

## **Impact of conservative agricultural practices on phosphorous availability on agricultural soil in Loess plateau, Belgium**

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IMPACT OF CONSERVATIVE AGRICULTURAL  
PRACTICES ON PHOSPHOROUS AVAILABILITY ON  
AGRICULTURAL SOIL IN LOESS PLATEAU,  
BELGIUM

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MARTIN VAN DEN ABBEELE

TRAVAIL DE FIN D'ÉTUDES PRÉSENTÉ EN VUE DE L'OBTENTION DU DIPLÔME  
DE MASTER BIOINGÉNIEUR EN SCIENCES ET TECHNOLOGIES DE  
L'ENVIRONNEMENT

Academical year 2019-2020

Co-Promoters:  
Pr. GILLES COLINET(ULg)  
Pr. CLEMENCE MARIAGE (ULg)

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## Abstract

Phosphorus management is a key feature in order to achieve sustainability in agro-systems. In this study, we focussed on the impacts on the soil phosphorus cycle of reduced tillage, cover crop implementation and crop residues restitution. We investigated soil parameters measured in three depths (0-10, 10-20 and 20-30 cm) throughout two long term field experiment started in 2008 and 2011 managed by the experimental farm of Gembloux Agro-Biotech . In addition, acid and alkaline phosphatase activities were measured on the 2020 samples in order to evaluate the biotic pathways of phosphorus mobilisation. Tillage management had the strongest impact on the available phosphorus content showing a concentration of nutrients and total organic carbon on surface. Phosphatase activity was stronger in surface in reduced tillage but lower in depth than conventional tillage. Overall our analysis suggest that conservative agricultural practices promises good leads to reduced the use of mineral P fertilizer in in cultivated Cutanic Luvisol of Belgium.

La gestion du phosphore est un aspect clé d'une production agricole durable. Dans cette étude, nous nous sommes intéressé aux impacts du labour réduit, de l'implantation de cultures de couverture et de la restitution des résidus de culture sur le cycle du phosphore dans les sols. Nous avons étudié les paramètres de sols mesurés sur trois profondeurs (0-10,10-20,20-30 cm) tout au long de deux essais longue durée menés à la ferme expérimentale de Gembloux, Agro Bio Tech commencés en 2008 et 2011. De plus, Les activités en phosphatase acide et alcaline ont été mesurées sur les échantillons de 2020 dans le but d'évaluer les voies métaboliques de mobilisation du phosphore. Les pratiques de labour réduit ont eue l'impact le plus important sur le contenu en phosphore disponible en montrant une concentration en nutriment et en carbone organique total en surface. L'activité en phosphatase a été plus importante en surface pour le labour réduit que le conventionnel. Globalement, notre analyse suggère que les pratiques culturales de conservation présentent des pistes prometteuses pour la réduction de l'utilisation de fertilisants phosphorés d'origine minérale dans des luvisols cutaniques cultivés de Belgique.

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### Abbreviation

Al	Aluminium
AMF	Arbuscular Mycorrhizal Fungi
Ca	Calcium
DSH	Décompactation et travail superficiel
CT	Conventional Tillage
DCP	Dicalcium phosphate
Eh	Electropotential
Fe	Iron
$\text{HPO}_4^{2-}$	Hydrogenophosphate ion
$\text{H}_2\text{PO}_4^-$	Dihydrogenophosphate ion
Ha	Hectare
N	Nitrate
P	Phosphorus
Pi	Phosphorus inorganic
Porg	Organic phosphorus
Pav	Available Phosphorus
Pase	Phosphatase
RT	Reduce Tillage
SOM	Soil Organic Matter
STEP	Station d'épuration d'eau (Wastewater treatment plant)
TOC	Total Organic Carbon
TCS	Technique Culturelle simplifiée (Simplified agricultural techniques)

# 1 Context and literature review

## 1.1 Phosphorus : a global issue

With the increase in worldwide population, and thus an ever growing demand for food, maintaining a sustainable global production is one of the major humanity's challenges of this century. From the middle of the 20th century to the beginning of the 21st, the use of fertilizer in agriculture, mainly in the N P K<sup>1</sup> forms, combined with the mechanisation and cheap energy has permitted a major increase in yield per Ha in a large part of the world. This increase in production efficiency coincided with an increase of the global population (fig1) [Amundson et al., 2015]. Currently, the shortage of resources and available land for agriculture, the threat posed by the climate changes and the degradation of landscape and biodiversity have lead to an increased consciousness of the fragile thread on which our food production system is based [IPCC, 2019].

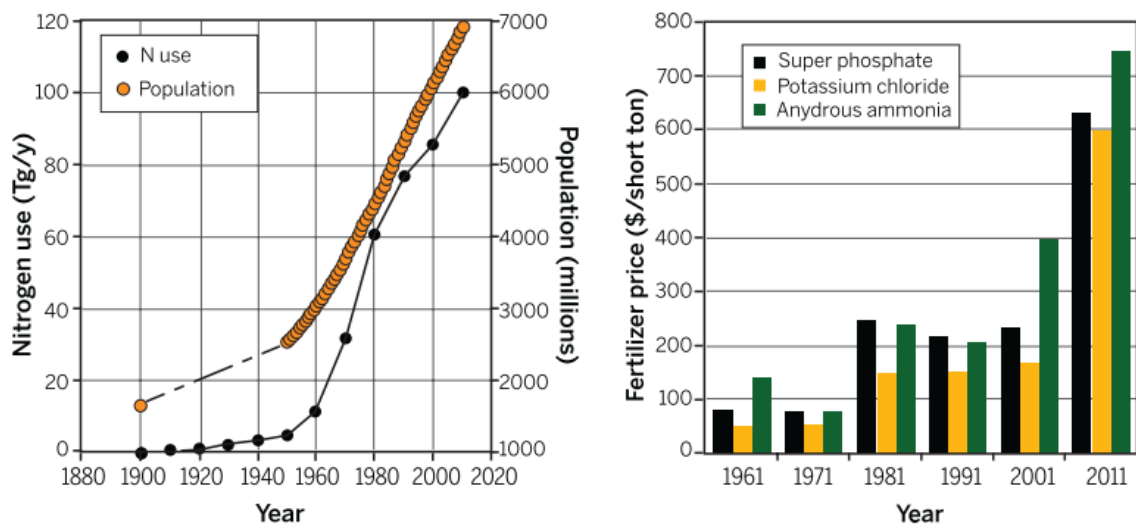


Figure 1: The post-World War II rise in fertilizer production and cost coincides with the spike in global human populations. From [Amundson et al., 2015]

Efficient use of fertilizer in agriculture is essential to reduce environmental impact and increase sustainability. Those fertilizers can be of various types, mainly concerning the 17 essential elements needed for plant growth. Comifer<sup>2</sup> sub-classified the major elements used in fertilization as : Carbon, Nitrogen, Phosphorus, Calcium, Potassium and the oligo-elements. Each of those elements presents its own biogeochemical cycle and different role in the vegetative life development for sometimes very secluded production chain.

Phosphorus possesses its own particularities and Challenges. The financial crisis of 2008 lead to a 800% increase cost of the commodities derived from phosphate rocks, which account for more than 80% of the phosphorus fertilizer commercialized [Smit et al., 2009]. This has shed light on the problematic that the management of this resource represents. A lot of studies have

<sup>1</sup>Nitrate, Phosphorus and Potassium

<sup>2</sup>Comité français d'étude et de développement de la fertilisation raisonnée

then aimed to clarify the main issues this resource brings [Amundson et al., 2015, Linderholm et al., 2012, Smit et al., 2009, Cordell and White, 2011] and investigated new ways for a more sustainable management in agriculture [Amundson et al., 2015, Neset and Cordell, 2012, Walan et al., 2014].

Phosphorus (P) is one of the fundamental building blocks of life on earth. It is one of the 17 essential nutrients for plant growth [Bhattacharya, 2019]. It's a vital component of compounds to build protein, plant structure, seed yield and genetic transfer [Dube et al., 2014] that cannot be substituted. Classified as one of the major nutrients, it is a limiting factor for plant growth in many ecosystems [Elser et al., 2007]. This means it is often deficient for crop and is required in agrosystems in relatively large amount [Bhattacharya, 2019]. The total phosphorus concentration in crops generally varies between 0.1% and 0.5% [Bhattacharya, 2019].

Phosphorus cannot be manufactured or destroyed and one of its particularities is that it's cycle does not have a gaseous phase. The main origin of P in ecosystems comes from the alteration of the bedrock. In natural ecosystems, phosphorus is recycled through organic degradation and uptake by plants over months or years. Historically, in agrosystems, the external P output from the harvest removed from the field was compensated by the application of litter and other manures that could be found around site [Smit et al., 2009]. This way of maintaining good phosphorus balance in the soils can be considered a sustainable approach, with the condition of having : (1) a sufficient amount of available P in the soil and (2) the capacity to bring enough P-rich material on the field. Although this methodology is still in application in most places around the world, a major shift in food production has been witnessed since P fertilizer of mineral origin started of being produced [Smit et al., 2009].

