm$ 0.10
WR	1.43 $\pm$ 0.43	1.54 $\pm$ 0.17
$S_{Dexter}$	0.0663 $\pm$ 0.0216	0.0551 $\pm$ 0.0122



**CHAPITRE 3 - Impacts des pratiques d'ingénierie pédologique  
et des surfaces asphaltées sur les communautés lombriciennes**

## **Objectifs de l'étude**

Les objectifs de cette étude étaient de (i) caractériser les communautés lombriciennes dans des Anthrosols d'accompagnement de voirie reconstitués à deux périodes différentes ainsi que dans des prairies pseudo-naturelles situées à proximité, et (ii) d'évaluer les impacts sur les communautés lombriciennes de deux types de surface asphaltées séparant les Anthrosols de potentiels réservoirs.

## **Méthodologie**

Les sites d'étude sont les mêmes que ceux proposés dans le chapitre 1, à savoir des Anthrosols de 4 et 20 ans reconstitués de manière similaire. Les vers de terre ont été échantillonnés manuellement par extraction de blocs de sol de manière à couvrir l'ensemble des linéaires de sol étudiés (54 blocs échantillonnés par type d'Anthrosol et 60 blocs pour l'ensemble des prairies). Au sein des Anthrosols, les prélèvements ont lieu au sein de tronçons (i) partiellement isolés par rapport à un potentiel réservoir (i.e., séparation par un trottoir de 3 m de large), et (ii) totalement isolés par rapport à un potentiel réservoir (i.e., séparation par une route de 6 m de large).

## **Principaux résultats**

Le processus d'ingénierie pédologique menant à des Anthrosols reconstitués a globalement négativement impacté la structure des communautés lombriciennes, favorisant une prépondérance significative des espèces endogées représentant 90% de l'abondance totale tous sols confondus. Dans le cas des sols de 20 ans, les paramètres lombriciens (abondance, biomasse, richesse totales) étaient significativement inférieurs dans les sols totalement isolés par rapport aux sols partiellement isolés, avec notamment l'absence de trois catégories

écologiques (épigés, épi-anéciques, anéciques stricts). Aucune différence n'a été observée dans les sols de 4 ans en en fonction du paramètre d'isolement.

**Title: Earthworm communities of reconstructed soils in urban areas are impacted by soil engineering processes and asphalt surfaces**

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## **Abstract**

Providing soil ecosystem services such as temperature and water regulation is a key issue in cities for the well-being of citizens. To optimise urban soil functions, one solution offered by soil engineering is to excavate deep trenches in existing urban soils and to reconstruct soils based on the model of natural soils. However, this process involves soil stripping and stockpiling which is particularly detrimental for earthworm communities, considered as key actors of soil restoration. Moreover, urban development is associated with landscape fragmentation by asphalt surfaces such as roads whose negative impacts have been highlighted on many organisms, but their consequences on earthworm communities of reconstructed soils have been little studied. In that respect, this study aimed (i) to evaluate earthworm communities between surrounding pseudo-natural grassland soils and in a recent (4-year-old) and an old (20-year-old) reconstructed roadside soil, and (ii) to evaluate in each reconstructed roadside soil the impacts of asphalt surfaces surrounding them on earthworm communities. We studied an urban landscape in the Paris region, France. We sampled earthworms along two linear reconstructed roadside soils and in surrounding pseudo-natural grassland soils. Reconstructed roadside soils were separated from a potential reservoir either by an asphalt sidewalk of 3 metre wide with no curb or by an asphalt road of 6 metre wide with a 20 cm high curb. Soil engineering processes negatively impacted the structure of earthworm communities of reconstructed roadside soils as endogeic species represented 90% of the total abundance. Regarding the effect of asphalt surfaces in the 20-year-old reconstructed roadside soil, earthworm parameters were significantly lower and three ecological categories were absent in soils bordered by a wide asphalt road in comparison with soils bordered by a narrow sidewalk. No differences were observed in 4-year-old reconstructed roadside soils regardless of the nearby asphalt surface. Overall, we highlighted that soil engineering processes and asphalt surfaces characteristics may

be key elements to consider in urban planning to enhance earthworm communities and ecosystem services associated with urban soils.

**Keywords: urbanisation, soil artificialisation, landscape fragmentation**

## 1. Introduction

Urban soils play an essential role in the well-being of citizens, especially unsealed soils (e.g., vegetated pseudo-natural or engineered soils) which ensure many functions and ecosystem services (Morel et al., 2015; Ugolini et al., 2020). For example, they take part in the regulation of water flows through infiltration and retention, and in the regulation of urban microclimate through the vegetation that grows in green spaces (e.g., parks and gardens; Gómez-Baggethun and Barton, 2013; Calzolari et al., 2020). However, maintaining soil functions is a key issue in urban contexts but can be challenging due to the soil degradation caused by urban development (e.g., soil compaction by construction machinery, soil stripping, accumulation of stones and building materials; Craul, 1985; Pavao-Zuckerman, 2008). Thus, various soil engineering strategies (i.e., from basic to advanced soil engineering strategies; Maréchal et al., 2021) are developed by urban planners depending on their recognition and citizens' awareness of functions provided by urban soils (Poyat, 2018). One of the strategies is to reconstruct soils on the model of natural soils to achieve similar levels of soil functionality. Firstly, deep trenches are dug in existing urban soils (at least 150 cm deep) to remove unfavourable material for vegetation (e.g., soils with many artefacts, high bulk density; Lehmann and Stahr, 2007). Secondly, trenches are filled with two complementary layers of topsoil (i.e., organo-mineral material) which have been stripped, stockpiled, transported, and amended with green waste compost for the surface layer (Marié and Rossignol, 1997; Unep, 2012). In all soil engineering strategies, soil fauna, and earthworms in particular, are well recognised as key actors to improve soil properties and accelerate soil restoration (Scullion and Malik, 2000; Frouz et al., 2006; Boyer and Wratten, 2010).

Earthworm communities in recently reconstructed soils based on the model of natural soils are subject to different soil engineering processes which can impact them. For example, soil stripping and stockpiling achieved by mechanised handling destroys earthworm habitat and

kills individuals, leading to a collapse of earthworm abundance and biomass (Scullion et al., 1988; Brun et al., 1991). Besides, soil stockpiling has adverse effects on earthworms due to, among others, low organic matter content (Pižl, 2001; Edwards, 2004), and anaerobic conditions caused by compaction (Abdul-Kareem and McRae, 1984; Boyer and Wratten, 2010). On the contrary, some practices, such as avoiding mixing topsoil and subsoil with machinery or incorporating green waste compost into the soil, improve physicochemical conditions and food supply which can enhance earthworm abundance and individual biomass (Abdul-Kareem and McRae, 1984; Ashwood et al., 2017). However, most studies on the recovery of earthworm communities after such soil engineering processes have been carried out in post-mining sites but none of them have been achieved in urban contexts (Armstrong and Bragg, 1984; Hüttl and Weber, 2001; Russell et al., 2016). Indeed, in urban contexts, the recovery of earthworm communities could be depending on individuals that survived through soil engineering processes, but also on active and passive earthworm dispersal from a potential surrounding reservoir (Pižl, 2001; Eijsackers, 2010, 2011). Earthworm active and passive dispersal could be compromised by the fragmentation of urban landscape due to anthropic infrastructures (e.g., buildings, roads; Collinge, 2009) and could be dependent on earthworm species (Eijsackers, 2011). For example, asphalt surfaces such as roads can constitute obstacles for earthworm displacement (Dupont et al., 2017), as well as they can induce high evapotranspiration rates resulting in soil dryness that is detrimental to earthworms (Tenenbaum et al., 2006; Pouyat et al., 2010). As far as we know, no studies focused on the impacts of asphalt surfaces on earthworm dispersal between reconstructed soils and potential surrounding reservoirs.

Firstly, we aimed to assess earthworm communities in a recent (4-year-old) and an old (20-year-old) linear reconstructed roadside, and surrounding pseudo-natural grassland soils and soil in the Paris region. We expected a lower earthworm abundance, biomass, diversity, and individual biomass in reconstructed roadside soils than in pseudo-natural grassland soils due to



disturbances caused by soil engineering. Secondly, we aimed to assess in each reconstructed soil, the impacts of two types of asphalt surfaces separating them from a potential surrounding reservoir, one by a narrow asphalt sidewalk of 3 metre wide without curb and another by a wide asphalt road of 6 metre with a 20 cm high curb on earthworm communities. We hypothesized lower abundance, biomass, diversity, and individual biomass in reconstructed roadside soils separated from a potential reservoir by a wide asphalt road than a narrow asphalt sidewalk due to more adverse conditions for dispersal.

## **2. Materials and methods**

### ***2.1. Study area***

The study sites were located in the Paris region (France) and were divided between Palaiseau (48° 71' 42'' N; 2° 19' 64'' E) and Saint-Quentin-en-Yvelines (48° 75' 75'' N; 2° 07' 25'' E) cities. The substratum corresponds to thick loess deposits (> 3m) (Mégnyen, 1989). The temperate climate of the region is associated with mean annual precipitation and temperature of 563 mm and 11.2°C, respectively (Météo France, 2008-2018).

We selected (i) a 4-year-old linear roadside soil (4RS) constructed in 2015 along the Gaspar Monge Boulevard in Palaiseau, and (ii) a 20-year-old linear roadside soil (20RS) constructed in 1999 along the Europe Avenue in Saint-Quentin-en-Yvelines. Each linear roadside soil was reconstructed following a similar engineering process prescribed by the same company. Firstly, trenches of 150 cm deep were excavated in urban soils present before the reconstruction. Secondly, 125 cm of stripped and stockpiled silty topsoil (organo-mineral material) was added in the trenches. In both 4RS and 20RS, the topsoil used for the soil reconstruction was the result of stripping in dry conditions of 30 cm of soil in the right-of-way of the development project. Stripped topsoil was stockpiled for 2 years in stocks over 2.5 metres high. Thirdly, a 25 cm-thick surface horizon of topsoil mixed with green-waste compost was implemented. Finally, an

additional layer of 5 cm of organic mulch (respectively green waste shreds for 4RS, and pine barks for 20RS) and a permanent plant cover were implemented on the surface. The cumulated length of 4RS and 20RS sections were 970 m and 790 m, respectively. 4RS were planted with 3 main tree species (*Quercus robur*, *Carpinus betulus* *Fagus sylvatica*,) every 4 metres, and only one tree species (*Tilia pallida*) was planted in 20RS every 7 metres. Details on reconstructed roadside soil design are given in Table S1. In addition, 10 pseudo-natural grassland soils (PNG) were selected as controls within an 8 km radius and according to the access authorisation by the managers. Selected PNG were left untouched by any disturbances for at least 20 years (e.g., natural reserve, old urban parks) which was confirmed by the analysis of historical aerial photographs and preliminary soil profile observations achieved with a soil auger. In addition, 9 soil profile descriptions were performed in 4RS, 9 in 20RS, and one in each PNG (NF EN ISO 25177; AFNOR, 2019) by digging 110 cm deep soil pits. Both, 4RS and 20RS soils were named Hortic Anthrosols (Organotransportic, Siltic), and PNG were named Haplic Luvisols (WRB, 2015; Table S1).

Within each linear reconstructed roadside soil, we selected sections separated from a potential surrounding reservoir by two types of asphalted surfaces: (i) either by a narrow asphalt sidewalk (3-metre-wide) with no curb (4RS-NAS and 20RS-NAS, respectively); (ii) or by a wide asphalt road (6-metre-wide) bordered by a curb of 20 cm high (4RS-WAR and 20RS-WAR, respectively; Fig. S1). The depth of road infrastructure is greater (approx. > 50 cm) than sidewalks (approx. < 20 cm; Packard, 1984). We sampled earthworms and soils all along the selected roadside sections. Thus, we studied 5 groups with different number of replicates for each group depending on the number of intersections and length of the sections: (i) PNG (n=10), (ii) 4RS-NAS (n=9), (iii) 4RS-WAR (n=9), (iv) 20RS-NAS (n=6), and (v) 20RS-WAR (n=12) (Table S1; Fig.S1).

## ***2.2. Earthworm sampling and identification***

We sampled earthworms within PNG, 4RS, and 20RS in spring 2019. Blocks of soil (20 cm × 20 cm × 25 cm, length × width × depth) were extracted and hand-sorted to collect earthworms. In 4RS and 20RS, one replicate corresponded to the extraction of 3 blocks of soil between two consecutive trees (i.e., inter-tree) of the same species. In order to balance the sampling between the 3 tree species present in 4RS, 3 inter-trees of each tree species were selected in NAS as well as in WAR (i.e., 9 inter-trees in 4RS-NAS, and 9 inter-trees in 4RS-WAR). Blocks were equidistant between them and located in the middle of the tree trench (Fig. S1). In PNG, one replicate corresponded to the extraction of 6 blocks of soil in the diagonal of a 3 m x 15 m rectangle, located at least 10 m from any edge (e.g., hedge, path; Fig. S1). A total of 54 blocks of soil were extracted in each reconstructed roadside soil and 60 blocks of soil for the 10 PNG were extracted. A 96% ethanol solution was used to store earthworms. Each earthworm was counted, weighed (fresh weight with gut content, ± 10 mg), associated with a stage of development (juvenile, sub-adult and adult), an ecological category (epigeic, epianecic, anecic-strict or endogeic; Bouché, 1972, 1977; Jégou et al., 1998; Hoeffner et al., 2019) and a sub-species. If the sub-species of juveniles was indeterminate, we limited in a first step the identification to the genus. In a second step, juveniles were distributed into sub-species according to the proportions of sub-adults and adults present on each replicate. To obtain an estimation per square metre, earthworm total abundance and biomass, and abundance and biomass per ecological category were multiplied by 25. We calculated the total species richness and the species evenness index ( $J' = \frac{H'}{H'_{max}}$ , with H' as Shannon diversity index) to analyse earthworm diversity.

## ***2.3. Soil sampling and physicochemical properties***

For each reconstructed roadside soil, soil samples were composed of the blocks extracted for earthworm sampling (between 0 and 25 cm deep). To constitute 9 replicates per reconstructed roadside soil, two series of 3 blocks were grouped regardless of the type of the asphalt surfaces bordering reconstructed soils. Regarding pseudo-natural grassland soils, 1 composite of sub-samples from the 6 blocks extracted per site was sent for analysis at AUREA Agrosociences laboratory (Orléans, France). The analyses consisted of measurements of pH (water; NF ISO 10390; AFNOR, 2005), soil organic matter (SOM; NF ISO 14235; AFNOR, 1998a), total nitrogen ( $N_{\text{tot}}$ ; NF ISO 13878; AFNOR, 1998b), cation exchangeable capacity (CEC; NFX 31–130; AFNOR, 1999), and particle-size distribution (clay  $<2 \mu\text{m}$ ; silt  $2 < \mu\text{m} < 50$ ; sand  $50 < \mu\text{m} < 200$ , %; NFX 31–107 without decarbonatation; AFNOR, 2003). In comparison with 4RS and 20RS, PNG had the lowest physicochemical values (between -2 and -16%; Table 1), except for clay and sand percentages which were the lowest in 20RS and 4RS respectively (at least -18% and -34% respectively; Table1). Regarding reconstructed roadside soils, 20RS had higher values of SOM,  $N_{\text{tot}}$ , C/N, and CEC than 4RS (at least +20%; Table 1).

**Table 1** Main physicochemical characteristics of reconstructed roadside soils (4RS and 20RS) and pseudo-natural grassland soils (PNG). Values are mean  $\pm$  standard deviation. SOM = soil organic matter;  $N_{\text{tot}}$  = total nitrogen; C:N = organic carbon / total nitrogen ration ratio; CEC = cation exchangeable capacity.