The extraction of mineral P has permitted to extend the cultivation to areas with a natural deficiency in P but in turn, induced a higher dependency with the mining of phosphate rock. Even being the 11th most represented element in the earth crust [Cordell and White, 2011], the deposits with a sufficient concentration in phosphate to be considered an extractable resource are few and well localised. Indeed, Phosphate rock resources occur principally as sedimentary marine phosphorites [Jasinski, 2019] of which the ones with a suitable concentration of P are scarce. Their formation occurs by the transfer of soluble and particulate P from erosion and run-off to the ocean that eventually settles in the sediments. The recycling of these sediments depends on the slow reshaping of the earth's surface making the primary P cycle dependent on tectonic uplift. The cycle closes after  $10^7$  to  $10^8$  years as the P-containing rocks are re-exposed to denudation [Smit et al., 2009] making the use of these deposits a non-renewable material at human time scale.

It is highly unlikely that new potentially exploitable deposits will be found in the future. The global production of phosphate rock in 2018 was around 27000 Mt [Jasinski, 2019] with China and Morocco accounting for more than 50% with the later detaining around 85% of the worldwide reserves known today [Jasinski, 2019]. A risk of depletion is real but the date is hard to assert. Estimation varies between 50 and 1000 year depending mainly on the growth rate taken in computation, worldwide reserves appreciation and the projection in growths of P fertilizer use [Bhattacharya, 2019, Smit et al., 2009, Cordell and White, 2011, Jasinski, 2019].

Another important issue of phosphorus application in agriculture is the risk of eutrophication due to an increased amount of P in natural waters. This mainly occurs due to diffuse loss of phosphorus from field to the water system in cases of over-fertilization in soil with low fixation capacity. Being the limiting factor for algae growth [De Bolle, 2013], the environmental risk must be taken into account in phosphorus fertilization planning.

It is certain that, as long as humanity exists, there will be a global demand for phosphorus [Cordell and White, 2011]. All those features emphasize the need to improve efficiency of phosphorus fertilization. Although a large amount of advances has been made, a lot of work still remains to understand the dynamic of P in the plant-soil system relative to the impact of new agricultural practices used in conservation agriculture. [Faucon et al., 2015] pointed out different strategies that are now in the process of evaluation: cultivar selection of higher P uptake efficiency, intercropping to vary P-sources and new fertilizer type of organic and inorganic forms. With those, at the farmer level, different management practices such as simplified groundwork or implementation of cover crop and residues management have to be explored.

### 1.2 Phosphorus cycle in agriculture

A lot of work have been done in past centuries to understand the different aspects of the phosphorus cycle at the agro-ecosystem level. The absence of gaseous phase and its occurrence in mainly inorganic forms make it differ greatly to the nitrogen (N) cycle present almost exclusively in organic form in soils [Morel et al., 2017]. Taken together, the overall P dynamics in the soil-plant system is a function of the integrative effects of P transformation, availability and utilisation caused by soil, rhizosphere and plant processes [Shen et al., 2011]. Those processes can be divided into three different stages that can be studied separately : the dynamic processes determining (1) P availability in the soil and in the rhizosphere, (2) P mobilisation and (3) P uptake and utilisation by plants.

Figure 2 shows a simplified scheme of P cycle in a crop system. Solid constituents represent the main part of P stock in soil. Studies on french soils suggest values between 665 and 8820 kg P.ha<sup>-1</sup> are contained in the ploughed layer in a cultivated plot [Morel et al., 2017]. In contrast, the amount of P taken by a crop each year is estimated to around 20 kg P.ha<sup>-1</sup> [Morel et al., 2017]. This indicates that the majority of P content in the soil is fixed in forms often non-available to plant.

The major inputs come from the fertilization applied to the field, being in a mineral form, manure or more recently mud from wastewater treatment stations (STEP). The efficiency depends on the form of the fertilizer applied. In comparison, amending phosphate rock directly has shown consistent smaller effect on the succeeding crop whereas more soluble forms of phosphorus are more readily available by plants [Bhattacharya, 2019]. This aspect is explained in more detail in section 1.2.4.

The outputs are of two types : (1) the exportation of P within the harvest which represents the major part (20 to 40 kg P.ha<sup>-1</sup> for wheat), and (2) the loss by lateral transfer of P, runoff, sub-surface flow and vertical P migration. The second is in most cases negligible ( $\pm 1.5$  kg P.ha<sup>-1</sup>) but needs to be taken into account in soils with low buffer capacity. These numbers

are only valid as indicator and do not represent the extreme variability encountered on the field [Morel et al., 2017].

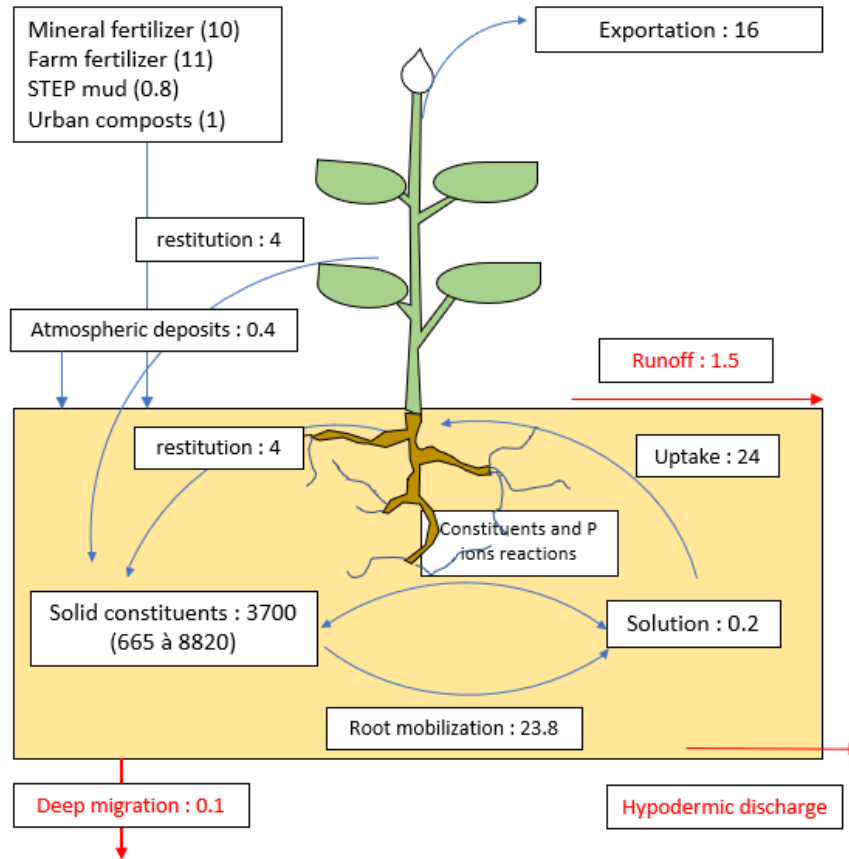


Figure 2: Schematic representation of P stocks and fluxes engaged in the P cycle for a cultural field in France. Fluxes are expressed in  $\text{kg P}\cdot\text{ha}^{-1}\cdot\text{year}^{-1}$ . Numbers have been obtained on data base analysis [Senthilkumar et al., 2012]. The average stock of  $3700 \text{ kg P}\cdot\text{ha}^{-1}$  in the soil corresponds to the ploughed layer in a cultural field. Three-quarter are in mineral forms, a little less than a quarter in organic form and P in Microbial biomass represents only tens of kilograms per hectare. Adapted from [Morel et al., 2017]

### 1.2.1 Forms of phosphorus in soils

To better understand the dynamics linked with the availability of P in a cultural plot, it is important to first define its different forms. P is present in the soil in solution or in solid form. The P concentration in solution can vary of a factor 1000 between  $0.001 \text{ mg/L}$  in tropical acidic soil to  $1 \text{ mg/L}$  in excessively fertilized temperate soil [Morel et al., 2017]. In solution, P can be in a free ionic, mineral or organic form. In agricultural fields of a pH between 5 and 8.4, the main ionic forms are dihydrogenophosphate ion ( $\text{H}_2\text{PO}_4^-$ ) and to a lesser extent hydrogenophosphate ion ( $\text{HPO}_4^{2-}$ ) [Johnston et al., 2014]. The proportion varies depending on the equilibrium constant of the different acid-base couples of orthophosphoric acid. Organic components containing P associated with colloids and polyphosphates have also been detected in solution [Morel et al., 2017]. This part of P is often referred to as the directly available pool for plant.

The solid phase, which represents the majority of P, is distinguished in inorganic and organic forms which bear a different fate in the soil. Inorganic P (Pi) represent 35% to 70% of total P content in the soil. It is composed of primary minerals such apatite and variscite between others<sup>3</sup> and secondary minerals of Calcium (Ca), Iron (Fe) or Aluminium (Al) bases. They constitute the main reserve which releases P to more available forms by weathering [Shen et al., 2011]. The primary minerals are often very stable which means their dissolution rate are too slow to meet crop demand [Shen et al., 2011]. The dissolution rate of the secondary minerals containing P, in contrast, vary mainly according to pH and particle size. Solubility of Fe and Al-phosphate increases with the pH where the Ca-Phosphate decreases [Shen et al., 2011].

The rest of inorganic P is adsorbed on clay or Fe/Al oxides and calcium carbonates. Their release in the solution is regulated mainly by desorption reaction. Depending on the strength of the bond, the P will be more or less available to plant. [Johnston et al., 2014] have defined a scheme categorizing the forms of Pi in the soil into "pools" or "fractions" according to their plant availability (or extractability using chemical reagents) (fig 3) [Bhattacharya, 2019, Johnston et al., 2014]. The available fraction of P in the soil is often referred as the combination of pool 1 (soil solution P) and pool 2 (Surface-sorbed P).