Code	pH	SOM (%)	$N_{\text{tot}}$ (%)	C:N	CEC (meq/100g)	Clay (%)	Silt (%)	Sand (%)
4RS	8.2 $\pm$ 0.1	3.4 $\pm$ 0.5	0.19 $\pm$ 0.02	10.7 $\pm$ 0.9	14.1 $\pm$ 0.8	21.5 $\pm$ 1.0	67.7 $\pm$ 1.4	7.3 $\pm$ 0.4
20RS	7.4 $\pm$ 0.2	8.5 $\pm$ 1.3	0.38 $\pm$ 0.09	13.5 $\pm$ 1.1	23.5 $\pm$ 2.8	16.1 $\pm$ 1.6	63.9 $\pm$ 2.0	11.1 $\pm$ 1.4
PNG	6.5 $\pm$ 0.4	2.9 $\pm$ 0.7	0.16 $\pm$ 0.03	10.4 $\pm$ 0.9	12.0 $\pm$ 1.9	19.6 $\pm$ 2.1	62.4 $\pm$ 15.0	15.1 $\pm$ 13.9

## ***2.4. Statistical analyses***

As reconstructed roadside soils studied (4RS and 20RS) were designed similarly by the same company but at different times and locations, statistical analyses focused on comparisons within each reconstructed roadside soil (respectively 4RS-NAS vs 4RS-WAR, and 20RS-NAS vs 20RS-WAR) without attempting to compare reconstructed roadside soils to each other. Before analyses, conditions of normality and homoscedasticity were checked to apply adapted tests. We applied Wilcoxon tests for non-parametric data to evaluate total abundance, total biomass, species richness, species evenness index, and abundance, biomass, species richness of ecological categories between PNG and all reconstructed roadside soils combined (RS). We also applied Wilcoxon tests to compare the same earthworm parameters between 4RS-NAS and 4RS-WAR, and between 20RS-NAS and 20RS-WAR. We performed an NMDS analysis calculated with a dissimilarity matrix (Bray-Curtis distances) to compare the species composition of earthworm communities among 4RS-NAS, 4RS-WAR, 20RS-NAS, and 20RS-WAR. Regarding earthworm individual biomass, only adults of earthworm species present in at least 3 blocks of 4RS-NAS, 4RS-WAR, 20RS-NAS and 20RS-WAR were selected. Then, we compared species by species differences in earthworm individual biomass. We used Wilcoxon tests to compare the same earthworm species between 4RS-NAS and 4RS-WAR, and between 20RS-NAS and 20RS-WAR (Table S2). All statistical analyses were performed using R software (R Core Team, 2019). In all cases, significance was evaluated at  $P < 0.05$ .

## **3. Results**

Regarding pseudo-natural grassland soils (PNG), total earthworm biomass and species richness were as expected significantly higher than in all reconstructed roadside soils (RS) combined (respectively  $112 \pm 47$  and  $60 \pm 41$  g.m<sup>-2</sup>;  $7 \pm 1$  and  $5 \pm 2$  mean earthworm species;  $W = 62.5$  and  $42.0$ ,  $P < 0.002$ ; Table 2) due to a higher epigeic, epi-anecic, and strict-anecic

earthworm biomass and species richness (at least +28%;  $W = 5.0$  to  $106.0$ ,  $P < 0.039$ ; Table 2). Epigeic, epi-aneic, and strict-aneic earthworm abundance were also higher in PNG than in RS (at least +5%;  $W = 7.0$  to  $107.5$ ,  $P < 0.026$ ; Table 2). Contrary to our hypothesis, total abundance and species evenness index were not significantly different between PNG and RS (respectively  $354 \pm 101$  and  $296 \pm 228$  individuals  $m^{-2}$ ;  $0.8 \pm 0.1$  and  $0.7 \pm 0.2$ ;  $W = 116.0$  and  $208.5$ ,  $P > 0.091$ ; Table 2), as well as endogeic abundance, biomass, and species richness ( $W = 143.0$  to  $182.5$ ,  $P > 0.313$ ; Table 2). Across all PNG and RS sites, 19 species of earthworms were observed (respectively 14 in and 15 species in PNG and RS), including 10 species common to both PNG and RS. Four species were only present in PNG (*Eiseniella tetraedra*, *Satchellius mammalis*, *Lumbricus festivus*, and *Allolobophora minima*), and 5 species only present in RS (*Aporrectodea caliginosa meridionalis*, *Allolobophora chlorotica poststepheba*, *Allolobophora limicola*, *Octolasion cyaneum*, and *Octolasion lacteum lacteum*; Table S2). Overall, endogeic abundance represented 71% and 90% of the total abundance in PNG and RS respectively. Regarding adult individual biomass, *Allolobophora rosea rosea* was significantly heavier in RS than in PNG (+19%,  $W = 1454$ ,  $P = 0.032$ ; Table S3) while no significant difference was observed for *Aporrectodea caliginosa caliginosa* ( $W = 653.5$ ,  $P = 0.655$ ; Table S3).

**Table 2** Earthworm total abundance (individuals  $m^{-2}$ ), total biomass ( $g \cdot m^{-2}$ ), species richness, species evenness index, and abundance, biomass and species richness per ecological category in PNG ( $n = 10$ ) and RS ( $n = 36$ ). Values are mean  $\pm$  standard deviation. Different letters denote significant differences among groups with  $a > b$ . W-values and p-values were given from Wilcoxon tests. PNG = pseudo-natural grassland soils; RS = reconstructed roadside soils.

	<b>PNG</b>	<b>RS</b>	<b>Wilcoxon</b>	<b>p-value</b>
<b>Total abundance</b>	<b>354 ± 101 a</b>	<b>296 ± 228 a</b>	<b>W = 116.0</b>	<b>0.091</b>
Epigeic abundance	40 ± 21 a	21 ± 41 b	W = 77.5	0.005
Epi-aneic abundance	6 ± 5 a	6 ± 13 b	W = 107.5	0.026
Strict-aneic abundance	59 ± 24 a	4 ± 12 b	W = 7.0	<0.001
Endogeic abundance	253 ± 99 a	265 ± 188 a	W = 160.0	0.603
<b>Total biomass</b>	<b>112.4 ± 47.1 a</b>	<b>60.2 ± 41.3 b</b>	<b>W = 62.5</b>	<b>0.002</b>
Epigeic biomass	2.0 ± 1.1 a	1.8 ± 4.3 b	W = 105.5	0.039
Epi-aneic biomass	3.2 ± 4.4 a	2.3 ± 6.2 b	W = 106.0	0.021
Strict-aneic biomass	55.3 ± 24.7 a	4.0 ± 10.2 b	W = 5.0	<0.001
Endogeic biomass	52.2 ± 30.0 a	52.2 ± 29.9 a	W = 182.5	0.958
<b>Species richness</b>	<b>7.4 ± 1.0 a</b>	<b>4.5 ± 2.3 b</b>	<b>W = 42.0</b>	<b>&lt;0.001</b>
Epigeic richness	1.7 ± 0.5 a	0.7 ± 0.8 b	W = 60.5	<0.001
Epi-aneic richness	0.8 ± 0.4 a	0.3 ± 0.6 b	W = 93.0	0.007
Strict-aneic richness	1.3 ± 0.5 a	0.2 ± 0.5 b	W = 29.5	<0.001
Endogeic richness	3.6 ± 0.8 a	3.3 ± 1.2 a	W = 143.0	0.313
<b>Species evenness index</b>	<b>0.8 ± 0.1 a</b>	<b>0.7 ± 0.2 a</b>	<b>W = 208.5</b>	<b>0.455</b>

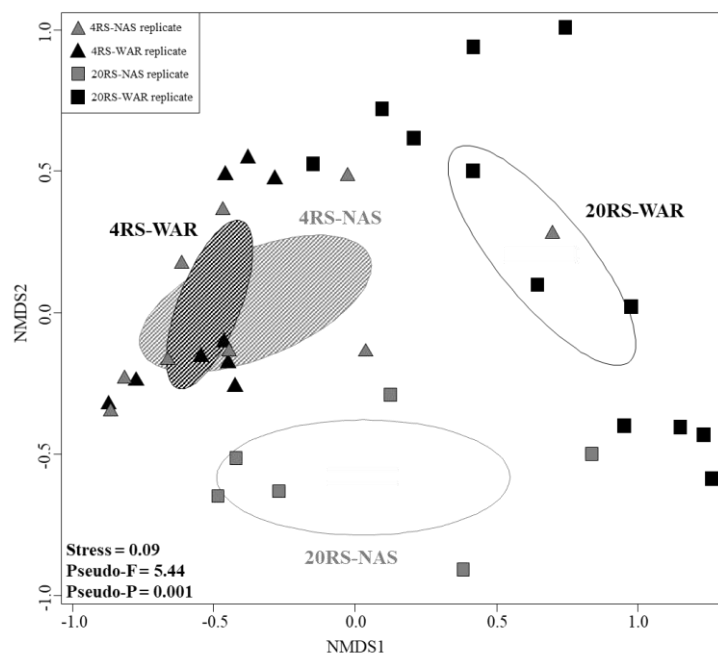
Regarding 20RS, total earthworm abundance, total biomass, and species richness were significantly higher in NAS than in WAR in accordance with our hypothesis (respectively  $513 \pm 343$  and  $135 \pm 60$  individuals  $m^{-2}$ ;  $113.9 \pm 63.3$  and  $42.4 \pm 20.0$   $g \cdot m^{-2}$ ;  $7.2 \pm 2.6$  and  $2.5 \pm 0.8$  species;  $W = 0.5$  to  $11.0$ ,  $P < 0.035$ ; Table 3), as epigeic, epi-aneic, and strict-aneic earthworms were absent in WAR (Table 3). In addition, endogeic abundance, biomass, and species richness were significantly higher in NAS than in WAR (at least +40%,  $W = 0.5$  to  $13.0$ ,  $P < 0.035$ ; Table 3). Overall, the structure of earthworm communities was significantly different between NAS and WAR ( $Pseudo-P < 0.001$ ; Fig.1). Among the 12 species present in NAS, only 4 species were present in WAR (*Aporrectodea caliginosa caliginosa*, *Allolobophora icterica*, *Allolobophora rosea rosea*, and *Octolasion cyaneum*; Table S2). Their abundance was similar in NAS and WAR, excepted *Aporrectodea caliginosa caliginosa* which was less abundant in WAR (-93%) but had a significantly higher adult individual biomass in WAR than in NAS (+54%,  $t = -6.4$ ,  $P < 0.001$ ; Table S4). Similarly, adult individual biomass of *Allolobophora rosea rosea* was significantly higher in WAR compared to NAS (+33%,  $t = -2.5$ ,  $P = 0.015$ ; Table S4). Besides, *Allolobophora rosea rosea* represented in association with *Allolobophora icterica* 91% of the total abundance in WAR against 26% in NAS (Table S2).

Regarding 4RS, no significant differences were observed between NAS and WAR in terms of total earthworm abundance, total biomass, species richness, and species evenness index contrary to our hypothesis ( $W = 43.0$  to  $55.0$ ,  $P > 0.222$ ; Table 3). Epigeic, epi-anecic, strict-anecic, and endogeic abundance, biomass, species richness, and structure of communities were also not significantly different between NAS and WAR ( $W = 34.5$  to  $59.5$ ,  $P > 0.102$ ; Table 3 and Fig.1). All species common to NAS and WAR were present in similar abundance, excepted *Allolobophora chlorotica chlorotica* which was more abundant in WAR (+62%). Overall, two endogeic species, *Aporrectodea caliginosa caliginosa* and *Allolobophora rosea rosea*, were the most abundant in both NAS and WAR (respectively 77% and 75% of the total abundance; Table S2) and their adult individual biomass was not significantly different between NAS and WAR ( $t = 0.4$  to  $1.5$ ,  $P > 0.142$ ; Table S4). On the contrary, *Allolobophora chlorotica poststepheba* and *Octolasion lacteum lacteum* occurred only one time in low abundance ( $\leq 2$  individuals  $m^{-2}$ ) and only in NAS.

**Table 3** Earthworm total abundance (individuals  $m^{-2}$ ), biomass ( $g \cdot m^{-2}$ ), species richness, species evenness index, and abundance, biomass and species richness per ecological category in 4RS-NAS ( $n = 9$ ), 4RS-WAR ( $n = 9$ ), 20RS-NAS ( $n = 6$ ), 20RS-WAR ( $n = 12$ ). Values are mean  $\pm$  standard deviation. Different letters denote significant differences among groups with  $a > b$ . W-values and p-values were given from Wilcoxon tests between 4RS-NAS and 4RS-WAR, and between 20RS-NAS, 20RS-WAR. 4RS-NAS = 4-year-old reconstructed roadside soils bordered by a narrow asphalt sidewalk; 4RS-WAR = 4-year-old reconstructed roadside soils bordered by a wide asphalt road; 20RS-NAS = 20-year-old reconstructed roadside soils bordered by a narrow asphalt sidewalk; 20RS-WAR = 20-year-old reconstructed roadside soils bordered by a wide asphalt road.



	<b>4RS-NAS</b>	<b>4RS-WAR</b>	<b>Wilcoxon</b>	<b>p-value</b>	<b>20RS-NAS</b>	<b>20RS-WAR</b>	<b>Wilcoxon</b>	<b>p-value</b>
<b>Total abundance</b>	<b>306 ± 218 a</b>	<b>359 ± 148 a</b>	<b>W = 52.5</b>	<b>0.31</b>	<b>513 ± 343 a</b>	<b>135 ± 60 b</b>	<b>W = 11.0</b>	<b>0.022</b>
Epigeic abundance	17 ± 17 a	19 ± 22 a	W = 41.5	0.964	72 ± 80 a	0 ± 0 b	W = 0.0	<0.001
Epi-aneic abundance	7 ± 15 a	3 ± 4 a	W = 42.0	0.911	21 ± 19 a	0 ± 0 b	W = 12.0	0.003
Strict-aneic abundance	3 ± 6 a	5 ± 7 a	W = 45.5	0.614	14 ± 27 a	0 ± 0 b	W = 24.0	0.049
Endogeic abundance	279 ± 210 a	332 ± 133 a	W = 55.5	0.199	406 ± 257 a	135 ± 60 b	W = 11.5	0.024
<b>Total biomass</b>	<b>49.3 ± 34.3 a</b>	<b>59.4 ± 21.8 a</b>	<b>W = 55.0</b>	<b>0.222</b>	<b>113.9 ± 63.3 a</b>	<b>42.4 ± 20.0 b</b>	<b>W = 13.0</b>	<b>0.035</b>
Epigeic biomass	1.6 ± 1.8 a	1.4 ± 1.7 a	W = 38.0	0.856	6.7 ± 8.9 a	0.0 ± 0.0 b	W = 0.0	<0.001
Epi-aneic biomass	2.7 ± 6.1 a	0.4 ± 0.7 a	W = 42.0	0.911	8.9 ± 11.6 a	0.0 ± 0.0 b	W = 12	0.003
Strict-aneic biomass	2.2 ± 5.5 a	6.2 ± 11.8 a	W = 46.0	0.576	11.7 ± 18.6 a	0.0 ± 0.0 b	W = 24.0	0.049
Endogeic biomass	42.8 ± 31.1 a	51.4 ± 16.5 a	W = 59.5	0.102	86.6 ± 39.1 a	42.4 ± 20.0 b	W = 13.0	0.035
<b>Species richness</b>	<b>4.9 ± 2.0 a</b>	<b>4.9 ± 1.4 a</b>	<b>W = 43.0</b>	<b>0.857</b>	<b>7.2 ± 2.6 a</b>	<b>2.5 ± 0.8 b</b>	<b>W = 0.5</b>	<b>&lt;0.001</b>
Epigeic richness	1.0 ± 0.9 a	0.7 ± 0.8 a	W = 34.5	0.606	1.3 ± 0.5 a	0.0 ± 0.0 b	W = 0.0	<0.001
Epi-aneic richness	0.2 ± 0.4 a	0.3 ± 0.5 a	W = 45.0	0.649	1.2 ± 1.0 a	0.0 ± 0.0 b	W = 12.0	0.002
Strict-aneic richness	0.2 ± 0.2 a	0.3 ± 0.5 a	W = 45.0	0.649	0.5 ± 0.8 a	0.0 ± 0.0 b	W = 24.0	0.049
Endogeic richness	3.4 ± 1.2 a	3.4 ± 1.0 a	W = 43.0	0.853	4.2 ± 1.2 a	2.5 ± 0.8 b	W = 0.5	<0.001
<b>Species evenness index</b>	<b>0.7 ± 0.1 a</b>	<b>0.7 ± 0.2 a</b>	<b>W = 38.5</b>	<b>0.895</b>	<b>0.8 ± 0.1 a</b>	<b>0.6 ± 0.3 a</b>	<b>W = 54.5</b>	<b>0.092</b>



**Fig. 1** Non-metric multidimensional scaling ordinations showing similarities, based on Bray-Curtis distance index, of earthworm communities of each reconstructed roadside soil. Significant dissimilarities between groups were assessed by PERMANOVA. Pseudo-F values and associated P-values are indicated. Ellipses correspond to groups (4RS-NAS = 4-year-old reconstructed roadside soils bordered by a narrow asphalt sidewalk; 4RS-WAR = 4-year-old reconstructed roadside soils bordered by a wide asphalt road; 20RS-NAS = 20-year-old

reconstructed roadside soils bordered by a narrow asphalt sidewalk; 20RS-WAR = 20-year-old reconstructed roadside soils bordered by a wide asphalt road.).

## 4. Discussion

### *4.1. Reconstructed soils based on the model of natural soils are beneficial for earthworm communities, especially for endogeic earthworms*

Total earthworm abundance and total species richness were relatively similar in pseudo-natural grassland soils and both reconstructed roadside soils combined (respectively 354 and 296 individuals m<sup>-2</sup>; 14 and 15 species), and surprisingly higher than values observed in other studies in urban soils (Pižl and Schlaghamerský, 2007; Vergnes et al., 2017; Tóth et al., 2020). For example, Tiho and Josens (2000) observed in a long lawn surrounded by two roads located in Brussels a mean earthworm abundance of 152 individuals m<sup>-2</sup> and 9 species, and Xie et al. (2018) observed in 36 residential areas in Beijing a mean earthworm abundance of 45 individuals m<sup>-2</sup> and 11 species. The present study highlights the suitability of reconstructed habitats based on the model of natural soils to earthworm development until reaching values of local grasslands which are known to favour high earthworm abundance in temperate climates (Decaëns et al., 2008; Cluzeau et al., 2012). This is probably explained by the surface horizon made from a mixture of stripped topsoil and compost from green waste may provide sufficient food resources (e.g., 3.5% and 8.5% of organic matter in 4-year-old and 20-year-old reconstructed soils respectively; Lapied et al., 2009; Ashwood et al., 2017) and the presence of a permanent cover and organic mulch also provides food resources, shelter, and limits temperature fluctuations (Pélosi et al., 2009; Schreck et al., 2012). Besides, few recent soil disturbances (e.g., pedestrian activity, the non-use of phytosanitary products) could favour earthworm development in reconstructed roadside soils (Pižl and Schlaghamerský, 2007; Peigné et al., 2009; Pelosi et al., 2013). The high total species richness in reconstructed roadside

soils could be explained by the presence of several rare species in the Parisian basin (e.g., *Aporrectodea caliginosa meridionalis*, *Allolobophora chlorotica postepheba*; Bouché, 1972) that may have been brought by passive dispersal (e.g., planting clods, the addition of topsoil; James and Hendrix, 2004). Our results highlight the capacity of most reconstructed roadside soils to be reservoirs of earthworm biodiversity as the four ecological categories were observed in pseudo-natural grassland soils as well as in reconstructed roadside soils. This could be highly beneficial for many soil functions such as soil organic matter turnover or water infiltration (Brown et al., 2000; Lavelle and Spain, 2001; Capowiez et al., 2009), and positively impact the delivery of ecosystem services in urban areas (Blouin et al., 2013; De Vries et al., 2013). Overall, high earthworm abundance and species richness should be particularly targeted by managers of urban areas to ensure better self-regulation of green spaces and, on a larger scale, citizens' well-being (Taylor and Hochuli, 2015).

Despite the presence of the four ecological categories in reconstructed roadside soils, endogeic earthworms largely predominated earthworm communities (90% of the total abundance), contrary to pseudo-natural grassland soils (71% of the total abundance). This imbalance, as visible in 4-year-old soils as in 20-year-old reconstructed roadside soils, could be a consequence of soil engineering processes. Indeed, endogeic earthworms are more likely to survive in stockpiles than deep-burrowing earthworms (i.e., epi-anecic, and strict-anecic earthworms) and surface dwellers (i.e., epigeic earthworms) due to unsuitable habitats in deeper layers (e.g., compaction, anaerobic conditions; Abdul-Kareem and McRae, 1984; Boyer et al., 2011) and the absence of soil cover (Chan, 2001; Pélosi et al., 2009). For example, Armstrong and Bragg (1984) highlighted the predominance of an endogeic species (*Allolobophora chlorotica*) in a 7-year-old reconstructed soil from stockpiles. Thus, as observed in our study sites, ubiquitous species such as *Aporrectodea caliginosa caliginosa*, and *Allolobophora rosea rosea* are more likely to be found in association and high abundance after soil reconstruction

(Söchtig and Larink, 1992; Fonte et al., 2009; Valckx et al., 2009). Soil engineering also impacted their ecological traits as individual biomass of *Allolobophora rosea rosea* was higher in RS than in PNG while *Aporrectodea caliginosa caliginosa* was not impacted. This could be explained by differences in terms of food resources (Butt et al., 1994; Lowe and Butt, 2002). Given this, more studies should be considered on these two species to understand their feeding behaviour. Besides, epigeic and anecic earthworms are particularly vulnerable to mechanical damage induced by machinery which can kill them during soil handling (Scullion et al., 1988; Brun et al., 1991), similarly as ploughing in agricultural areas (Chan, 2001; Briones and Schmidt, 2017). It is also possible that these species were present in low abundance in the stripped topsoil (e.g., due to agricultural practices, quiescence) (Marié and Rossignol, 1997). Consequently, a too low residual pool of individuals per species after the re-spread of stockpiled soils may not allow a sufficient reproduction rate to maintain some species, in particular anecic cross-fertilization hermaphrodite species with long generation time and low fecundity (Satchell, 1967; Barois et al., 1999; Diaz Cosin et al., 2011). Surprisingly, the abundance of anecic earthworms remained low even in 20-year-old reconstructed roadside soils in comparison to surrounding pseudo-natural grasslands (e.g., 14 and 59 individuals m<sup>-2</sup> of strict-anecic earthworms, respectively). For example, Hoeffner et al. (2021) showed that the abundance of the strict-anecic species *Aporrectodea longa longa* increased from 8 to 113 individuals m<sup>-2</sup> in only 3 years after the stop of soil disturbances due to the introduction of grassland into the crop rotations. Thus, habitat conditions in reconstructed roadside soils were suitable for endogeic but not for epigeic and anecic earthworms, perhaps because of the quality of the organic matter, the soil moisture, or the narrowness of the soil. Overall, as large-diameter species highly take part in the creation of large galleries into the soil (François Bastardie et al., 2003), it can be assumed that their low number will not allow a high contribution to soil functions associated with this gallery network, such as soil structuration (Blanchart et al., 1999; Capowiez et al.,

2012), water infiltration (Jouquet et al., 2012; Pelíšek, 2018) or organic matter turnover (Pey et al., 2013; Hoeffner et al., 2019).

#### ***4.2. Wide asphalt roads have negative effects on earthworm parameters in 20- but not in 4-year-old reconstructed soils***

Regarding 20-year-old reconstructed roadside soils, epigeic, epi-anecic, and strict-anecic earthworms were absent in soils bordered by a wide asphalt road whereas they were all present in soils bordered by a narrow asphalt sidewalk. Besides, endogeic abundance, biomass, and species richness were lower in soils bordered by roads. This contrast between similarly engineered roadside soils could be explained by several factors. Roads could constitute major barriers to earthworm displacement (Muñoz et al., 2015; Dupont et al., 2017), inducing high mortality rates when crossing the road due to the regular passage of cars or exposition to extreme conditions when earthworms remain on asphalt surfaces (i.e., drought), associated with an incapacity to access to the reconstructed roadside soils due to the 20 cm-high curb. The emigration of individuals outside the reconstructed roadside soil may also disadvantage epigeic and anecic earthworms which mostly emigrate by surface displacement (Caro et al., 2013a; Jiménez et al., 2014) contrary to endogeic earthworms which mostly live, feed, and disperse into the soil and are less likely to get out reconstructed roadside soils (Eijsackers, 2011). Habitat conditions may also be particularly adverse in mid-road soils, for example, soil moisture may be low due to deep road foundations limiting water flows, combined with a drying effect due to evapotranspiration from tree lines (Marchionni et al., 2019). The lower abundance of endogeic earthworms in soils bordered by a wide asphalt road than by a narrow asphalt sidewalk may explain the higher individual biomass of adults observed in this study. Indeed, it has been shown that the decrease in earthworm abundance contributes to the increase in individual biomass because of a decrease in intra- and/or interspecific competition to feed on soil organic

matter (Butt et al., 1994; Cardinael et al., 2019). Unfortunately, the consequences of this difference in individual biomass on the development of these populations or the soil functioning remain to be studied. Sidewalks could lead to similar constraints for earthworm development and displacement but exchanges with nearby reservoirs may be possible given their narrow width (3 m) and shallow foundations (approx. < 20 cm), allowing displacement by crawling on the surface or burrowing into the soil (Chuang and Chen, 2008; Eijsackers, 2011). The absence of curbs or level differences between the sidewalk and surrounding soils may also allow surface and subsurface water flows (Pickett et al., 2011), maintaining sufficient soil moisture for earthworms.

Regarding 4-year-old reconstructed roadside soils, earthworm communities were relatively similar regardless of the type of asphalt surface surrounding the soil. This result could be explained by a too recent soil reconstruction to observe any effects previously discussed on earthworm communities. Nonetheless, changes in the structure of communities can be expected as observed in 20-year-old reconstructed roadside soils. Overall, the effects of soil engineering and asphalt surfaces on earthworm communities should be considered in the long term in the context of sustainable urban development.

## **5. Conclusion**

In reconstructed roadside soils, total earthworm abundance and species richness reached levels comparable to surrounding pseudo-natural grassland soils which underlined a high-quality habitat provided by soil engineering. However, soil engineering processes (e.g., soil stripping, stockpiling) negatively impacted the structure of earthworm communities of reconstructed roadside soils as endogeic species represented 90% of the total abundance, mostly ubiquitous species (e.g., *Aporrectodea caliginosa caliginosa*, *Allolobophora rosea rosea*). The present study highlighted the negative impacts of wide asphalt roads on communities in 20-

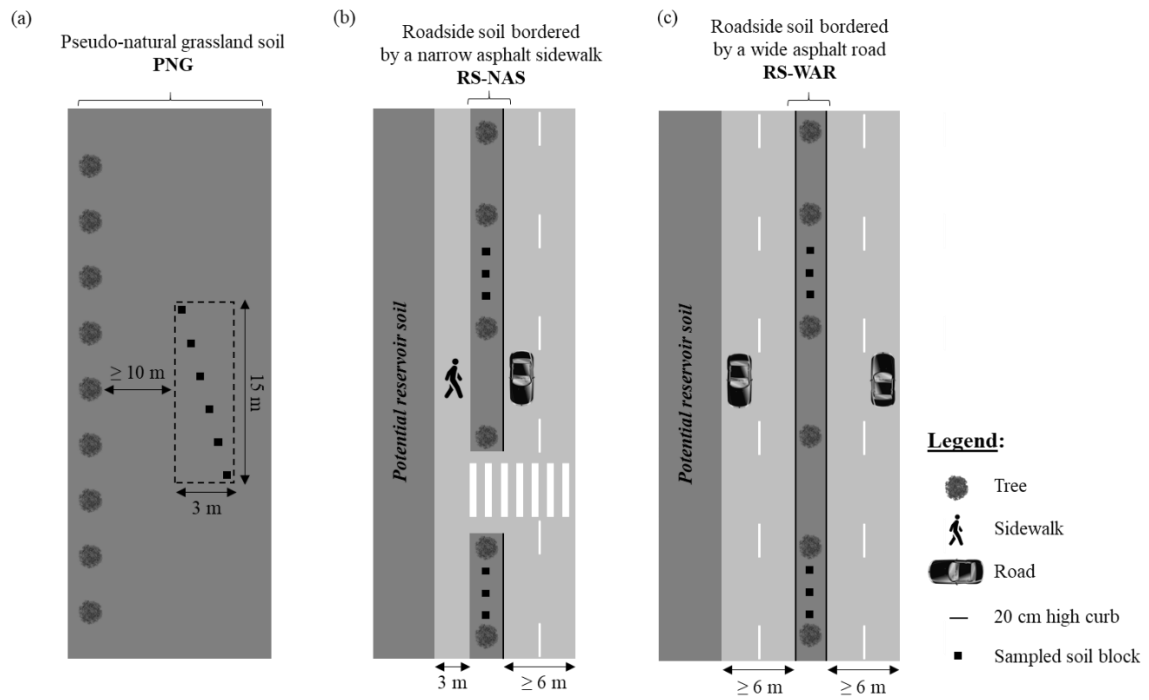
year-old reconstructed roadside soils as earthworm parameters (total abundance, total biomass, species richness) were significantly lower and three ecological categories were absent in comparison with soils bordered by a narrow sidewalk. No difference in earthworm parameters was observed in 4-year-old reconstructed roadside soils regardless of the nearby asphalt surface. Overall, favouring earthworm communities in urban areas requires the preservation or the reconstruction of high-quality habitats and ecological continuities to allow their displacement and establishment.

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## SUPPORTING INFORMATION

**Fig. S1** Diagram of the sampling plan: (a) pseudo-natural grassland soils (PNG); (b) reconstructed roadside soils bordered by a narrow asphalt sidewalk (RS-NAS); (c) reconstructed roadside soils bordered by a wide asphalt road (RS-WAR).





**Table S1** Description of the reconstructed roadside soils studied.

Code	Soil type	Soil name (IUSS Working Group, 2007)	Age (years)	Cumulated length (m)	Number of sections	Width (m)	Inter-tree (m)	Cover	Tree species
4RS	Engineered	Hortic Anthrosol (Organotransportic, Siltic)	4	970	9	1.8	4	Climbing ivy Grasses	<i>Quercus robur</i> <i>Fagus sylvatica</i> <i>Carpinus betulus</i>
20RS	Engineered	Hortic Anthrosol (Organotransportic, Siltic)	20	790	2	1.8	7	Creeping ivy	<i>Tilia pallida</i>

**Table S2** Name, first description, associated ecological category, total abundance (individuals m<sup>-2</sup>), and total occurrence (Occ.) of the 19 earthworm species of the study sites. Values of abundance are mean ± standard deviation, and are given for PNG, 4RS-NAS, 4RS-WAR, 20RS-NAS, and 20RS-WAR. Values of occurrence (Occ.) are given for PNG, 4RS, and 20RS. PNG = pseudo-natural grassland soils; 4RS = 4-year-old reconstructed roadside soils; 20RS = 20-year-old reconstructed roadside soil; 4RS-NAS = 4-year-old reconstructed roadside soils bordered by a narrow asphalt sidewalk; 4RS-WAR = 4-year-old reconstructed roadside soils bordered by a wide asphalt road; 20RS-NAS = 20-year-old reconstructed roadside soils bordered by a narrow asphalt sidewalk; 20RS-WAR = 20-year-old reconstructed roadside soils bordered by a wide asphalt road.