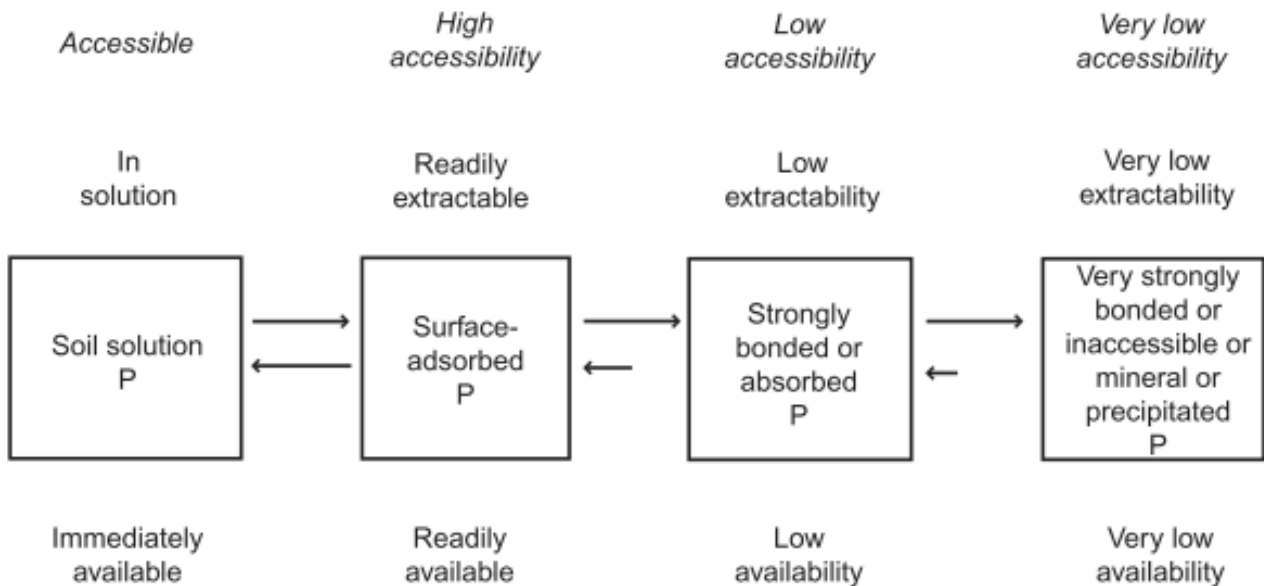


Figure 3: Inorganic phosphorus forms in soil categorized in terms of accessibility, extractability, and plant availability taken from [Johnston et al., 2014]

Organic P (Porg) accounts for 30% to 65% of the total P in the soil [Shen et al., 2011]. It is mainly composed of stabilized forms as inositol phosphates, often called phytates, and phosphonates and active forms as orthophosphate diesters, labile orthomonoesters and organic polyphosphates [Shen et al., 2011, Morel et al., 2017]. The Porg is released into the soil solution through mineralisation processes mediated by soil organisms, plants roots and phosphatases secretions [Shen et al., 2011].

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<sup>3</sup>Hundreds of phosphated minerals have been numbered [Morel et al., 2017]



### 1.2.2 Evaluation of the phyto-available pool of P in a cultural plot

The extraction of P by plants is the major flux of P exportation in a cultural field. Phosphorus is taken up from the soil solution by plant roots principally as orthophosphate ions. According to [Johnston et al., 2014], the most important factors controlling the rate of uptake of P by plant are the concentration of P in solution and the buffer capacity of the soil. The latter controls the rate at which P is released in the solution by desorption/dissolution reactions [Johnston et al., 2014]. The buffer capacity of a soil depends on moisture, temperature, surface physical-chemical properties, pH and Electropotential (Eh).

A common way to characterize soil P is by using P fractionation method using a succession of chemical extractants (NaHCO<sub>3</sub>, HCl, NaOH in different concentrations) but it is a lengthy procedure demanding time and effort not adequate for routine measurements. However, routine measurements of available P is the base for adequate fertilization planning and through-time monitoring. Those monitoring are, in turn, the bases on which the study of processes involved in P fluctuation can be carried out.

Phosphorus effectively available to plant is the sum of P ions in solution and the weakly bonded P on soil particles. Many methods are available to evaluate routinely this pool in the soil. Most of them use some kind of chemical extractant [Renneson et al., 2016, Wuenscher et al., 2015]. The accuracy of this type of methodology poses a serious analytical problem. Not all extractant are fitted for all kind of soils. Acid types often over-evaluate the real amount of P available in the soil by extracting strongly bonded P usually non-available to plant. It is therefore highly recommended consistency in the chemical method chosen for monitoring soils and council fertilization [Johnston et al., 2014, Faucon et al., 2015].

The Olsen method use NaHCO<sub>3</sub> and is the most commonly used worldwide in order to evaluate the available P pool. It often correlates well with Plant P uptake and yield but was initially developed for calcareous soils. In Belgium, routinely measurement of Pav in soil uses mainly two reactants : (1) Lakanen and Ervö method using ammonium acetate and EDTA [Lakanen and Erviö, 1971] and (2) acetate-lactate [Egner et al., 1960].

### 1.2.3 Factor influencing P availability

Long term studies on the processes influencing the replenishment of phosphate ions in soil solution and the evolution of the different pools have shown that in/out fluxes influence the long term dynamic of Pi without perturbing the long term pool of Porg which stays stable [Morel et al., 2017]. Figure 4 shows an integrated view of the different pools and process involved in the soil/rhizosphere continuum. The processes are divided into the soil, rhizosphere and plant processes.

#### 1.2.3.1 Soil processes

In the soil, the dominant mechanism influencing the concentration of P in the solution is molecular diffusion controlled by the gradient of P concentration ( $\pm 10\mu\text{M}$ ). When the plant roots extract hydrogenophosphate ions from soil solution, ions adsorbed on the soil particle will be released and diffused. At the contrary, when a soluble fertilization is applied, the soluble phosphate will be fixed into the soil. The rate is a negative exponential meaning the P will

be rapidly fixed. In contrast, the dissolution of insoluble primary phosphate mineral and the mineralisation of Porg play a marginal role [Morel et al., 2017].

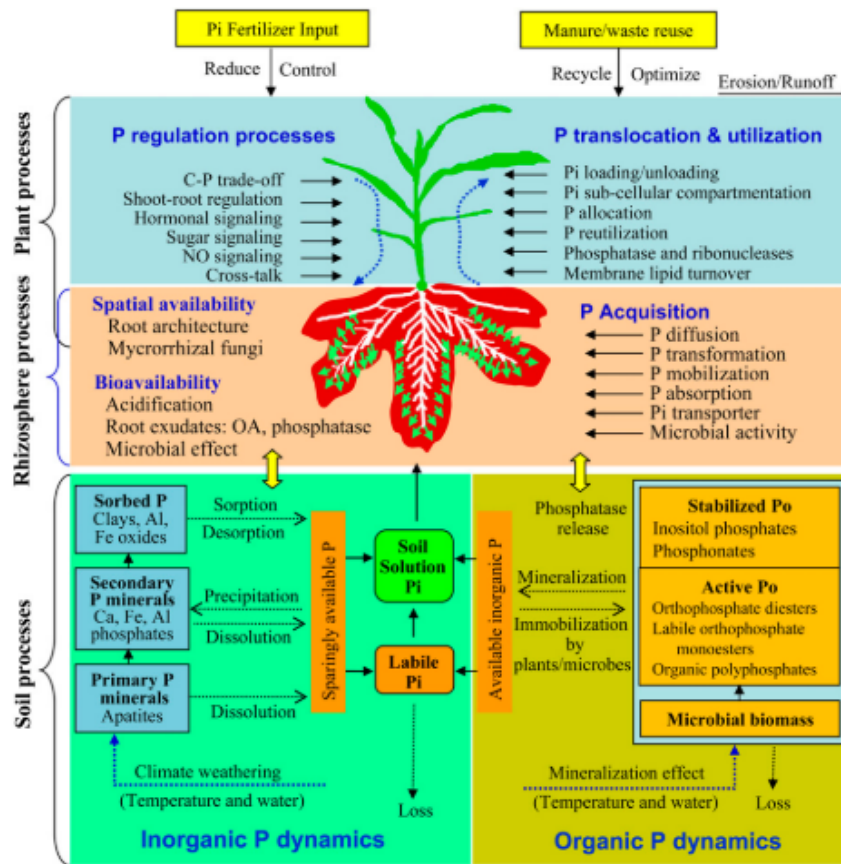


Figure 4: P dynamics in the soil/rhizosphere-plant continuum. C-P, Carbon-P; NO, nitric oxide; OA, organic acids. From [Shen et al., 2011]

P availability in neutral to calcareous soil is controlled mainly by precipitation/dissolution reaction. P can also be adsorbed on surface of calcium carbonate. The precipitation of P in DCP (Dicalcium phosphate) readily available for plant slowly transform into HAP octocalcium phosphate Hydroxyapatite much more stable [Shen et al., 2011]. In acidic soils, the availability of Pi is mainly controlled by sorption/desorption reaction of P adsorbed with Fe and Al oxides.