Genus	Species	Authority	Ecological category	Abundance PNG	Occ. (/10)	Abundance 4RS-NAS	Abundance 4RS-WAR	Occ. (/18)	Abundance 20RS-NAS	Abundance 20RS-WAR	Occ. (/18)
<i>Eiseniella</i>	<i>tetraedra</i>	Savigny (1826)	epigeic	3 ± 3	3	0 ± 0	0 ± 0	0	0 ± 0	0 ± 0	0
<i>Satchellius</i>	<i>mammalis</i>	Savigny (1826)	epigeic	7 ± 13	3	0 ± 0	0 ± 0	0	0 ± 0	0 ± 0	0
<i>Lumbricus</i>	<i>castaneus</i>	Savigny (1826)	epigeic	23 ± 18	9	11 ± 14	17 ± 20	11	38 ± 34	0 ± 0	5
<i>Lumbricus</i>	<i>castaneus disjonctus</i>	Savigny (1826)	epigeic	2 ± 5	2	6 ± 11	3 ± 6	5	35 ± 51	0 ± 0	3
<i>Lumbricus</i>	<i>rubellus rubellus</i>	Hoffmeister (1843)	epi-aneic	5 ± 5	6	7 ± 15	3 ± 4	5	17 ± 17	0 ± 0	4
<i>Lumbricus</i>	<i>terrestris</i>	Linnaeus (1758)	epi-aneic	1 ± 3	1	0 ± 0	0 ± 0	0	4 ± 5	0 ± 0	3
<i>Lumbricus</i>	<i>festivus</i>	Savigny (1826)	epi-aneic	0 ± 1	1	0 ± 0	0 ± 0	0	0 ± 0	0 ± 0	0
<i>Aporrectodea</i>	<i>giardi</i>	Savigny (1826)	strict-aneic	50 ± 26	9	0 ± 0	0 ± 0	0	13 ± 23	0 ± 0	2
<i>Aporrectodea</i>	<i>longa longa</i>	Ude (1885)	strict-aneic	8 ± 11	4	3 ± 6	5 ± 7	5	0 ± 0	0 ± 0	0
<i>Aporrectodea</i>	<i>caliginosa meridionalis</i>	Bouché (1972)	strict-aneic	0 ± 0	0	0 ± 0	0 ± 0	0	1 ± 3	0 ± 0	1
<i>Aporrectodea</i>	<i>caliginosa caliginosa</i>	Savigny (1826)	endogeic	59 ± 25	10	169 ± 159	180 ± 106	18	161 ± 132	11 ± 15	11
<i>Allolobophora</i>	<i>chlorotica chlorotica</i>	Savigny (1826)	endogeic	9 ± 17	4	17 ± 30	45 ± 55	9	8 ± 14	0 ± 0	2
<i>Allolobophora</i>	<i>chlorotica postepheba</i>	Bouché (1972)	endogeic	0 ± 0	0	1 ± 3	0 ± 0	1	0 ± 0	0 ± 0	0
<i>Allolobophora</i>	<i>icterica</i>	Savigny (1826)	endogeic	132 ± 96	10	20 ± 35	12 ± 15	11	71 ± 55	65 ± 60	18
<i>Allolobophora</i>	<i>rosea rosea</i>	Savigny (1826)	endogeic	48 ± 73	7	68 ± 58	89 ± 86	15	61 ± 49	58 ± 54	16
<i>Allolobophora</i>	<i>limicola</i>	Michaelsen (1900)	endogeic	0 ± 0	0	3 ± 4	6 ± 9	7	100 ± 65	0 ± 0	5
<i>Allolobophora</i>	<i>minima</i>	Savigny (1826)	endogeic	6 ± 10	5	0 ± 0	0 ± 0	0	0 ± 0	0 ± 0	0
<i>Octolasion</i>	<i>cyaneum</i>	Savigny (1826)	endogeic	0 ± 0	0	0 ± 0	0 ± 0	0	4 ± 7	1 ± 2	3
<i>Octolasion</i>	<i>lacteum lacteum</i>	Orley (1885)	endogeic	0 ± 0	0	2 ± 6	0 ± 0	1	0 ± 0	0 ± 0	0

**Table S3** Individual biomass ( $\text{g}\cdot\text{m}^{-2}$ ) of *Aporrectodea caliginosa caliginosa* and *Allolobophora rosea rosea* across pseudo-natural grassland soils and reconstructed roadside soils. Values are mean  $\pm$  standard deviation. Different letters denote significant differences among groups with a > b. W-values and p-values were given from Wilcoxon tests between PNG and RS. PNG = pseudo-natural grassland soils; RS = all reconstructed roadside soils combined.

Genus	Species	Authority	Ecological category	PNG		RS		Wilcoxon	p-value
				Individual biomass	Number of adults	Individual biomass	Number of adults		
<i>Aporrectodea</i>	<i>caliginosa caliginosa</i>	Savigny (1826)	endogeic	0.38 $\pm$ 0.11 a	20	0.41 $\pm$ 0.18 a	70	W = 653.5	0.655
<i>Allolobophora</i>	<i>rosea rosea</i>	Savigny (1826)	endogeic	0.17 $\pm$ 0.06 b	46	0.21 $\pm$ 0.09 a	82	W = 1454	0.032

**Table S4** Individual biomass ( $\text{g}\cdot\text{m}^{-2}$ ) of *Aporrectodea caliginosa caliginosa* and *Allolobophora rosea rosea* across reconstructed roadside soils. Values are mean  $\pm$  standard deviation. Different letters denote significant differences among groups with a > b. t-values and p-values were given from Student tests between (a) 4RS-NAS and 4RS-WAR, and between (b) 20RS-NAS, 20RS-WAR. 4RS-NAS = 4-year-old reconstructed roadside soils bordered by a narrow asphalt sidewalk; 4RS-WAR = 4-year-old reconstructed roadside soils bordered by a wide asphalt road; 20RS-NAS = 20-year-old reconstructed roadside soils bordered by a narrow asphalt sidewalk; 20RS-WAR = 20-year-old reconstructed roadside soils bordered by a wide asphalt road.

(a)

Genus	Species	Authority	Ecological category	4RS-NAS		4RS-WAR		Student p-value	
				Individual biomass	Number of adults	Individual biomass	Number of adults		
<i>Aporrectodea</i>	<i>caliginosa caliginosa</i>	Savigny (1826)	endogeic	0.40 ± 0.10 a	18	0.38 ± 0.15 a	21	t = 0.4	0.721
<i>Allolobophora</i>	<i>rosea rosea</i>	Savigny (1826)	endogeic	0.18 ± 0.07 a	14	0.16 ± 0.04 a	26	t = 1.5	0.142

(b)

Genus	Species	Authority	Ecological category	20RS-NAS		20RS-WAR		Student p-value	
				Individual biomass	Number of adults	Individual biomass	Number of adults		
<i>Aporrectodea</i>	<i>caliginosa caliginosa</i>	Savigny (1826)	endogeic	0.36 ± 0.12 b	25	0.78 ± 0.22 a	6	t = -6.4	<0.001
<i>Allolobophora</i>	<i>rosea rosea</i>	Savigny (1826)	endogeic	0.18 ± 0.08 b	10	0.27 ± 0.09 a	32	t = -2.5	0.015

**CHAPITRE 4 - Réponse des communautés lombriciennes à  
l'ingénierie pédologique et à l'isolement des sols dans les paysages  
urbains**

Le quatrième chapitre correspond à un article publié dans la revue internationale *Ecological Engineering*.

### **Objectifs de l'étude**

Les objectifs de cette étude étaient de (i) caractériser les communautés lombriciennes dans des sols d'accompagnement de voirie issus de deux processus d'ingénierie contrastés, ainsi que dans des prairies pseudo-naturelles à proximité directe (i.e., potentiels réservoirs), et (ii) d'évaluer les impacts sur les communautés lombriciennes de deux types de surface asphaltées séparant les sols issus de l'ingénierie pédologique de potentiels réservoirs.

### **Méthodologie**

Les sites d'étude sont les mêmes que ceux proposés dans le chapitre 2, à savoir des Luvisols anthropisés de 30 ans et des Anthrosoles reconstitués de 20 ans. Les vers de terre ont été échantillonnés manuellement par extraction de blocs de sol de manière à couvrir l'ensemble des linéaires de sol étudiés. Pour chacun des types de sol, les prélèvements ont lieu au sein de tronçons (i) partiellement isolés par rapport à un potentiel réservoir (i.e., séparation par un trottoir de 3 m de large), et (ii) totalement isolés par rapport à un potentiel réservoir (i.e., séparation par une route de 6 m de large).

### **Principaux résultats**

L'ingénierie pédologique, tous types de sol confondus, n'a pas affecté l'abondance lombricienne totale tandis que la richesse totale a été négativement impactée par rapport aux sols pseudo-naturels situés à proximité directe. L'isolement total a eu un impact différent sur les communautés lombricienne selon le processus d'ingénierie considéré. Dans les Anthrosoles reconstitués issus d'un processus élaboré, l'abondance totale et la richesse spécifique étaient

significativement plus faibles en isolement total qu'en isolement partiel, alors qu'aucune différence n'a été observée selon les conditions d'isolement dans les Luvisols anthropisés issus d'un processus basique.

**Title: Response of earthworm communities to soil engineering and soil isolation in urban landscapes**

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## **Abstract**

Engineered soils provide numerous ecosystem services in urban landscapes, such as water regulation and plant growth. They are constructed to optimize soil physicochemical properties but their biological properties are given little consideration. In particular, earthworm communities may be highly impacted by soil engineering processes and soil isolation caused by asphalted surfaces separating soils, and in particular roadside soils, from pseudo-natural soils. In this context, this study aimed to evaluate (i) the effects of soil engineering processes applied to construct roadside soils, and (ii) the effects of soil isolation from pseudo-natural soils by asphalted surfaces on earthworm communities. The study was conducted in an urban landscape in the suburbs of Paris, France. We sampled earthworms in roadside soils from two distinct soil engineering processes: basic engineering (BER) associated with shallow stripping depth and advanced engineering (AER) associated with deep stripping depth and organic amendment. Within each soil engineering process, two levels of soil isolation were defined depending on the asphalted surface separating roadside soils from pseudo-natural soils: light isolation (LI) in case of a narrow cycle path, and high isolation (HI) in case of a wide road. Soil engineering did not affect the total earthworm abundance whereas the total species richness was negatively impacted in comparison to nearby pseudo-natural soils. High soil isolation differently impacted earthworm communities depending on the engineering process. Regarding AER, earthworm abundance and species richness were significantly lower in HI than in LI while no differences were observed between BER-LI and BER-HI. Overall, engineering processes define the soil's ability to host earthworms, and soil isolation defines soil ability to be colonized from nearby environments. Considering the contribution of earthworms to the provision of ecosystem services, both soil engineering and soil isolation should be taken into account in urban projects to optimize their development in urban landscapes.

Keywords: urban soils, pedological engineering, connectivity, ecological continuity, brown network

## **1. Introduction**

In response to global changes, urban landscapes integrate climate-resilient engineered soils (e.g., parks, roundabouts, roadsides), especially to balance water flows and provide thermal comfort through vegetation (Demuzere et al., 2014; Kim et al., 2016). Many ecosystem services such as water regulation or support for plant growth are mainly provided by urban soils depending on their physical, chemical, and biological properties (Dominati et al., 2010). Engineered soil properties result from processes which include, among others, soil stripping, transportation, and implementation in one or several layers (Marié and Rossignol, 1997; Vidal-Beaudet and Rossignol, 2018). Therefore, the construction of engineered soils in the urban landscape is a key element to maintain functional soils and address urban issues (Morel et al., 2015; Blanchart et al., 2018; Calzolari et al., 2020). If the physical and chemical components of engineered soils have been studied (Damas and Coulon, 2016; Séré et al., 2017), the biological component, despite being a key player in soil processes (Balser and Firestone, 2005; Ernst et al., 2009), remains largely unexplored.

Among soil biological actors, earthworms are widespread in temperate climates and are involved in numerous ecosystem services (Lavelle et al., 2006; Blouin et al., 2013; Bertrand et al., 2015), acting in particular on water regulation (Ernst et al., 2009; Pelíšek, 2018) and plant growth (Scheu, 2003; van Groenigen et al., 2014). They are usually classified into four main categories (Bouché, 1972, 1977; Jégou et al., 1998): epigeic earthworms live and feed on organic matter of the soil surface, endogeic earthworms create horizontal burrows and feed on soil organic matter, epi-anecic earthworms rather feed on fresh surface organic matter whereas strict-anecic earthworms preferentially feed on aged organic matter already incorporated into

the soil (Bouché, 1977; Jégou et al., 1998). Earthworm communities of urban soils are poorly described and the few studies available highlighted the effects of soil physicochemical properties (Amossé et al., 2016; Audusseau et al., 2020), land-uses (Li et al., 2020; Tóth et al., 2020), and land management (Smetak et al., 2007; Vergnes et al., 2017). For example, Smetak et al. (2007) observed that additions of nitrogenous fertilizer in urban parks were associated with an increase of an anecic species abundance. However, to our knowledge, there is no study on the effect of engineering processes on earthworm communities in urban landscapes.

Several studies have been undertaken at a large scale to evaluate the impact of landscape diversity on earthworm communities (Joschko et al., 2006; Frazão et al., 2017; Hoeffner et al., 2021). For example, Vanbergen et al. (2007) highlighted a positive correlation between earthworm diversity and habitat richness at landscape-scale (1 km<sup>2</sup>); between earthworm abundance and plant species richness at a local scale (200 m<sup>2</sup>). In the urban context, Xie et al. (2018) observed that the ratio of asphalted surface and green space surface within different buffers from 200 m to 1 km was negatively correlated with an anecic species. However, the majority of the previous studies did not report the impact of landscape on earthworm communities at a very fine scale (Vanbergen et al., 2007; Lüscher et al., 2014; Frazão et al., 2017; Xie et al., 2018). In particular, the effect of asphalted surfaces such as roads or cycle paths surrounding roadside soils on earthworm communities remains unclear. Tiho and Josens (2000) highlighted similar earthworm communities between soils surrounded by two roads and soils of local urban parks, whereas Dupont et al. (2017) reported that for two endogeic species roads could constitute barriers to earthworm dispersal at a regional scale (~130 km<sup>2</sup>). Thus, earthworm communities of roadside soils and the effect of surrounding landscapes at a very fine scale are still to be explored.

The first objective of the present study was to evaluate the effects of soil engineering processes applied to construct roadside soils on earthworm communities (total abundance, total

species richness, evenness index, and abundance within each earthworm ecological category). We hypothesized that the more steps the engineering process includes, the more degraded earthworm communities are likely to be. The second objective was to evaluate the effects of soil isolation by asphalted surfaces on earthworm communities. We hypothesized that earthworm communities in highly isolated roadside soils (i.e., separated from pseudo-natural soils by a wide road) are likely to be more degraded than earthworm communities in lightly isolated roadside soils (i.e., separated from pseudo-natural soils by a narrow cycle path). We conducted a study in an urban landscape of Paris' suburbs, France. Earthworms were sampled in pseudo-natural grassland soils (controls) and along linear roadside soils from two different soil engineering processes: basic engineering involving shallow soil stripping and the addition of a single unmodified soil layer; advanced engineering involving deep soil stripping, the addition of two soil layers and an organic amendment in the surface soil layer. Within each engineering process, we studied soil isolation by comparing lightly and highly isolated roadside soils.

## **2. Materials and methods**

### ***2.1. Study sites and sampling design***

The study area is located around the city of Paris (France), in the commune of Guyancourt (48° 76' 18'' N; 2° 07' 23'' E) which belongs to the urban community of Saint-Quentin-en-Yvelines, built as a "new city" in the 1970s to avoid urban concentration in Paris. The city expands on large agricultural parcels where the substrate is characterized by fine loess deposits more than 3 metres deep (Mégnyen, 1989). Guyancourt is structured by three main avenues: two from north-east to south-west, and one from west to east. The climate is temperate, with a mean annual precipitation of 563 mm and a mean annual temperature of 11.2°C, a monthly

minimum of  $-2.3^{\circ}\text{C}$  and a monthly maximum of  $26.4^{\circ}\text{C}$  (data from Météo France, 2008-2018 Trappes station).

Two linear roadside soils made by following distinct engineering processes were selected: (i) basic soil engineering process (Léon Blum Avenue) which is commonly used in the urban context and (ii) advanced soil engineering process (Europe Avenue) which is rarely used in the urban context. These roadside soils were selected for the detailed historical knowledge available (Table S1), their proximity (less than 2 km apart), their great age (30 and 20 years old respectively), and the presence of adjacent pseudo-natural grassland soils. Basic engineered roadside soils (BER) were implemented at the end of the 1980s. The existing soil was stripped to a depth of 30 cm to reach the desired road level. The topsoil was reconstituted with an addition of 20 to 30 cm of organo-mineral material (silty topsoil). Advanced engineered roadside soils (AER) were implemented at the end of the 1990s. The existing soil was stripped to a depth of 150 cm. The soil was reconstituted from bottom to top with 125 cm of organo-mineral material (silty topsoil), 25 cm of a mixture of organo-mineral material and compost from green waste, and finally a thin layer of pine bark mulch (Table S1). In both cases, the organo-mineral material came from the perimeter of the urban project and corresponds to topsoil stripped down to 30 cm, stored in a windrow, and then transported to the roadside location. Six soil profile descriptions per engineering process (BER and AER) were performed in 110 cm deep pedological pits (NF EN ISO 25177; AFNOR, 2019). Soils were identified according to the IUSS Working Group (WRB, 2015). In addition, for each roadside soil, nearby pseudo-natural grasslands soils (BER-PNG and AER-PNG, respectively) left untouched by urban engineering at least for 30 and 20 years old respectively were selected.

Within each of the two types of engineered roadside soils, two levels of isolation from adjacent pseudo-natural grassland soils were selected: (i) lightly isolated roadside soils (BER-LI and AER-LI, respectively) between a 6 m wide asphalt road with a 20 cm high curb and a 3

m wide asphalt cycle path with no curb; (ii) highly isolated roadside soils (BER-HI and AER-HI, respectively) between two asphalt roads, each 6 m wide with a 20 cm high curb (Fig. S1). Details concerning land use and management of each group are shown in Table S1. Earthworms and soil were sampled in order to cover all of the selected avenue portions. Each replicate was separated from 60 m and the number of replicates for each group varied depending on the avenue's layouts (i.e., intersections) and length. Thus, 6 groups were studied: (i) BER-PNG (n=10), (ii) BER-LI (n=10), (iii) BER-HI (n=5), (iv) AER-PNG (n=7), (v) AER-LI (n=7), and (vi) AER-HI (n=7).

## ***2.2. Earthworm sampling and laboratory analyses***

Each earthworm sampling consisted of extracting 6 blocks of soil (20 cm × 20 cm × 25 cm, length × width × depth) and hand-sorting to collect earthworms. Sampling of soil blocks was undertaken at a distance of 2 m from a tree and equidistant from the edge of the tree trench. Earthworms were stored in ethanol (96% solution). In the laboratory, earthworms were counted, assigned to a stage of development (juvenile, sub-adult and adult), identified at the sub-species level and assigned to its ecological categories: epigeic, epi-anecic (genus *Lumbricus*), anecic-strict (genus *Aporrectodea*) and endogeic (Bouché, 1972, 1977; Jégou et al., 1998; Hoeffner et al., 2019). For juveniles that could not be determined at the sub-species, they were first assigned to a genus and an ecological category, then distributed in proportion of sub-adults and adults identified at the sub-species level in each group. The number of hand-sorted earthworms was multiplied by 25 to obtain an estimation per square metre. Earthworm diversity was analysed by calculating the total species richness, and the evenness index. The evenness index ( $J'$ ) was calculated as follows:  $J' = \frac{H'}{H'_{max}}$ , where  $H'$  is the Shannon diversity index. Abundance was also analysed at the scale of ecological categories.

### ***2.3. Soil sampling and physicochemical analyses***

Soil samples were taken for analysis from the soil where earthworms had been collected. Six sub-samples of soil (between 0 and 25 cm) were pooled and homogenized to consider one composite sample per site. Composites were analysed by the analytical laboratory AUREA AgroSciences (Orléans, France) for particle-size distribution (clay  $<2\ \mu\text{m}$ ; silt  $2\ \mu\text{m}<50$ ; sand  $50\ \mu\text{m}<200$ , %; NFX 31-107 without decarbonatation; AFNOR, 2003), soil organic matter (SOM; NF ISO 14235; AFNOR, 1998) and pH (water; NF ISO 10390; AFNOR, 2005).

### ***2.4. Statistical analyses***

Wilcoxon tests were applied to compare total abundance, total species richness, evenness index, and abundance of ecological categories between BER-PNG and BER, and between AER-PNG and AER. Two-ways ANOVAs for parametric data were applied to test the factors “soil engineering”, “soil isolation”, and their interaction on total abundance, total species richness, and evenness index between BER-LI, BER-HI, AER-LI, and AER-HI. We used a type III sum of squares ANOVA because the sampling design was unbalanced. In case of significant interaction, one-way ANOVA, and pairwise multiple comparisons were performed to highlight the significant differences between mean variables of BER-LI, BER-HI, AER-LI, and AER-HI. Given the large contrasts of abundance between groups at the scale of ecological categories, Kruskal-Wallis tests for non-parametric data were applied, followed by Dunn tests for post-hoc pairwise comparisons to identify the significant differences between BER-LI, BER-HI, AER-LI, and AER-HI.

The species composition of earthworm communities among BER-PNG, BER-LI, BER-HI, AER-PNG, AER-LI, and AER-HI was compared thanks to a data matrix of pairwise comparisons among samples calculated using the Bray-Curtis distance index. Non-Metric multidimensional Scaling (NMDS) was then used to find the best low-dimensional

representation of the distance matrix. The null hypothesis of no difference in patterns of earthworm community structure was tested with a permutational multivariate analysis of variance (PERMANOVA). PERMANOVAs were run on the Bray-Curtis distance with 1000 permutations per analysis.

All statistical analyses were performed on R software (R Core Team, 2019). Significance was evaluated in all cases at  $P < 0.05$ .

### **3. Results**

#### ***3.1. Physicochemical characteristics of soils***

BER-PNG and AER-PNG were identified as Haplic Luvisols, with a silt content of  $63 \% \pm 3.5$  ( $\pm$  SD), a pH of  $7.8 \pm 0.3$  and an organic matter content of  $1.8 \% \pm 0.4$ . BER and AER were similar in pH ( $7.4 \pm 0.6$  and  $7.2 \pm 0.2$ , respectively) and soil texture, with a dominant percentage of silt ( $71 \% \pm 4.0$  and  $67 \% \pm 2.4$  respectively), while the percentage of organic matter was higher AER in than in BER ( $6.4 \% \pm 1.0$  and  $1.8 \% \pm 0.3$ , respectively) (Table S2). BER were identified as Terric Luvisols (Transportic, Siltic) and AER were identified as Hortic Anthrosols (Organotransportic, Siltic) (Table S1).

#### ***3.2. Earthworm communities of the study area***

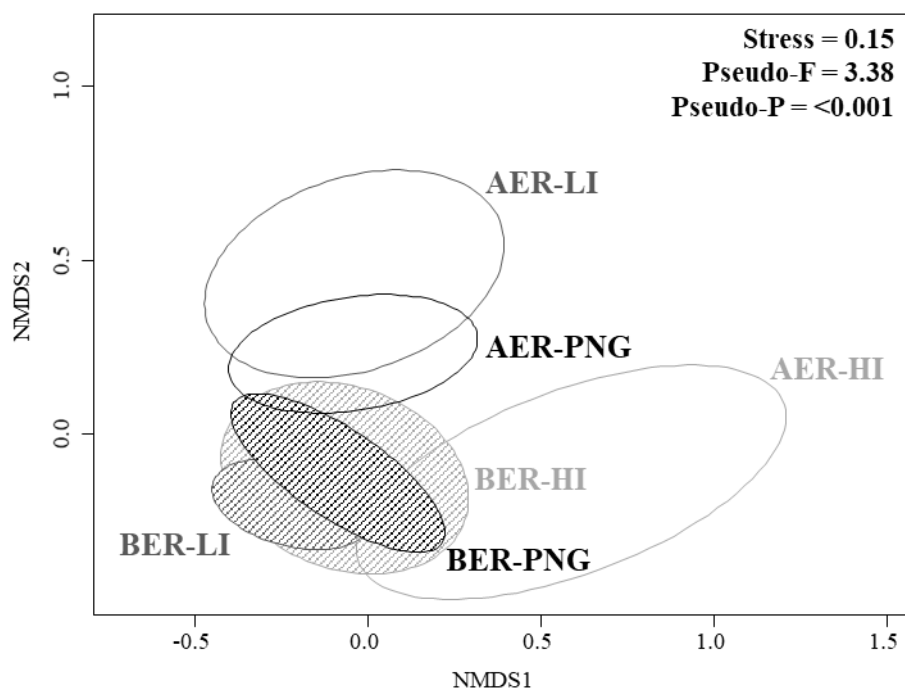
Over the whole study area, the mean earthworm abundance and species richness were  $369 \pm 190$  individuals  $m^{-2}$  and  $5.6 \pm 1.8$  species, respectively (Table 1, Table S3). Fourteen species were observed (Table S4): three epigeic (dominated by *Lumbricus castaneus*), two epi-anecic (*Lumbricus rubellus rubellus* and *Lumbricus terrestris*), two strict-anecic (*Aporrectodea giardi* and *Aporrectodea longa longa*), and seven endogeic earthworms (dominated by *Aporrectodea icterica*).



Regarding both pseudo-natural grassland soils, earthworm abundance was  $377 \pm 150$  individuals  $m^{-2}$  and  $485 \pm 163$  individuals  $m^{-2}$  in BER-PNG and AER-PNG respectively, and was not different to their respective engineered roadside soils ( $W = 59.0$  and  $29.0$  respectively,  $P > 0.05$ ; Table S3). Earthworm species richness was significantly higher in both pseudo-natural grassland soils compared to their respective engineered roadside soils (at least +34 %;  $W = 6.0$  and  $12.5$  respectively,  $P < 0.05$ ; Table S3), especially concerning epi-aneic species in BER and strict-aneic species in AER. In addition, evenness index was significantly higher in BER-PNG than in BER (+14 %,  $W = 31.0$ ,  $P = 0.016$ ; Table S3) while no difference was observed between AER-PNG and AER ( $W = 36.5$ ,  $P > 0.05$ ; Table S3). Among ecological categories, epi-aneic abundance was significantly higher in BER-PNG than in BER (+93 %,  $W = 35.0$ ,  $P = 0.006$ ; Table S3), strict-aneic abundance was significantly higher in AER-PNG than in AER (+72 %,  $W = 15.5$ ,  $P = 0.011$ ; Table S3) while no differences were observed between grassland soils and their respective engineered roadside soils regarding epigeic and endogeic abundance ( $W < 39.0$ ,  $P > 0.05$ ; Table S3).

Regarding lightly isolated roadside soils (LI), earthworm species richness and evenness index were significantly lower in BER-LI than in AER-LI (at least +31 %,  $F = 24.4$  and  $10.3$ , respectively,  $P < 0.004$ ) while earthworm abundance was not affected by the engineering process (Table 1). Among ecological categories, epigeic and epi-aneic abundances were significantly lower in BER-LI than in AER-LI (at least +88 %,  $K = 12.9$  and  $15.8$ , respectively,  $P < 0.005$ ; Table 2) while strict-aneic and endogeic abundance were not affected by the engineering process. In addition, earthworm communities were different in BER-LI and AER-LI (Fig. 1). Regarding highly isolated roadside soils (HI), strict-aneic abundance was significantly higher in BER-HI than in AER-HI (+100 %,  $K = 15.1$ ,  $P = 0.002$ ; Table 2) while earthworm abundance, species richness, evenness index, as well as epigeic, epi-aneic and endogeic abundance were similar between BER-HI and AER-HI (Table 1, Table 2).

In basic engineered roadside soils (BER) no differences were observed in both BER-LI or BER-HI regarding earthworm abundance, species richness, evenness index, structure of communities, as well as epigeic, epi-anecic, strict-anecic and endogeic abundance (Table 1, Table 2, Fig.1). Regarding advanced engineered roadside soils (AER), earthworm abundance and species richness were significantly higher in AER-LI than in AER-HI (at least +54 %,  $F = 10.8$  and  $24.4$  respectively,  $P < 0.003$ ) while evenness index was not affected by soil isolation (Table 1). Among ecological categories, epi-anecic and endogeic abundance were significantly higher in AER-LI than in AER-HI (+at least 64 %,  $K = 11.6$  and  $15.8$ ,  $P < 0.009$ ). In addition, earthworm communities were different between AER-LI and AER-HI (Fig. 1) in particular due to the absence of epigeic and strict-anecic earthworms in AER-HI (Table 1).



**Fig. 1** Non-metric multidimensional scaling ordinations showing similarities, based on Bray-Curtis distance index, of earthworm communities of each group. Significant dissimilarities between groups were assessed by PERMANOVA. Pseudo-F values and associated P-values are indicated. Ellipses correspond to groups (BER-PNG = pseudo-natural grassland soils nearby

basic engineered roadside soils; BER-LI = lightly isolated basic engineered roadside soils; BER-HI = highly isolated basic engineered roadside soils; AER-PNG = pseudo-natural grassland soils nearby advanced engineered roadside soils; AER-LI = lightly isolated advanced engineered roadside soils; AER-HI = highly isolated advanced engineered roadside soils).

**Table 1** Effects of soil engineering and soil isolation on earthworm communities. Values are mean  $\pm$  standard deviation. F-values and P-values from Anova type III were given for total abundance, total species richness, and evenness index (Eng = Engineering; Iso = Isolation). Different letters denote significant differences regarding the interaction Eng\*Iso among groups, with a > b > c (post-hoc Tukey test results).

	Basic engineered roadside soils (BER)		Advanced engineered roadside soils (AER)		Eng		Iso		Eng * Iso	
	Light isolation (BER-LI, n = 10)	High isolation (BER-HI, n = 5)	Light isolation (AER-LI, n = 7)	High isolation (AER-HI, n = 7)	Anova	p-value	Anova	p-value	Anova	p-value
Total abundance (ind.m <sup>-2</sup> )	327 $\pm$ 104 ab	340 $\pm$ 181 ab	538 $\pm$ 239 a	153 $\pm$ 92 b	F = 0.0	0.848	F = 9.5	0.005	F = 10.8	0.003
Total species richness	4.6 $\pm$ 0.5 b	4.4 $\pm$ 1.1 bc	6.7 $\pm$ 1.0 a	3.1 $\pm$ 1.1 c	F = 1.6	0.221	F = 30.5	9.6e-06	F = 24.4	4.4e-06
Evenness index	0.5 $\pm$ 0.1 b	0.7 $\pm$ 0.1 ab	0.8 $\pm$ 0.1 a	0.6 $\pm$ 0.2 ab	F = 3.5	0.073	F = 0.3	0.570	F = 10.3	0.004

**Table 2** Effects of soil engineering and soil isolation on earthworm abundance per ecological categories (ind.m<sup>-2</sup>). Values are mean ± standard deviation. K-values and P-values from Kruskal-Wallis test were given for epigeic, epi-anecic, strict-anecic, and endogeic abundance. Different letters denote significant differences among groups with a > b (post-hoc Dunn test results).