### 1.2.3.2 Rhizosphere and plant processes

The capacity for a plant to gather P from the soil is greatly determined by its genotype [Hallama et al., 2019]. The main mechanisms at play are : (1) Soil exploration by roots and mycorrhizal hyphae, (2) Mobilisation of sparingly soluble Pi and Porg by oxidation of  $H^+/OH^-$  and carboxylates and (3) mineralisation of Porg by phosphatases [Hallama et al., 2019, Richardson et al., 2011]. The processes engaged in P acquisition vary greatly from a species of plant to another. The soil microbial community can enhance those mechanisms.

The most important trait influencing P uptake is the top soil exploration and root hair density [Richardson et al., 2011]. Root development is greatly influenced by the genotype but Arbuscular mycorrhizal fungi (AMF) and microorganisms also play a fundamental role. The

first one can act as extensions of the root and the second one promotes root growth and modify root architecture via signaling molecules in the rhizosphere [Hallama et al., 2019].

To get P from the sparingly-available  $P_i$  and  $P_{org}$  pools, plants and microbes use what is called P-mining strategies that enhance their desorption and solubilisation. The main mechanism is the exudation of low-molecular-weight organic anions (carboxylates) to dissolve precipitates and chelate metal cations that impair P availability [Hallama et al., 2019]. By facilitating the release of P sorbed via ligand-exchange reactions [Hinsinger, 2001] and blocking binding site on soil particles, carboxylates can increase the concentration of P in the solution [Hallama et al., 2019]. Emission of solubilizing ions seems to be related to P availability and Al toxicity [Richardson et al., 2011] but differs strongly with genotypes and soils [Hallama et al., 2019].

The implication of pH variation on the P dynamics is difficult to predict due to the complex biochemistry of P in soil with the various processes occurring simultaneously [Hallama et al., 2019]. The exudation of  $H^+$  and  $OH^- / HCO_3^-$  changes the pH of the soil solution around the rhizosphere. This modification, in turn, influences the variable surface charge of minerals and soil organic matter (SOM) and can increase P in solution [Hallama et al., 2019]. The emission of proton has only a positive effect in calcareous soil by dissolving Ca-phosphate mineral such as apatite.

Overall, the process of exudation of different organic anions and acidification seems to be complementary but the exact mechanics and their results is hard to assert. Others mechanisms are involved like microorganisms that can be direct sources or sinks of carboxylates and the remobilization of P from the microbial biomass by soil fauna [Hallama et al., 2019].

In addition to the mobilisation processes, the large amount of  $P_{org}$  in the soil needs also to be accessed. To be present in solution and becoming available to plants, the  $P_{org}$  needs to undergo enzymatic hydrolysis using phosphatase and phytase [Richardson et al., 2011]. The production of such enzymes is greatly dependent on the genotype of the plant and their exudation is limited to few centimeters from the roots due to sorption processes. Other strategies imply exudation of sugars further in the soil to nourish micro-organism capable to emit such enzyme. Their natural contribution to the mineralisation is undisputed but their contribution are often hard to assess due to analytical constraints. Acid phosphatase can be emitted by plant and microorganisms where alkaline phosphatases are only produced by microorganisms [Hallama et al., 2019].

The strategies used by plants vary greatly between species. This has lead to the creation of aptitude classes to characterize different cash crops according to their demands in available P in the soil for optimal yield [Morel et al., 2017].

### 1.2.3.3 Influence of soil biota on P cycle

Soil P availability can also be influenced by soil biota [Richardson et al., 2011]. The main effect is double : it can decrease the amount of P in solution by immobilisation when manure is applied in the soil on one hand, and release P from SOM due to the stimulation of mineralisation of  $P_{org}$  from soil biomass and its subsequent hydrolysis by microbial and plant derived phosphatases on the other [Richardson et al., 2011]. Current knowledge tends to say that both plant and soil determine microbial community composition [Hallama et al., 2019].

The impacts of the meso and macro-fauna can also be non-negligible. They have been subject of great focus in past decades and their important role in the nutrient dynamics is more and more recognized [Faucon et al., 2015]. Studies have shown a higher P availability and marked changes in the biogeochemical status of P in the soil in the presence of earthworms. Their presence seems to decrease the amount of organic P and increase the Pi content [Chapuis-Lardy et al., 2011]. In contrast, collembola seems to reduce P uptake in presence of AMF and their presence has been positively correlated with P concentration in the environment [Faucon et al., 2015].

### 1.2.4 Phosphorus Fertilization : process and efficiency

The main goal of soil P management is to assure the expression of potential yield permitted by other limiting factors without affecting the environment by over-fertilization. Other way to phrase it is to get to the agronomic threshold without reaching the environmental threshold (fig 5) or to balance the exportation from harvest and keeping a good level of available P in the soil. Due to the process of precipitation/adsorption and immobilisation by microorganisms, the amount of P added to the soil is quickly fixed into non-available forms for plant making an efficiency return usually low ( $\pm 20\%$ ) [Johnston et al., 2014, Bhattacharya, 2019].

Agronomic threshold is often defined as the critical P value [Johnston et al., 2014, Bai et al., 2013, Xin-kai et al., 2012, Tang et al., 2009]. Critical P values represents the amount of measured available soil P from which any increase will not affect the succeeding yield (fig 5). Critical P values vary from plant to plant and types of soil. Their values are usually obtained through local long term experiments [Johnston et al., 2014].

Environmental threshold (fig 5) corresponds to the point where the soil buffer capacity is overloaded and any extra dose of P fertilizer will most likely results in P leaching and runoff impacting greatly the environment. This threshold is difficult to quantify and greatly dependent of the pedo-climatic context, especially the soil buffer capacity. The main issue with overfertilization is the environmental impact it represents, mainly by polluting the water system. Over-fertilization seems to have other impact on the soil such as limiting the mycorrhizal development [Grant et al., 2005].

Soil analysis is an essential tool in fertilization council. The main idea is to calculate the offer in available P into the soil and the demand from the following crop. Currently, in Belgium, the results from soils analysis are compared to pivot values depending on the soil pH and texture in order to evaluate the needs of the field. P fertilization efficiency is often really low because of the fixing processes. But the fixed P is not lost and long term fertilization experiment showed that it is becoming available throughout cropping seasons [Johnston et al., 2014, Poulton et al., 2013, Bruwier, 2019, Neyroud and Lischer, 2003].

The type of fertilizer also has great importance. The rate at which a fertilizer of P is fixed and then released into the soil is greatly dependent on the component solubility [Morel et al., 2017]. Good indicators for the selection of the product to apply are the soil pH and the total Ca content [Morel et al., 2017]. Quantities to apply are calculated according to the availability of the product and the estimation of the following crop uptake and exportation. The application of organic compounds such as manure and mud from STEP pose a difficulty by their variable P content and the different pathway taken by its release into bio-available forms.

In this purpose, [Renneson, 2018] calculated in her thesis equivalence ratio between inorganic and organic fertilizer. Her results gave a value of 1 for manure applied on soils in the Loess plateau in Belgium.

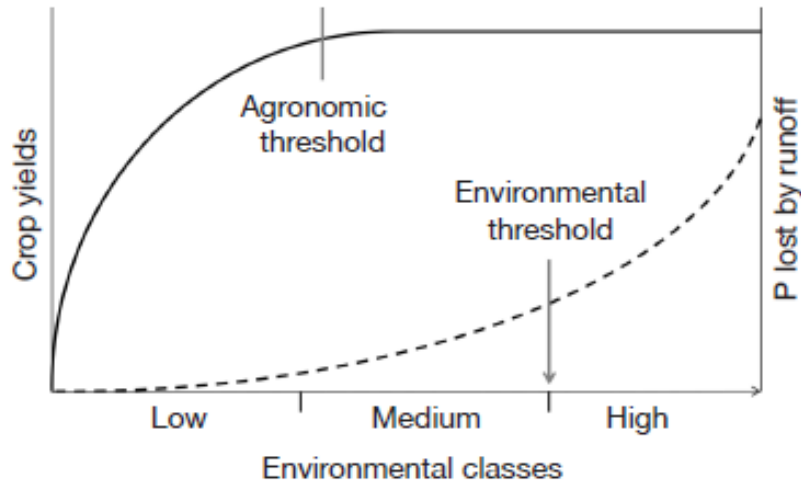


Figure 5: Representation of agronomic and environmental threshold's determination [Renneson, 2018]

### 1.3 Influences of agronomic measures on P Cycle

Phosphorus can be a great limiting factor in crop production and its management is not straightforward. Different management practices can be used to increase P use efficiency at the field level (fig 6). Apart from simple fertilization, where the modalities in fertilizer choice and application method can have strong influences, those strategies are divided into two main areas : (1) the root/rhizosphere management and (2) soil based P management [Shen et al., 2011]. Those strategies, when applied in the good conditions could drastically reduce critical P value at the same time of the environmental impact.

From a farmer perspective, those managements translate themselves into a reduced number of tools called agronomic practices or measures. These measures in intensive agricultural land concern mainly : (1) tillage practices, (2) cover crop implementation, (3) residue management and (4) cultivar selection of P efficient or P mobilisation species. Their impact on P cycle in agriculture has already been reviewed [Horst et al., 2001, Faucon et al., 2015, Hallama et al., 2019] but a lot of the mechanisms involved and their combined contributions remain unclear making prediction of their impacts on P availability and uptake by plant hard to assert.

The direct impact on the soil comes mainly from tillage practices and residue management but the cultivar selection, in a cover crop for example, can also influence the P dynamic. All of these measures have a relative impact on physical, chemical and biological properties of the soil. Their effect (positive, negative or neutral) on the nutrient dynamic, i.e. Phosphorus, depends greatly on the pedo-climatic context of the field on which they are applied [Hiel et al., 2018, Hallama et al., 2019, Faucon et al., 2015]. This indicates that local assessment is needed if accurate management guidelines are to be defined.

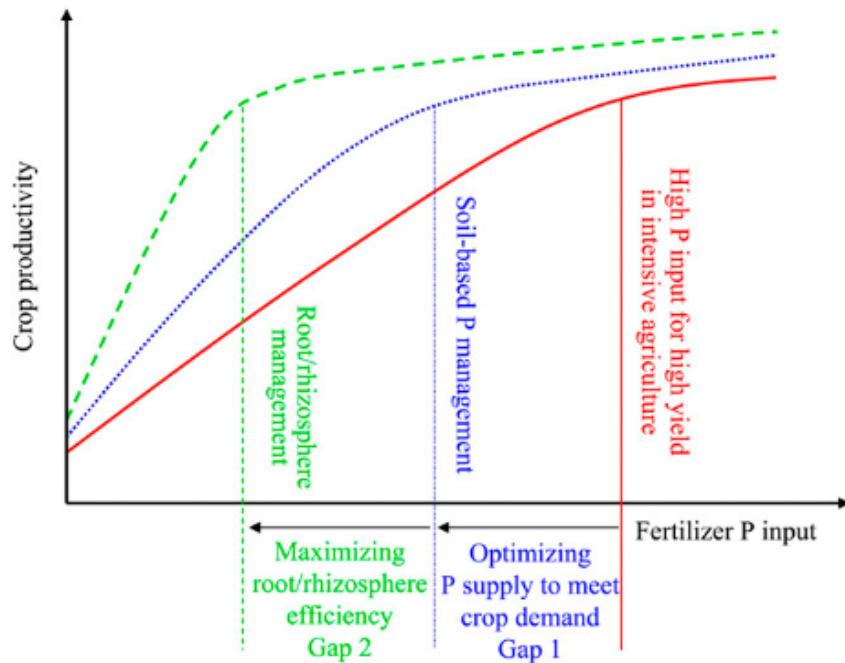


Figure 6: Conceptual model of root/rhizosphere and soil-based nutrient managements for improving P-use efficiency and crop productivity in intensive agriculture. Gap 1 for saving P input can be achieved by soil-based nutrient management for optimizing P supply to meet crop demand. Gap 2 can be realized by root/rhizosphere management for improving P-use efficiency and crop production through exploitation of root/rhizosphere efficiency and further saving P resource input. The red line (solid curve) represents crop productivity response to high-P input under intensive agriculture. The blue line (dotted curve) represents crop productivity response to P input under soil-based P management. The green line (dashed curve) represents crop productivity response to P input under root/rhizosphere management. Taken from [Shen et al., 2011]

### 1.3.1 Management practices : soil tillage

Ploughing plays a major role in modern agriculture. Its beneficial impacts are various as soil structural improvement, weed and pathogen control, preparation of seedbed and influencing nutrient cycle by mixing previous crop residues. But it represents also a large cost in the technical itinerary for crop production and increases risk of soil erosion and compaction. In the sake of conservation agriculture, other forms of tillage practices less invasive and costly can be applied. Although studies on the impact of reduce tillage in temperate Europe shows an average decrease of 4.5 % in yield [Van den Putte et al., 2010], economies it represents on other area make it an interesting option.

Impact of reduce tillage (RT) on the P cycle is still not completely understood. The main positive impact could be to limit erosion leading to P losses and runoff although some studies tend to show an increase in P loss in RT systems [Pittelkow et al., 2014, Cambouris et al., 2017]. In conventional tillage (CT), the residues of previous crop are mixed within the first layer of the soil which get homogenized at each mixing. Direct sowing or complete absence of groundwork, in contrast, can lead gradually to a thin superficial concentration of P by leaving

the residue in decomposition. Long term consequences of this stratification of P in the cultural profile is still poorly evaluated on the stock levels available to roots [Morel et al., 2017].

It is often considered that reduced tillage affects positively earthworms and their abundance and thus affects positively the availability of P. But other studies realized in Belgium could not confirm this hypothesis [Lemtiri et al., 2018].

### 1.3.2 Management practices : residue management

The main idea behind residue management is to remove the least material possible from the field to avoid nutrients losses. The mechanism is to return and incorporate into the soil by mulching or ploughing the residues from the previous crop to keep the nutrient balance close to zero and reduce the need of fertilization.

The impact of residue management on the P uptake by plant is dispersed. It can be positive, negative or neutral [Hiel et al., 2018]. In RT, it can increase the stratification of SOM and accumulate P in the surface by enhancing mineralisation by microorganisms [Tiecher et al., 2012]. But residue fate seems to have stronger effects under climates with less water and drier conditions than Belgium [Pittelkow et al., 2014]. In this region residue fate seems to have less impact on the nutrient cycle and crop performance than the tillage type [Brennan et al., 2014]. Nonetheless, its effect on P dynamic deemed to be investigated, in particular with the introduction of cover crops from which the residues are left on the field after the off-season.

### 1.3.3 Management practices : cover crop

A cover crop can be defined as close-growing crops that provide soil protection, and soil improvement between periods of normal crop production, or between trees in orchards and vines in vineyards [SSSA, 1997]. Their biomass is left on the field after the off-season providing various benefits as erosion reduction, soil organic matter (SOM) build-up, weed and pathogen control, and nutrient management [Hallama et al., 2019]. Their use in Belgium has been greatly increasing in the interest of regulating the nitrous cycle with the obligation of implementing what is called CIPAN<sup>4</sup> or catch crop. Cover crop can be of various types, leguminous or non-leguminous, or a combination of multiple species [Fageria et al., 2005]. Legume cover crop are usually used as a source of N for the following crop where grasses are mainly used to reduce NO<sub>3</sub> leaching and erosion [Fageria et al., 2005]. They appear to have some effect on the phosphorus cycle in the soil but the mechanisms involved are still not yet well understood and their impact hard to predict [Hallama et al., 2019].

Depending on the species used, evidences have shown an improvement in P uptake of the succeeding crop [Takeda et al., 2009, Fageria et al., 2005, Dube et al., 2014]. [Takeda et al., 2009] has observed an increase in phosphatase activity in the soil with a cereal rye cover crop but no effect was observed with rapeseed. [Dube et al., 2014] showed a significant increase of phosphorus concentration in young maize following a cover crop of grazing vetch (*Vicia dasycarpa* L.) and oats (*Avena sativa* L.) but no significant effect on the available pool of P extract by 0.5 M NaHCO<sub>3</sub> (Olsen P). On the other hand, some studies have showed a negative effect of a ruzigrass cover crop on the following soybean crop by reducing P mobility and P resupply [Almeida et al., 2019]. [Tiecher et al., 2012] analysed the phosphatase activity and used

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<sup>4</sup>Culture Intercalaire Piège à Nitrate

P fractionation to assess the long term impact of cover crop application with different tillage practices in a field trial on acidic soil in Brazil. Its results showed an increase in microbial P and phosphatase activity with cover crop especially in reduce tillage.

Overall, cover crop have varying results on the nutrient dynamic but can, in some cases, successfully improve crop yield and P uptake. [Hallama et al., 2019] did a meta analysis on the role of cover crop and soil microorganism on P cycle. Their conclusions emphasize the importance of the site condition and agronomic management. They identify three pathways that act simultaneously and correspond to the hypothesis advanced by [Faucon et al., 2015] (fig 7) : (1) the plant-storage pathway, (2) soil microbe pathway and (3) the biochemical rhizosphere modification pathway.

For the plant storage pathway, cover crops accumulate soil P, sometimes from sparingly available pools, in their biomass. The succeeding mineralisation of the P-rich litter provides then available P for the main crop. The dynamic of the mineralisation is determined mostly by the P concentration in the biomass. This pathway is more relevant for high biomass cover crops such as Poaceae, Brassicaceae and Fabaceae. Poaceae have usually low P concentration that may explain their low efficacy in P enhancer [Hallama et al., 2019].

On the other hand, Poaceae have a strong effect on soil micro-organisms, as for the Fabaceae. By enhancing soil microbial abundance and activity (extracellular phosphatase activity), they can improve P availability for the succeeding crop. Their effect is greater in soils with low P availability where the remaining promoted AMF can be beneficial for the succeeding crop. This corresponds to the microbial pathway.

The last pathway (biochemical rhizosphere modification), describes the process with which some species are capable of mining P pools improving soil P availability even during the main crop phase [Hallama et al., 2019]. The most famous species might be *Lupinus sp.* which changes the rhizosphere by producing large quantities of citrate, one of the most effective organic anion for P mobilisation [Hallama et al., 2019].