	Basic engineered roadside soils (BER)		Advanced engineered roadside soils (AER)		Kruskal-Wallis	p-value
	Light isolation (BER-LI, n = 10)	High isolation (BER-HI, n = 5)	Light isolation (AER-LI, n = 7)	High isolation (AER-HI, n = 7)		
Epigeic abundance	6 ± 5 b	28 ± 31 ab	50 ± 69 a	0 ± 0 b	K = 12.9	0.005
Epi-anecic abundance	1 ± 3 b	0 ± 0 b	28 ± 40 a	2 ± 2 b	K = 15.8	0.001
Strict-anecic abundance	41 ± 33 a	60 ± 57 a	38 ± 37 a	0 ± 0 b	K = 15.1	0.002
Endogeic abundance	280 ± 96 a	252 ± 122 ab	422 ± 157 a	151 ± 92 b	K = 11.6	0.009

#### 4. Discussion

In the present study, mean earthworm abundance and species richness ranged from 153 to 538 individuals  $m^{-2}$  and from 3.1 to 7.7 species. The lowest mean earthworm abundance and species richness observed are close to those observed in previous urban studies (Amossé et al., 2016; Xie et al., 2018), for example Vergnes et al. (2017) found a mean of 93.4 individuals  $m^{-2}$  and 4.6 species in 20 urban parks of two districts of Paris. The highest mean earthworm abundance observed in the present study is higher than those observed in grasslands where earthworms are generally the most abundant in temperate climates (Decaëns et al., 2008), while the highest mean earthworm species richness observed is lower than those observed in grasslands. For examples, Didden (2001) found a mean of 384 individuals  $m^{-2}$  and 10 species in 20 grasslands in the Netherlands, and Cluzeau et al. (2012) found a mean of 350 individuals  $m^{-2}$  and 9.6 species among 47 grasslands in Brittany. These results underline the high variability of earthworm communities within a single city as observed in previous studies (Smetak et al., 2007; Xie et al., 2018). Indeed, urban landscapes are strongly influenced by numerous anthropogenic factors (e.g., historical land-use, local soil management, urban shapes) which differently affect earthworm communities at multiple spatial urban scales (Swan et al., 2011; Aronson et al., 2016) such as city, district or neighbourhood. Thus, earthworm communities of urban soils can be highly degraded as well as preserved, modifying their level of contribution to ecosystem services (Lavelle et al., 2006; Blouin et al., 2013).

Soil engineering did not affect the total earthworm abundance, as compared to their respective pseudo-natural grassland soils, earthworm abundance was similar in basic and advanced engineered roadsides soils. Thus, despite soil disturbances caused by engineering processes (e.g., soil stripping, soil transport) and regardless of engineering intensity (e.g., stripping depth, soil addition), earthworm abundance reached comparable levels to pseudo-natural soils after 20 and 30 years. This similar earthworm abundance between engineered

roadside soils and nearby pseudo-natural grassland soils can be explained by the absence of soil disturbance in recent decades such as pedestrian activity or soil manipulation, which favours earthworm development (Pižl and Schlaghamerský, 2007; Peigné et al., 2009). Besides, the presence of a permanent cover in both engineered roadsides soils could also be a favourable factor for earthworm development as it provides food resources and increases protection against climatic hazards (Pélosi et al., 2009; Schreck et al., 2012). However, soil engineering negatively affected total earthworm species richness which reinforces a previous study that showed in urban landscapes that earthworm species richness declines due to soil disturbance (i.e., soil engineering) and the loss of habitats due to urbanisation (Xie et al., 2018). Given this erosion of earthworm diversity, engineering processes could include population transfers from nearby pseudo-natural soils to engineered soils thanks to inoculation techniques that would compensate for losses due to constructions (Butt et al., 1997; Butt, 1999).

Soil isolation differently impacted earthworm communities in both engineered roadside soils. Regarding advanced engineered roadside soils, earthworm abundance was the highest in lightly isolated roadsides among all the roadside soils considered. This result highlights a particularly suitable environment for earthworm communities in advanced engineered roadside soils. Although the entire soil profile has been reconstituted to a depth of 150 cm, the use of topsoil and the organic amendment in the surface soil layer, highlighted by a high percentage of organic matter, allowed a high level of earthworm development by providing abundant trophic resources (Lapied et al., 2009; Ashwood et al., 2017). High levels of organic matter in advanced engineered soils could explain the high abundance of the geophagous endogeic earthworm *Aporrectodea caliginosa caliginosa*, which was almost two times more abundant in lightly isolated soils than in nearby pseudo-natural soils (Bouché, 1972; Doube et al., 1997). Earthworm abundance and species richness were in contrast the lowest in highly isolated advanced roadside soils which shows, despite highly suitable habitats provided by the advanced

engineering process, that roads have a negative effect on earthworm communities. This result could be explained by high mortality rates caused by cars and drought when earthworms remain on asphalt surfaces, added to their probable difficulty to climb the 20 cm-high curbs to access the soils. Besides, highly isolated advanced roadside soils are surrounded by roads on both sides and are elevated from them which can limit water runoff and soil moisture, considered as an essential factor to earthworm survival (Lee, 1985). Thus, resulting isolated earthworm communities were almost exclusively composed of endogeic species that are particularly tolerant to soil disturbances (Rosas-Medina et al., 2010; Briones and Schmidt, 2017). Overall, the absence of two ecological categories (epigeic and anecic earthworms) can negatively affect the development of other earthworm species in terms of trophic relations, as shown by Keith et al. (2018) who highlighted for example that the surface foraging activity of anecics was beneficial for a soil-feeding endogeic species. More globally, the lack of several earthworm ecological categories may be very damaging for the maintenance of soil functions in which they are involved such as organic matter decomposition (especially epigeic species) or water infiltration (especially anecic species) (Bastardie et al., 2003; Sharma et al., 2017). Regarding soil from basic engineering processes, no differences were observed between communities of lightly and highly isolated roadside soils, and both were very similar to nearby pseudo-natural grassland soils. Similar observations were made in Brussels by Tiho and Josens (2000), who observed similar earthworm communities between highly isolated roadside soils of Roosevelt Avenue and 19 pseudo-natural soils of local urban parks. Considering that basic roadside soils were stripped at shallow depth, it can be assumed that, as suggested by Tiho and Josens (2000), earthworm communities could have developed from a relict of the earthworm communities present before the road construction and have been preserved under the modified layer of the soil. Thus, the issue of soil isolation should be taken into account in urban projects, in particular



if initial habitats and pool of species are likely to be highly impacted along the construction process.

## **5. Conclusion**

This study highlights the combined effects of soil engineering and soil isolation on earthworm communities in urban landscapes. Soil engineering did not modify earthworm abundance 20 or 30 years after roadside construction while earthworm species richness was negatively impacted. Earthworm communities of highly isolated advanced roadside soils (i.e., surrounded by roads) were strongly negatively impacted while soil isolation did not impact basic engineered soils. In the context of growing urbanisation and landscape fragmentation, our results shows that urban soil biodiversity has to be taken into account all along the different steps of urban projects. From the start, soil and biodiversity diagnostics could help to define the quality of the habitat and identify the species already present on site; low impacting engineering processes could limit the degradation of earthworm populations during the project; the preservation of reservoir areas and the maintaining of connections between green spaces could help the colonisation process; adapted soil management (e.g., ensuring sufficient soil moisture) could maintain long-term high-quality habitats for soil biodiversity. To specify and confirm technical recommendations, further studies should be conducted on other urban shapes than more or less isolated roadsides, and different urban scales from the city to the neighbourhood should be explored.

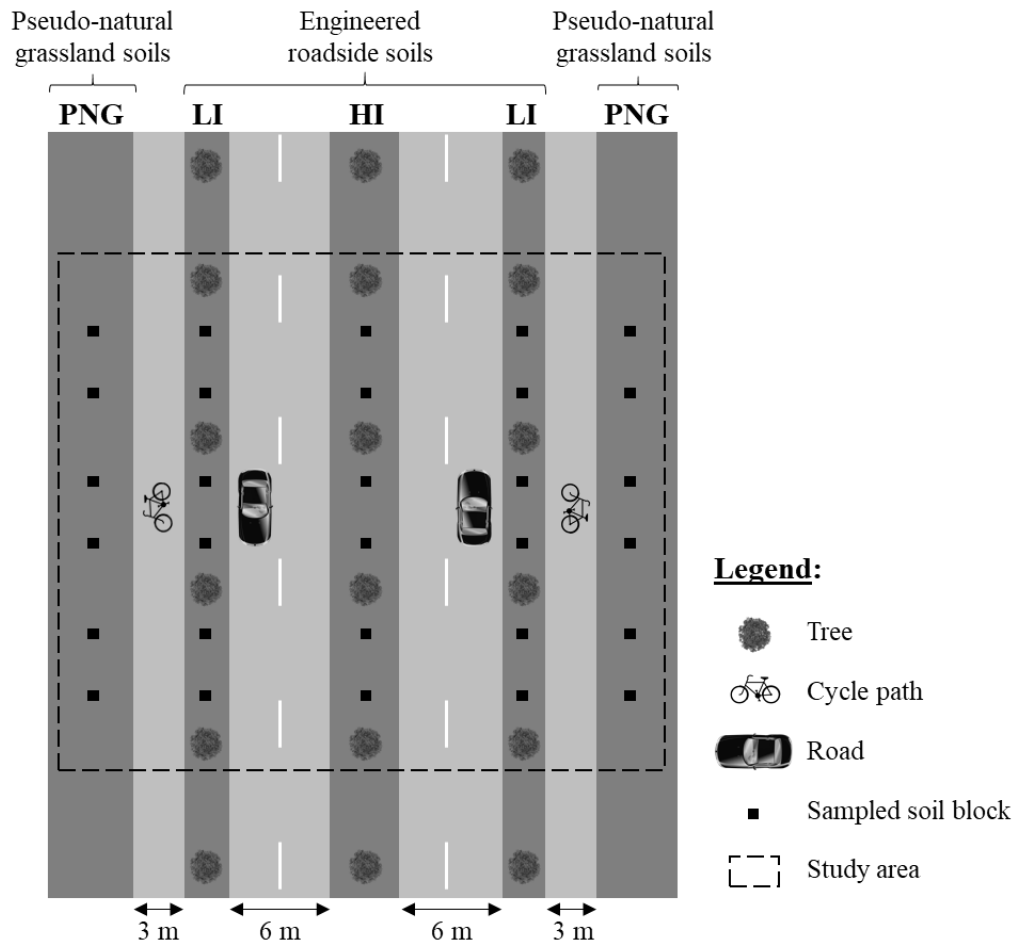
### **Acknowledgments**

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## SUPPORTING INFORMATION

**Fig. S1** Design of the sampling plan. PNG = pseudo-natural grassland soils; LI = light isolation; HI = high isolation.



**Table S1** Description of the study sites.

Code	Soil type	Engineering	Isolation	Soil name (IUSS, 2007)	Age (years)	Length (m)	Width (m)	Cover	Trees	Inter-tree (m)
BER-PNG	Pseudo-natural	-	-	Haplic Luvisol	>30	400	>100	Grass	-	-
AER-PNG	Pseudo-natural	-	-	Haplic Luvisol	>20	500	>100	Grass	-	-
BER-LI	Engineered	Basic	Light	Terric Luvisol (Transportic, Siltic)	30	400	2.45	Grass	<i>Acer platanoides</i>	10
BER-HI	Engineered	Basic	High	Terric Luvisol (Transportic, Siltic)	30	400	4	Grass	-	-
AER-LI	Engineered	Advanced	Light	Hortic Anthrosol (Organotransportic, Siltic)	20	500	1.8	Creeping ivy	<i>Tilia pallida</i>	7
AER-HI	Engineered	Advanced	High	Hortic Anthrosol (Organotransportic, Siltic)	20	500	1.8	Creeping ivy	<i>Tilia pallida</i>	7

**Table S2** Main physicochemical characteristics of pseudo-natural grassland soils and engineered roadside soils. Values are mean  $\pm$  standard deviation. PNG = pseudo-natural grassland soils; BER = basic engineered roadside soils; AER = advanced engineered roadside soils.

Code	Clay (%)	Silt (%)	Sand (%)	pH	Organic matter (%)
PNG	24 $\pm$ 4.4	63 $\pm$ 3.5	11 $\pm$ 4.1	7.8 $\pm$ 0.3	1.8 $\pm$ 0.4
BER	19 $\pm$ 2.1	71 $\pm$ 4.0	9 $\pm$ 3.0	7.4 $\pm$ 0.6	1.8 $\pm$ 0.3
AER	17 $\pm$ 1.5	67 $\pm$ 2.4	10 $\pm$ 1.9	7.2 $\pm$ 0.2	6.4 $\pm$ 1.0

**Table S3** Total abundance (ind.m<sup>-2</sup>), species richness, species evenness index and abundance per ecological category (ind.m<sup>-2</sup>) of engineered soils and associated control soils. Values are mean ± standard deviation. Different letters denote significant differences among groups with a > b (post-hoc Dunn test results). W-values and P-values from Wilcoxon test were given for total abundance, total richness, evenness index, and abundance per ecological category.

	BER-PNG (n = 10)	BER (n = 15)	Wilcoxon	p-value	AER-PNG (n = 7)	AER (n = 14)	Wilcoxon	p-value
Total abundance	377 ± 150 a	331 ± 128 a	W = 59.0	0.390	485 ± 163 a	345 ± 265 a	W = 29.0	0.149
Total species richness	6.8 ± 1.1 a	4.5 ± 0.7 b	W = 6.0	9.4e-05	7.7 ± 1.1 a	4.9 ± 2.1 b	W = 12.5	0.006
Species evenness index	0.7 ± 0.1 a	0.6 ± 0.2 b	W = 31.0	0.016	0.8 ± 0.1 a	0.7 ± 0.1 a	W = 36.5	0.370
Epigeic abundance	26 ± 26 a	13 ± 20 a	W = 42.5	0.073	53 ± 61 a	25 ± 54 a	W = 23.5	0.054
Epi-anecic abundance	7 ± 9 a	0.5 ± 2.1 b	W = 35.0	0.006	11 ± 13 ab	15 ± 31 a	W = 46.0	0.847
Strict-anecic abundance	59 ± 41 a	47 ± 42 a	W = 64.5	0.577	68 ± 47 a	19 ± 32 b	W = 15.5	0.011
Endogeic abundance	285 ± 132 a	270 ± 102 a	W = 71.0	0.846	352 ± 174 ab	287 ± 187 a	W = 39.0	0.488

**Table S4** Name, first description, associated ecological category and mean abundance (individuals m<sup>-2</sup> ± standard deviation) of the 14 earthworm species of the study sites.