Those pathways can be used simultaneously in a mixed cover crop. By implanting different species in a same cover, farmer could optimize its effect on the P cycle and create a positive plant/soil feedback. Figure 7 shows the potential positive plant-soil feedback of multifunctional cover crops to a wheat crop. The cover crop is composed of lentil (*Lens culinaris*), broadbean (*Vicia faba*), white mustard (*Sinapis alba*), and phacelia (*Phacelia tanacetifolia*) in a direct-seeded cropping system. T1 shows the relationship between functional plant diversity of a cover crop including P-mobilizing species, legumes, and diversity of morphological traits (root, stem, leaf) with P availability. This includes enhancement of mineralisation, desorption and solubilisation by emission of phosphatase/phytase, low-molecular-weight organic acids (LOA) and enhancement of soil biota [Faucon et al., 2015].

In T2, P availability is influenced by the residues of cover crop according to two processes : (1) synergistic effects of residue mixtures on litter decomposition and (2) fresh OM of crop residue mixtures would reduce soil organic P and increase P availability by SOM decomposition due to a litter-priming effect. In T3, the heterogeneity of crop residue mixtures decomposing at



the soil surface would have decomposition kinetics that are synchronous to P nutritional needs of the crop [Faucon et al., 2015].

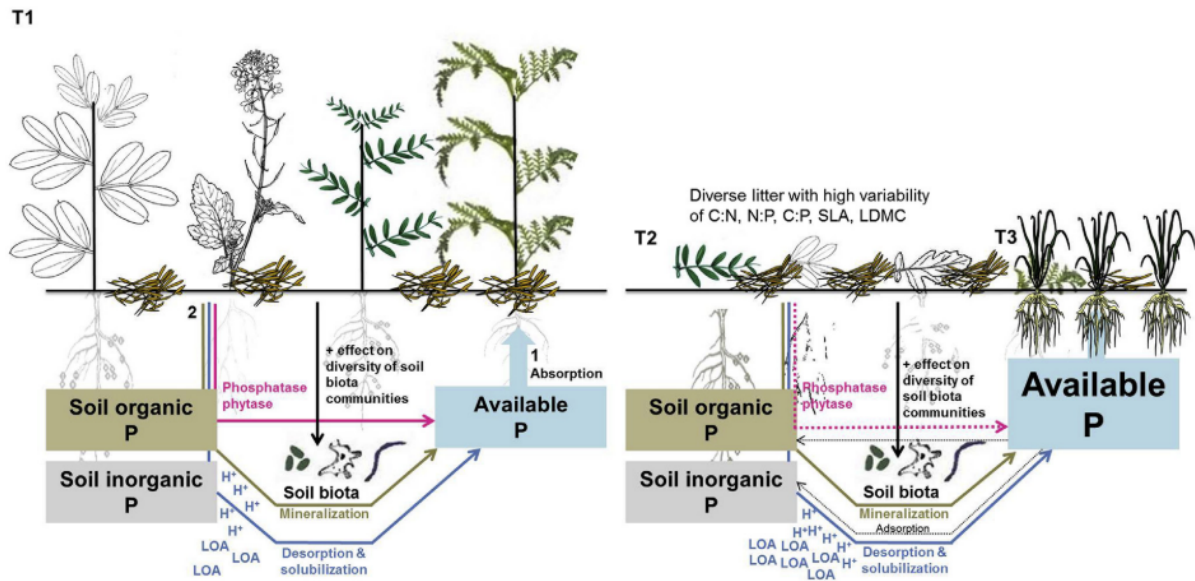


Figure 7: Positive plant/soil feedbacks of multifunctional cover crops to a wheat crop. T1, cover crop growing season, T2, mulching of the cover crop for seedbed preparation, T3, following cash crop growing season. LOA, low-molecular-weight organic acids, e.g., malate and oxalate, that decrease the sorption of phosphate ; SLA, specific leaf area; LDMC, leaf dry matter content. Taken from [Faucon et al., 2015]

## 2 Objective of the study

Conservation agriculture promotes a more integrated approach to the natural mechanisms occurring in the water-soil-plant system. This approach includes some agronomic measures such as reduce tillage or cover crop implementation, of which impacts on the P cycle in the soil is still difficult to predict. P dynamics being greatly dependent on soils properties. Local assessment under specific pedo-climatic conditions is necessary to come to relevant guidelines for modern agriculture [Hiel et al., 2018]. The main goal of this study is to evaluate the P budget of an agricultural field under different agronomic practices in order to distinguish trends that could favour one from another.

Two long term field experiments have been put in place by the faculty of Gembloux agrobiotech : "SolResidus" started in 2008 and "SolCouvert" started in 2011. The first one is testing different residue management systems under conventional and reduced tillage. The second is testing the long term impact of cover crop implementation under different tillage systems. These two experiments, located in the center of the most productive agricultural land in Belgium, offer an incredible opportunity to evaluate medium to long term impact of agricultural practices on the P dynamic.

For the SolResidus experiment, the monitoring of the pedological characteristics of the field trial has been done every 6 months from the start and every year since 2016. For solCouvert, the monitoring started in 2013 with a yearly periodicity. These data can help understand the impact on the P cycle from the different modalities in place.

The first hypothesis is to see a gradual stratification of the SOM and the nutrients of the soil with a higher concentration on the top layer in modalities of reduce tillage whereas a homogeneous repartition for conventional tillage. This can be verified with the pedological data gathered on the field. The first part of the study focus on those data in order to distinguish if : (1)those modalities influence the total Pav in the soil and (2) how did the Pav pool changed since the start of the experiment.

### **Main goal of the current study**

- Evaluate the long term impact on the Pav pool of the use of differential soil practices in an intensive agricultural land in Belgium
- Investigate the mechanisms influencing the P availability dynamic in an agricultural soil in the Loess plateau and their interaction under different agronomic systems.

Soils in the loess plateau are known for being naturally rich in P with often enough return on the fixed P to achieve crop needs. [Bruwier, 2019] has studied a centenary exhaustion trial also managed by the University of Gembloux Agro-Biotech using P fractionation. Her conclusion tended to show that the exported P from the available pool by the harvest was replenished from richer less available pools. Her study showed that the decrease in total P was explained mainly by the decrease of Porg with the Pi pools staying relatively constant through time. Those results underline the fact that the Porg pool is strongly solicited by crops.

## 2. OBJECTIVE OF THE STUDY

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To study the mechanisms involved in the P mobilisation, the phosphatase enzymatic activity was measured in order to see if the different modalities influence this soil parameter. The second part of the work focus then on the dynamics influencing the availability of P and their interaction with the different agronomic practices. The investigation of soil parameters and their fluctuation could help to answer the question : is there a better tillage systems in the phosphorus availability point of view ?

## 3 Methodology

### 3.1 Experimental site

The two experimental fields were established side by side in the vicinity of Gembloux, Belgium (50°33'50"N 49°42'44"E) and cover an area of 1.7 ha each of the experimental farm of Gembloux Agro-Bio Tech, University of Liège (fig 8). SolResidus was established in 2008 followed by SolCouvert in 2011. Yield and soil measurements started in 2010 in SolResidus. Figure 9 shows the monthly rainfall(mm), temperature (°C) values and normal rainfall and temperature (1971-2012) measured in Ernage (at 2.5 km). The area is located in a temperate climate. Both experiments were designed in a latin square with 4 repetitions by modality (fig 10).

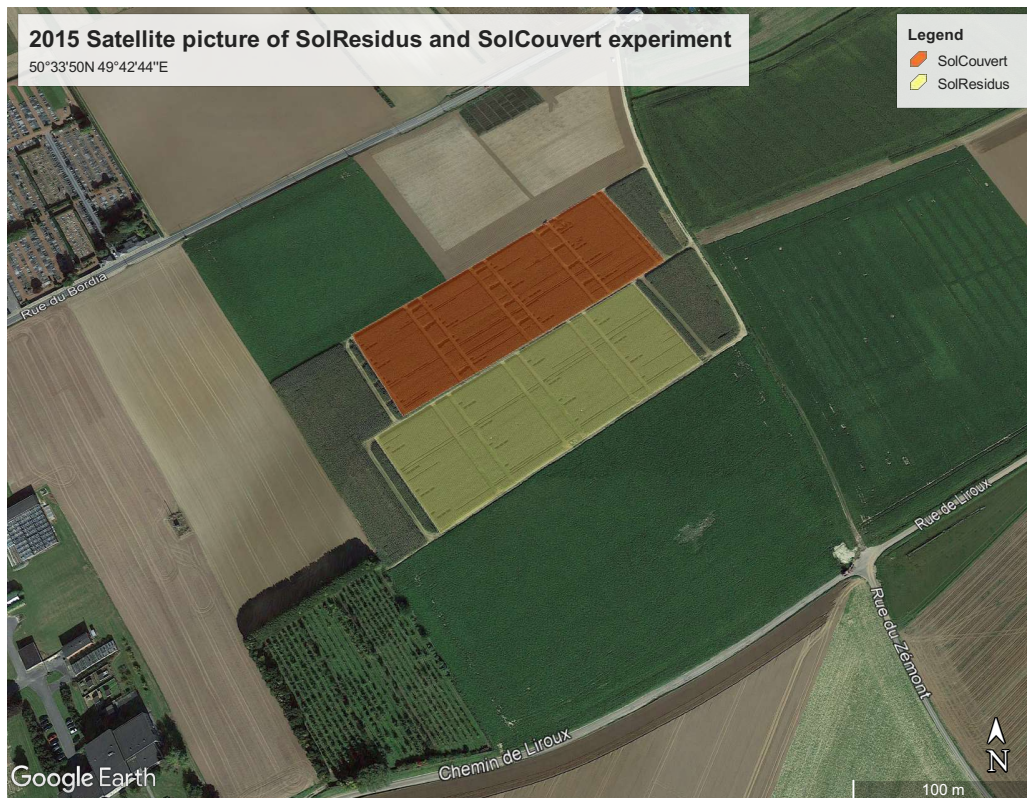


Figure 8: Localisation of the experimental sites SolCouvert et SolResidus

### 3. METHODOLOGY

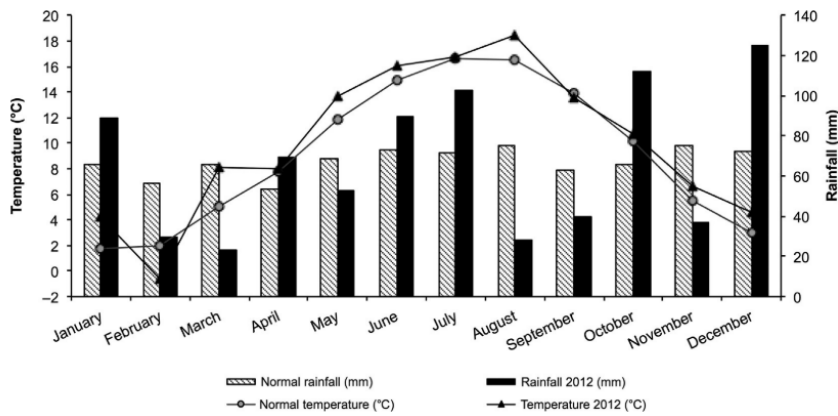


Figure 9: 2012 monthly rainfall (mm) and temperature (°C) values and normal rainfall and temperature (1971–2000) measured at Ernage. Data were recorded from a weather station located at Ernage-Gembloux by CRA-W (Walloon Agricultural Research Centre).

In the field trial SolResidus, the residue quantity depends on the exportation or not of the straw. These residues are then, or buried 10 cm deep by superficial work, or in 25 cm by conventional ploughing. Weeds and pathogen mushroom present on the residues follow the same path. Weed pressure and fungus pathologies can then influence differently potential yield for the next crop. Fresh SOM is also differently distributed in the soil that can result in different nutrient availability in the ryzosphere. Experimental disposition is shown in fig 10. The modalities are as follows and detailed in table 1 :

- Conventional tillage with residue (Lab In)
- Conventional tillage residue out (Lab Out)
- Reduced tillage residue in (TCS In)
- Reduced tillage residue out (TCS Out)

Table 1: Comparison between Conventional and reduced tillage treatments in SolResidus

Period	Conventional Tillage	Reduced Tillage
After harvest		Stubble breaking Tool : tine stubble cultivator Depth : 7-10 cm
Few days before sowing	<b>Ploughing</b> Tool : moldboard plough Depth : 25 cm	No ploughing
Sowing day	<b>Seedbed preparation</b> Tool: dual cultivator with tines and rolls in front of the tractor and rotary harrow followed by wedge ring roller. Depth: 7–10 cm The sowing machine is either mounted behind the wedge ring roller (cereals and faba bean) or either an extra passage with a precision spaced planter (maize) is done. Depth: 2-5 cm	

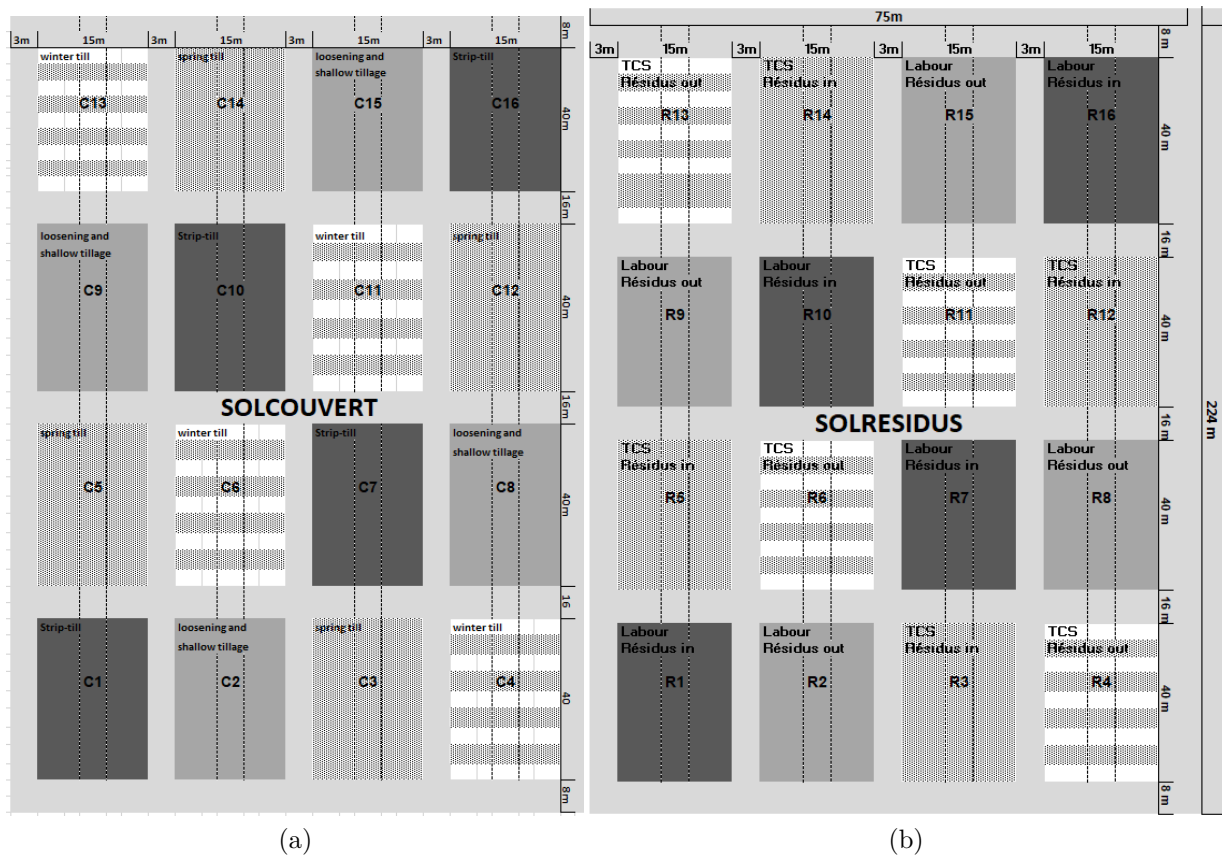


Figure 10: Experimental disposition (plots and modalities) of (a) SolCouvert and (b) SolR sidual

In the SolCouvert experiment, the implementation of a cover crop between wheat harvest and the spring crop allow an important OM production and a competition for weed. Four modalities of cover crop management are tested. The reference practice is an autumn ploughing to destroy the cover followed by a superficial seedbed preparation before plantation (winter ploughing). This allows to bury the fresh biomass from the cover and control poorly developed weeds. The second modality differ in the date of ploughing. In this one, the ploughing is applied just before seedbed preparation. In this itinerary, cover crops and weeds continued to develop. The third one comprise a loosening on 25 cm depth applied before cover crop followed by a superficial preparation for sowing. OM produced by the cover and weeds are mixed in the soil on a 10 cm depth. For the last one, called strip till, seedbed line is only worked on a width of 10 cm and a depth of 15 cm. Residue from the cover and weeds are left in the interlines. On the two last modalities, if the cover has not been destroyed by the cold, a chemical destruction is applied.

For SolCouvert the modalities are denominated as follows and detailed in table 2 :

- Winter Ploughing (WP)
- Spring Ploughing (SP)
- Strip-Till (ST)
- Loosening and shallow ploughing (DSH)

### 3. METHODOLOGY

Table 2: Comparison between tillage treatments in SolCouvert

Period	Spring Ploughing	Winter Ploughing	DSH	Strip Till
After harvest	Stubble breaking Tool : tine stubble cultivator Depth : 7-10 cm			
In Autumn	No ploughing	<b>Ploughing</b> Tool : moldboard plough Depth : 25 cm	<b>Loosening</b> Tool : tooth decompactor Depth : 25 cm	No ploughing
In Spring	<b>Ploughing</b> Tool : moldboard plough Depth : 25 cm	No ploughing		
Sowing day	<b>Seedbed preparation</b> Tool: dual cultivator with tines and rolls in front of the tractor and rotary harrow followed by wedge ring roller. Depth: 7–10 cm  The sowing machine is either mounted behind the wedge ring roller (cereals and faba bean) or either an extra passage with a precision spaced planter (maize) is done. Depth: 2-5 cm			<b>Strip Till</b> Tool : Strip Till Seed bed line worked on a width of 10 cm and a depth of 15 cm

#### 3.1.1 Management History

The successions of crops in the fields are presented in table 3. The ones marked in bold represent cover crops implemented between cash crops. Years without cover crops (2013-2014, 2015-2016, 2017-2018 and 2019-2020) are due to the cropping of winter wheat which is seeded before winter. The only fertilizations applied were of nitrate forms according to crop demand. No Phosphorous-type fertilization was applied. Only sugar lime was sprayed in December 2016 which contained some Phosphorus.