Genus	Species	Author	Ecological category	Control	Basic engineered roadside soils (BER)		Control	Advanced engineered roadside soils (AER)		Total occurrence
				BER-PNG (n = 10)	Light isolation (BER-LI, n = 10)	High isolation (BER-HI, n = 5)	AER-PNG (n = 7)	Light isolation (AER-LI, n = 7)	High isolation (AER-HI, n = 7)	
<i>Satchellius</i>	<i>mammalis</i>	Savigny (1826)	epigeic	1 ± 3	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	2
<i>Lumbricus</i>	<i>castaneus</i>	Savigny (1826)	epigeic	25 ± 25	6 ± 5	28 ± 31	37 ± 30	31 ± 28	0 ± 0	33
<i>Lumbricus</i>	<i>castaneus disjonctus</i>	Savigny (1826)	epigeic	0 ± 0	0 ± 0	0 ± 0	16 ± 36	20 ± 47	0 ± 0	5
<i>Lumbricus</i>	<i>rubellus rubellus</i>	Hoffmeister (1843)	epi-anecic	5 ± 9	1 ± 3	0 ± 0	10 ± 14	5 ± 9	0 ± 0	12
<i>Lumbricus</i>	<i>terrestris</i>	Linnaeus (1758)	epi-anecic	1 ± 4	0 ± 0	0 ± 0	1 ± 3	23 ± 41	2 ± 2	9
<i>Aporrectodea</i>	<i>giardi</i>	Savigny (1826)	strict-anecic	25 ± 28	19 ± 34	47 ± 30	22 ± 24	27 ± 32	0 ± 0	26
<i>Aporrectodea</i>	<i>longa longa</i>	Ude (1885)	strict-anecic	34 ± 39	22 ± 17	13 ± 30	46 ± 39	11 ± 11	0 ± 0	26
<i>Aporrectodea</i>	<i>caliginosa caliginosa</i>	Savigny (1826)	endogeic	30 ± 32	23 ± 22	41 ± 34	94 ± 57	168 ± 113	25 ± 27	41
<i>Allolobophora</i>	<i>chlorotica chlorotica</i>	Savigny (1826)	endogeic	25 ± 44	1 ± 3	0 ± 0	1 ± 2	0 ± 0	1 ± 2	8
<i>Allolobophora</i>	<i>icterica</i>	Savigny (1826)	endogeic	196 ± 74	251 ± 96	203 ± 101	215 ± 125	193 ± 126	106 ± 75	46
<i>Allolobophora</i>	<i>rosea rosea</i>	Savigny (1826)	endogeic	23 ± 22	5 ± 6	8 ± 9	32 ± 28	30 ± 13	20 ± 13	38
<i>Allolobophora</i>	<i>minima</i>	Savigny (1826)	endogeic	11 ± 21	0 ± 0	0 ± 0	7 ± 6	0 ± 0	0 ± 0	8
<i>Allolobophora</i>	<i>limicola</i>	Michaelsen (1900)	endogeic	0 ± 0	0 ± 0	0 ± 0	4 ± 8	31 ± 54	0 ± 0	4
<i>Octolasion</i>	<i>cyaneum</i>	Savigny (1826)	endogeic	0 ± 0	0 ± 0	0 ± 0	0 ± 0	1 ± 2	0 ± 0	1

# DISCUSSION ET PERSPECTIVES

## 1. Synthèse des résultats scientifiques

Au travers des quatre chapitres de cette thèse, nous avons caractérisé les propriétés physiques, chimiques et biologiques de sols issus de l'ingénierie pédologique selon (i) l'âge des sols et le (ii) le processus d'ingénierie appliqué (basique ou élaboré, impliquant un isolement partiel ou total). L'incorporation de compost dans l'horizon de surface des Anthrosols reconstitués de 4 ans était nettement bénéfique à leur fertilité. Entre 0 et 20 cm de profondeur en moyenne, la fertilité chimique du mélange-terre compost était notamment élevée, bénéficiant ainsi sa structuration grâce à une activité lombricienne stimulée par des ressources trophiques encore abondantes, contrairement à l'horizon sous-jacent non amendé, plus compact et deux fois moins bioturbé entre 20 et 50 cm de profondeur. Les communautés lombriciennes de ces sols étaient composées à 90% d'espèces endogées prédominées par *Aporrectodea caliginosa caliginosa* et *Allolobophora rosea rosea*, et aucun impact de l'isolement n'a été identifié sur les communautés lombriciennes. Les Anthrosols reconstitués de 20 ans se différenciaient de ceux de 4 ans par le développement d'une litière d'environ 8 cm d'épaisseur concentrant les ressources trophiques et d'un couvert végétal épais. En comparaison au mélange terre-compost de 4 ans, la fertilité chimique du mélange terre-compost de 20 ans présent entre 8 et 22 cm de profondeur en moyenne était inférieure, associée à une faible richesse nématofaunique et à un pourcentage de biostructures lombriciennes divisé par 1.5. L'isolement total des sols impliquait l'absence de trois catégories écologiques (épigés, épi-anéciques, anéciques stricts) et l'isolement partiel impliquait une abondance totale très élevée ( $> 500$  individus  $m^{-2}$ ), toutefois composée à 80% d'endogés. Les Luvisols anthropisés de 30 ans issus d'une ingénierie basique présentaient trois horizons différenciés, l'horizon de surface concentrant nettement l'activité racinaire et les ressources trophiques, tandis que leur répartition était plus homogène dans les

deux premiers horizons des Anthrosols reconstitués (i.e., litière et mélange terre-compost). A horizons équivalents, la fertilité physique et chimique était nettement favorisée dans les Anthrosols reconstitués par rapport aux Luvisols anthropisés. L'isolement partiel ou total des Luvisols anthropisés n'avait pas d'impacts sur les communautés lombriciennes.

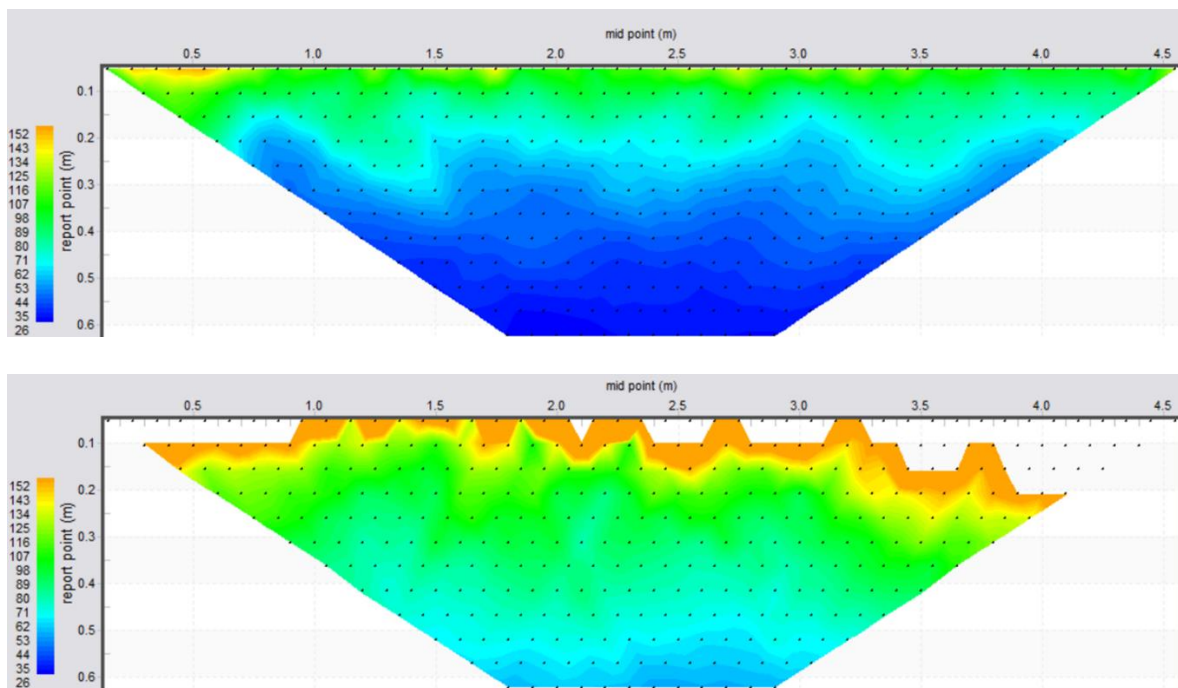
## **2. Suivi des sols et de la biodiversité en faveur de la multifonctionnalité des sols**

Dans le contexte de l'objectif Zéro Artificialisation Nette (ZAN), ces travaux de thèse ont permis de tester et de développer des outils de caractérisation des fonctions de fertilité et de réservoir de biodiversité de sols issus de l'ingénierie pédologique, apportant ainsi un retour d'expérience inédit sur l'efficacité de solutions potentielles de renaturation de sols. Cependant, les référentiels de données robustes sur les propriétés et la biodiversité des sols urbains sont peu nombreux ce qui peut rendre délicates l'interprétation des données et la proposition de préconisations en faveur de leur préservation et de leur gestion (Guilland et al., 2018; Arrouays et al., 2021). En effet, la constitution de référentiels de données urbaines est contrainte par les autorisations d'accès aux parcelles privées ou publiques, la présence de réseaux enterrés qui peuvent limiter l'ouverture de fosses pédologiques, ou encore la forte hétérogénéité des sols urbains rendant difficile la réalisation de répliquats vrais et impliquant la réalisation de nombreux points d'observation pour caractériser une zone urbaine. Toutefois, il existe différents leviers d'action pour développer l'acquisition de données pédobiologiques en milieu urbain.

Des méthodes non invasives telles que les approches géophysiques peuvent être envisagées en complément de fosses pédologiques pour appréhender l'hétérogénéité des sols urbains. La résistivité électrique apparente, par exemple, est corrélée à différents paramètres du sol tels que la teneur en eau, la texture et la teneur en matières organiques (Bronson et al., 2005; Friedman, 2005). Les mesures peuvent être réalisées par injection de courant dans le sol (tomographie de résistivité électrique ; e.g., Michot et al., 2003) comme illustré en Figure 10, ou encore par



génération d'ondes électromagnétiques à la surface du sol permettant de couvrir de vastes surfaces de sol rapidement (e.g., Domsch and Giebel, 2004). Les approches géophysiques apparaissent également prometteuses en complément d'échantillonnages manuels de vers de terre pour la caractérisation des communautés lombriciennes (Valckx et al., 2009; Joschko et al., 2010). Par exemple, Lardo et al. (2012) ont montré que la conductivité électrique du sol pourrait être un bon proxy de l'abondance et de la biomasse lombricienne et permettrait donc de fournir des cartes de la distribution des vers de terre pour des surfaces de sol importantes. Cette méthode réduirait considérablement le nombre d'échantillonnages et les coûts associés (e.g., main d'œuvre et matériel d'échantillonnage).



*Figure 10 - Illustration de pseudo-sections de tomographie de résistivité électrique en dispositif Wenner réalisées sur les Luvisols anthropisés (en haut) et les Anthrosols reconstitués de 20 ans (en bas)*

Les sciences participatives constituent également un levier d'action considérable pour compenser le manque de données en milieu urbain par le développement d'outils simples d'utilisation (Kartinen et al., 2013; Rossiter et al., 2015), favorisant la sensibilisation des habitants aux problématiques des sols et de la biodiversité souterraine de leur jardin, cours d'école ou parcs qu'ils fréquentent (Zoellick et al., 2012; Morales-Doyle and Frausto, 2019)..

Plusieurs programmes de sciences participatives se développent ainsi en Europe et aux Etats-Unis sur les sols (MySoil, LAND PKS, Clé de sol) et les vers de terre (OPAL Soil and Earthworm survey, OPVT, Worm Tracker, Worm Watch). L'identification des individus à l'espèce reste toutefois limitée aux spécialistes. L'accumulation de données lombriciennes en milieu urbain permettrait de développer des outils de gestion efficaces et d'envisager la construction d'outils de modélisation prédictive d'abondance, de présence de catégories écologiques ou encore de niveaux de contribution des lombriciens aux fonctions des sols selon les paramètres du milieu (Huang et al., 2010; Schneider and Schröder, 2012; Palm et al., 2013).

### **3. Cinétique de restauration des communautés lombriciennes**

Pour la première fois, cette étude menée dans les sols anthropisés d'accompagnement de voirie a permis de mettre en évidence une cinétique de restauration des communautés lombriciennes à l'issue de travaux d'aménagement (court et long terme). Cette cinétique dépendait entre autres des propriétés du milieu, des communautés lombriciennes ayant survécu aux phases de travaux (i.e., en ingénierie basique : individus préservés dans les parties inférieures du solum et apportés via les terres et les mottes d'arbres plantés ; i.e., en ingénierie élaborée individus uniquement apportés par les terres et les mottes d'arbres plantés) et de la capacité d'individus alentours à (re)coloniser le sol anthropisé. Sur la base de nos résultats, établis dans un contexte pédoclimatique précis, la Figure 11 permet d'illustrer la restauration des paramètres lombriciens (abondance, biomasse, richesse totale) suite à une phase travaux impliquant une ingénierie basique conservant une partie du profil de sol initial (en orange) et une ingénierie élaborée associée à une excavation de l'ensemble du profil de sol initial (en rouge). Ces résultats soulignent les effets fortement impactant des processus de travaux sur les

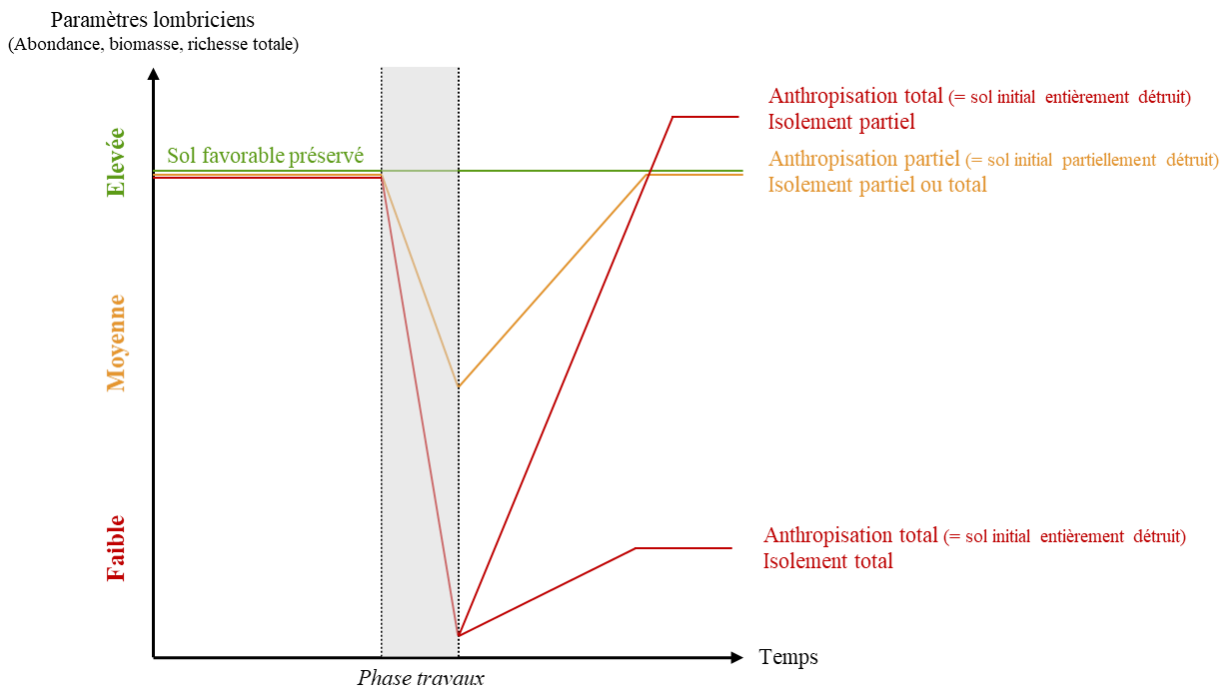


Figure 11 - Schéma simplifié de la cinétique de restauration des communautés lombriciennes suite à une phase travaux (d'après Maréchal et al., 2021)

communautés lombriciennes, qui peinent notamment à se restaurer à la suite d'une excavation de l'ensemble du profil de sol cumulée à des conditions d'isolement total, impactant ainsi les fonctions du sol réalisées par les lombriciens. Dans ces cas de figure, plusieurs solutions peuvent être explorées pour limiter la dégradation des communautés. Tout d'abord, des techniques d'inoculation permettraient de compenser un déficit d'abondance ou de catégories écologiques à un instant  $t$ . Dans la limite d'un milieu d'accueil favorable, il peut s'agir d'introduction de lombriciens adaptés au milieu d'accueil par prélèvements dans un milieu naturel local, ou encore d'implantation dans le sol d'accueil de blocs de sols contenant des lombriciens (Brun et al., 1987; Butt et al., 1995, 1997). Ensuite, l'accès des vers de terre aux sols de terre-plein centraux (i.e., en isolement total) par les voies de surface pourrait être facilité par la suppression d'obstacles verticaux ascendants comme des bordures de voirie. La réalisation d'un profil en travers avec un niveau plus bas au centre de la route faciliterait l'entraînement des lombriciens vers le centre de la route via les eaux de ruissellement et

limiterait la discontinuité entre les deux milieux séparés par la route (Figure 12). Ce dispositif serait notamment favorable aux espèces se dispersant par la surface (épigés, anéciques).



*Figure 12 - Exemple de noue d'infiltration en terre-plein central (Orsay) : le long de la noue central, la bordure séparant la zone plantée du revêtement de chaussée est arasée : elle ne fait pas obstacle à une dispersion active de surface*

Dans cette thèse, la dispersion active des lombriciens sous les surfaces minérales (routes, trottoirs, pistes cyclables) n'a pas été étudiée mais l'hypothèse d'une discontinuité forte a été sélectionnée. En effet, ces revêtements minéraux doivent être soutenus par une portance mécanique suffisante pour éviter sa déformation suite aux passages répétés des usagers, impliquant la compaction de couches sous-jacentes constituées de sables ou de graves traités à la chaux, constituant des environnements inhabitables pour les lombriciens. Cependant, il existe des solutions alternatives telles que les sols fertiles en mélanges terre-pierres, pour des infrastructures de type parking ou piste cyclable, qui permettraient le passage des lombriciens dans la terre entre le squelette minéral. Cette solution, qui reste à étudier en termes de dispersion lombricienne, pourrait s'assimiler à un « lombriduc » favorable notamment aux vers endogés restant continuellement dans le sol. Enfin, notre étude a mis en évidence l'intérêt de la conservation des continuités verticales via la préservation d'une partie du profil de sol lors de phases travaux. Des préconisations supplémentaires pourraient être envisagées dans l'emprise

des travaux pour éviter la compaction de ces zones en particulier (e.g., interdiction d'en faire une piste de chantier, décapages et remblaiements légers)

La mise en œuvre de ces solutions peut s'accompagner de suivis scientifiques pour évaluer la capacité d'un ouvrage à servir de corridor, rendant possible une dispersion active des individus à la surface du sol et/ou sous des revêtements minéraux, en assurant la continuité entre les milieux pour les organismes du sol à faible mobilité (i.e., identifier le caractère franchissable ou infranchissable des obstacles anthropiques). Pour ce faire, des observations directes de terrain par capture-marquage-recapture (Butt et al., 2009; Mathieu et al., 2018), ou l'outil génétique peuvent être employés (Dupont et al., 2015; Mathieu et al., 2018). Cette dernière approche se base sur l'étude de marqueurs moléculaires (i.e., séquences ADN très variables) dont la variation permet de comparer des profils génétiques entre populations voisines, d'identifier d'éventuels brassages génétiques occasionnés par des échanges effectifs, et ainsi de trancher sur l'efficacité de la continuité ou au contraire sur l'importance de la fragmentation (Torres-Leguizamon et al., 2014; Dupont et al., 2017).

#### **4. Préservation des sols et de la Trame Brune dans les projets d'aménagement via la séquence ERC**

Dans un contexte d'érosion majeure de la biodiversité mondiale, les indicateurs de fertilité et de biodiversité développés dans la thèse peuvent constituer des outils d'évaluation opérants de l'artificialisation des sols suite aux évolutions réglementaires récentes. Les enjeux de l'artificialisation ont été considérés dans ce travail à l'échelle du projet d'aménagement où l'ingénierie pédologique constitue une réponse à la renaturation de sols. Afin d'optimiser et de généraliser la préservation des sols et des continuités écologiques à cette échelle, des entrées réglementaires supplémentaires sont envisageables dans le cadre de l'évaluation

environnementale<sup>15</sup>. En effet, le volet pédologique des études d'impact se restreint le plus souvent à la délimitation de zones humides ainsi qu'aux diagnostics de pollution des sols, ces deux axes étant soumis à des obligations réglementaires régies par le code de l'environnement (articles L211-1 et L556-1 à L556-3). De plus, aucun indicateur qualitatif ou quantitatif permettant de mesurer les incidences, positives ou négatives, du projet sur les sols voire sur leur biodiversité n'est appliqué à ce jour. Pour pallier à ce manque de prise en compte des sols et de leur biodiversité dans les décisions d'aménagement, il serait possible d'ouvrir un compartiment réglementaire légitime et opérant dans le cours des études d'impacts menées par les acteurs de l'évaluation environnementale (e.g., écologues, pédologues). A l'issue de celle-ci, les contours d'une Trame Brune potentielle pourraient être dessinés grâce à l'identification réservoirs et de corridors. La mise en place du projet pourrait se faire par le prisme de la séquence ERC (« Eviter, Réduire, Compenser ») intégrant les sols et leur biodiversité, lombricienne dans notre cas d'étude, afin d'assurer des itinéraires techniques à faible impact (Figure 13). Une séquence ERC similaire pourrait être également prise en compte à l'échelle de la planification urbaine<sup>16</sup>, agissant en amont sur l'évitement de l'aménagement dans des zones à fort intérêt pédologique et biologique. La planification territoriale peut également opérer sur la densification du bâti et la hauteur des bâtiments, impliquant la construction de formes urbaines variées avec des impacts potentiellement différents sur les sols et leur biodiversité, dans une optique de réduction d'impact dans les zones aménagées. Une démarche vertueuse en faveur des sols et de leur biodiversité, de la planification territoriale aux projets d'aménagement, est proposée en Figure 14.

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<sup>15</sup> L'évaluation environnementale est un processus visant à intégrer l'environnement dans l'élaboration d'un projet, ou d'un document de planification, et ce dès les phases amont de réflexions. Elle doit rendre compte des effets potentiels ou avérés sur l'environnement du projet, du plan ou du programme et permet d'analyser et de justifier les choix retenus au regard des enjeux identifiés sur le territoire concerné (source : <https://www.ecologie.gouv.fr/levaluation-environnementale>)

<sup>16</sup> La planification urbaine et stratégique regroupe l'essentiel des documents de planification qui organisent l'aménagement de l'espace sur les territoires (source : <http://outil2amenagement.cerema.fr/planification-urbaine-et-strategique-r1.html>)



Figure 13 - Séquence ERC intégrant les sols et leur biodiversité lombricienne aux étapes de conception et de réalisation du projet d'aménagement



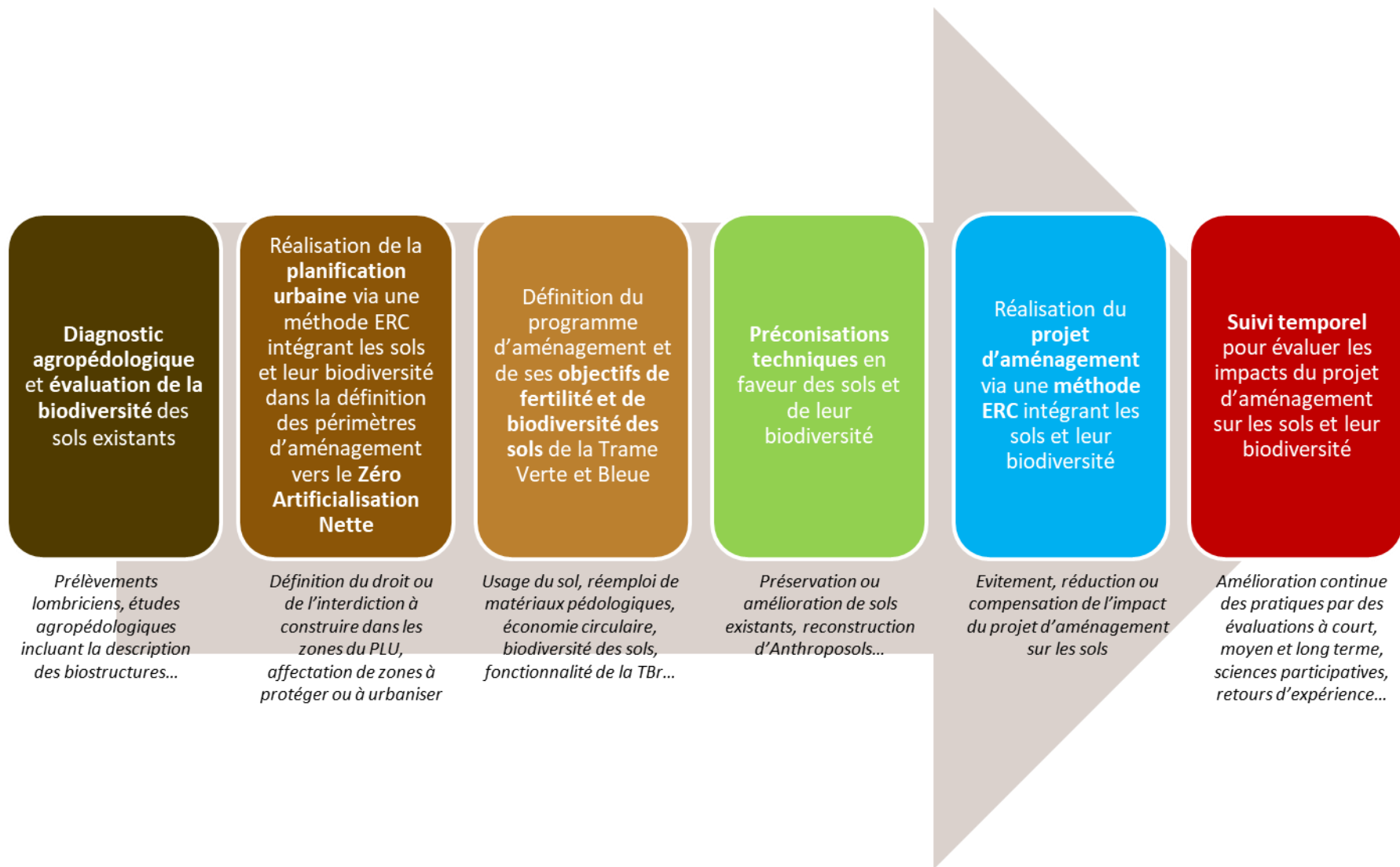


Figure 14 - Démarche vertueuse en faveur des sols et de leur biodiversité, de la planification territoriale aux projets d'aménagement



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**Titre :** Sols fertiles et Trame Brune en milieu urbain : Impacts de l'ingénierie pédologique sur les sols et les communautés lombriciennes

**Mots-clés :** Anthrosoles, artificialisation, renaturation, vers de terre, continuité écologique

**Résumé :** Les sols et la biodiversité qu'ils abritent fournissent de nombreux services écosystémiques participant à l'amélioration du cadre de vie en milieu urbain. Cependant, l'artificialisation croissante constitue un facteur majeur de dégradation des sols et d'érosion de la biodiversité. L'ingénierie pédologique apporte des réponses techniques pour la reconstitution de sols fertiles support de végétation. Peu d'études ont permis à l'heure actuelle de caractériser l'efficacité à court et long terme de ces processus d'ingénierie sur les propriétés physiques, chimiques et biologiques de ces sols en conditions réelles de projets d'aménagement. De plus, ces processus n'incluent aucune prise en compte de la biodiversité des sols, et en particulier des vers de terre, acteurs clés de l'écosystème sol. Ces derniers sont susceptibles d'être fortement impactés par les perturbations engendrées lors des étapes de reconstitution de sol mais également par la rupture des continuités écologiques inhérente au milieu urbain, isolant les sols et les populations lombriciennes par des barrières anthropiques telles que des routes ou des trottoirs. Le premier objectif de la thèse était de caractériser à l'aide d'une quarantaine d'indicateurs les composantes physiques, chimiques et biologiques de la fertilité de Luvisols anthropisés de 30 ans issus d'une ingénierie basique impliquant un décapage peu profond et aucun amendement organique, et des

Anthrosoles reconstitués de 4 et 20 ans issus d'une ingénierie élaborée impliquant un décapage profond et un amendement organique. Le deuxième objectif était de caractériser les communautés lombriciennes dans ces mêmes sols et d'appréhender les impacts de leur isolement, partiel ou total, par rapport à de potentiels réservoirs. Les résultats montraient dans les Anthrosoles un effet très positif de l'amendement organique sur la fertilité, stimulant particulièrement l'activité lombricienne dans les horizons de surface des sols de 4 ans. Vingt ans après leur mise en place, la fertilité des Anthrosoles était supérieure à celle de Luvisols anthropisés non amendés, notamment en termes d'activité lombricienne et racinaire. L'isolement n'avait aucun impact sur les communautés lombriciennes des Anthrosoles de 4 ans et les Luvisols anthropisés tandis que l'isolement total engendrait une faible abondance et la perte de trois catégories écologiques dans les Anthrosoles de 20 ans. Cette thèse met en évidence les impacts contrastés des processus d'ingénierie pédologique sur les fonctions de fertilité et de réservoir de biodiversité lombricienne ainsi que sur l'intégration des sols au sein d'une Trame Brune urbaine permettant les déplacements des lombriciens au sein de la matrice urbaine.

**Title:** Fertile soils and brown infrastructure in urban areas: impacts of soil engineering on soils and earthworm communities

**Key-words:** Anthrosoles, artificialisation, renaturation, earthworms, ecological continuity

**Abstract:** Soils and their biodiversity provide numerous ecosystem services which improve the well-being of citizens in urban areas. However, increasing artificialisation is a major factor in soil degradation and biodiversity erosion. Soil engineering provides technical solutions for the reconstitution of fertile soils supporting vegetation. Few studies have been conducted to characterise the short- and long-term efficiency of these engineering processes on the physical, chemical and biological properties of these soils, under real conditions of development projects. Moreover, these processes do not take into account soil biodiversity, and in particular earthworms, key actors of the soil ecosystem. They are likely to be strongly impacted by the disturbances caused by soil reconstitution stages but also by the ecological discontinuities inherent to the urban environment, isolating soils and earthworm populations by anthropogenic barriers such as roads or sidewalks. The first objective of the thesis was to characterise the physical, chemical and biological components of the fertility of 30-year-old anthropised Luvisols resulting from a basic engineering process involving shallow stripping and no amendment, and re-

constituted Anthrosoles of 4 and 20 years old resulting from an elaborate engineering process involving deep stripping and organic amendment. The second objective was to characterise the earthworm communities in these soils and to understand the impacts of their isolation, partial or total, from potential reservoirs. The results showed a very positive effect of the organic amendment on fertility in Anthrosoles, particularly stimulating earthworm activity in the surface horizons of 4-year-old soils. Twenty years after their implementation, the fertility of Anthrosoles was higher than that of unamended anthropised Luvisols, particularly in terms of earthworm and root activities. Isolation had no impact on the earthworm communities of the 4-year-old Anthrosoles and the anthropised Luvisols, while total isolation resulted in low abundance and the loss of three ecological categories in the 20-year-old Anthrosoles. This thesis highlights the contrasting impacts of soil engineering processes on the functions of fertility and earthworm biodiversity reservoir, as well as on the integration of soils within a brown infrastructure allowing earthworm dispersal within the urban matrix.