Table 3: Crop rotation applied on the field trial; crops in bold representing the cover crops. SR = SolResidus and SC = SolCouvert.

Period	Crop	Field
2008-09	<b>Rapeseed (<i>Brassica napus</i>)</b>	SR
2009-10	Winter wheat ( <i>Triticum aestivum</i> )	SR
2010-11	Winter wheat ( <i>Triticum aestivum</i> )	SR
2011-12	Winter wheat ( <i>Triticum aestivum</i> )	SR
2012-13	<b>Mustard (<i>Sinapis alba</i>)</b>	SR and SC
2013	Faba bean ( <i>Vicia Faba</i> )	SR
2013	Sugar beet ( <i>Beta vulgaris</i> )	SC
2013-2014	Winter wheat ( <i>Triticum aestivum</i> )	SR and SC
2014-2015	<b>Oats (<i>Avena sativa</i>) and peas (<i>Pisum sativum</i>)</b>	SR and SC
2015	Maize ( <i>Zea mays</i> )	SR and SC
2015-2016	Winter wheat ( <i>Triticum aestivum</i> )	SR and SC
2016-2017	<b>Oats (<i>Avena strigona</i>) and Peas (<i>Pisum sativum</i>)</b>	SR and SC
2017	Sugar beet ( <i>Beta vulgaris</i> )	SR and SC
2017-2018	Winter wheat ( <i>Triticum aestivum</i> )	SR and SC
2018-2019	<b>SIE mix : Vetch 50%, japanese oats 42 % and Phacelia 8 %</b>	SR and SC
2019	Maize ( <i>Zea mays</i> )	SR and SC
2019-2020	Winter wheat ( <i>Triticum aestivum</i> )	SR and SC

## 3.2 Pedological context

The soil for both experiment is a Cutanic Luvisol [WRB, 2015]. According to the 2012 soil analysis, the soil was silty with favourable natural drainage, containing 70–80% of silt, clay content of 18–22% of clay and 5–10% of sand. In 2013, a measuring campaign has been conducted on the SolResidus experimental field giving precious information to interpret the current data. This campaign included a pedological characterization, an evaluation of spatial nutrient variability and temporal dynamic of nutrient (available elements) [Colinet et al., 2013].

For the pedological characterisation, one pit was dug for every modality until 2m depth in April 2011 and one face was described according to a standardized procedure [Delecour and Kindermans, 1980]. Samples were taken in every horizon for laboratory determinations. Soil bulk density (BD) was estimated from soil cylinders (100 cm<sup>3</sup>) taken in three replicates at various depths according to horizons. Main soil characteristics are summarized in table 4.

Ploughing depth used to reach 25 cm before experimentation and even 35cm in the past. No E horizon can therefore be observed under the plough layer. Textural B horizon contains 5 to 10 percent more clay than the A horizon, present a blocky subangular (30 mm size clods) friable structure, and is porous (mainly small size pores) but compact.

Three sub-layers were described and sampled within the A horizon in order to study the effects of tillage modes. The first two sub-layers are very similar under ploughing while the 0-10cm sub-layer is less dense and richer in organic matter under reduced tillage. Numerous clods were platy under modes without residue restitution while biogenic clods (granular and subangular blocky) were more important under the other modes.



Table 4: Main soil properties in the field trial of SolResidus according to cultural practices (Lab: plough / TCS: no plough / IN: crop residues left in the field / OUT: crop residues exported off the field)

	Lab In	TCS In	Lab Out	TCS Out
Topsoil (0 to 25/35 cm)				
Texture*	Silt Loam (Clay: 14-16%; Silt: 75-80%; Sand: 5-6%)			
pH*	Neutral (6,5 – 7,0)			
TOC* (g/100g) / Bulk Density*				
0-10 cm	1,1	1,3   34	1,1   1,38	1,4   1,26
10-25 cm	1,1   1,41	1,1   1,56	1,1	1,2   1,50
25-35 cm	1,1	1,1	1,1   1,53	1,2   1,44
Structure				
0-10 cm	Gran. + Blck sub.	Gran. + Platy	Platy + Gran.	Granular
10-25 cm	Gran. + Blck sub.	Blck ang.	Platy + Gran	Platy
25-35 cm	Platy	Blck ang.	Blck ang. + Gran.	Platy
Subsoil (35-100cm): textural B and B to C transition horizons				
Texture*	Silt Loam (Clay: 20-25%; Silt: 70-75%; Sand: 3-6%)			
pH*	Slightly acidic (6,2 – 6,5)			
TOC* (g/100g) / Bulk Density*				
	0,1 – 0,5 (decreases with depth) / 1,50 – 1,66			
Structure	Blocky angular to Blocky subangular / Compact			

\*Texture measured with automated Robinson pipette; pH: 2:5 w:v ratio, 0,1N KCl, 2h contact;  
 TOC: Total Organic Carbon after Walkley-Black ; Bulk density in 100cm<sup>3</sup>-cylinders;  
 Structure: Gran.: Granular ; Blck ang. : Blocky angular, Blck sub. : Blocky subangular.

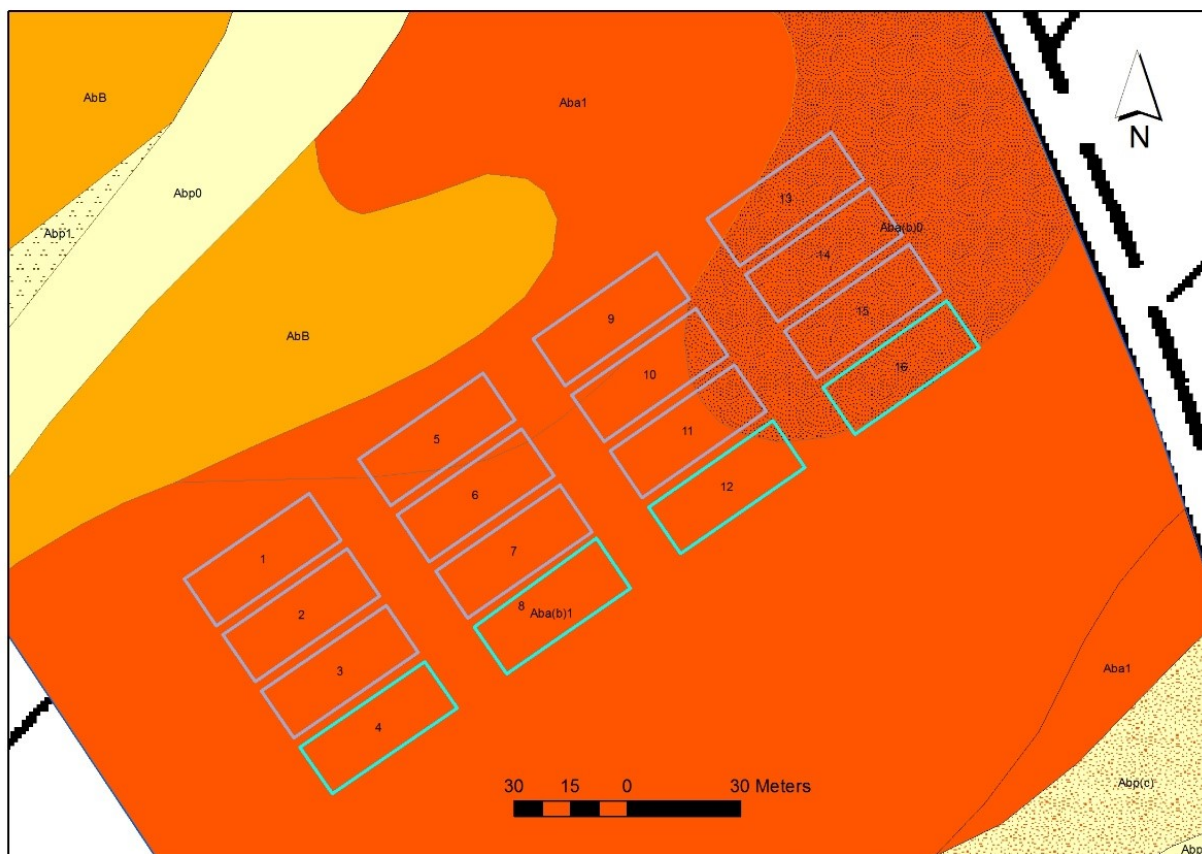


Figure 11: : Plots localisation of SolResidus experiment on soil map. The 16 plots are located on loamy soils with BT horizon with different erosion levels ( $Aba(b)0 < Aba(b)1 < Aba1$ ) and the presence ( $Aba(b)$ ) or not ( $Aba$ ) of taches in BT horizon. A pit has been dug in plots 4,8,12 and 16 during lysimeter implantation.

### 3.2.1 Cartography of the chemical fertility of the experimental parcel of Sol-Residus

The cartography of the chemical fertility of the experimental parcel was conducted on the pH, Total Organic Carbon (TOC) and available elements to evaluate spatial variabilities. This constitutes an important prerequisite to alleviate any bias in the interpretation of further data by incorporating innate differences of the medium.

To do so, 107 samples in an random-stratified scheme have been taken on the all area and analysed. Maps have been created by ordinary krigage with ArcGIS 9.3. Statistical parameter have then been computed for each plot (ANNEXE B). A variance analysis has then been made on the Mean values for each plot obtained from predicted values. In case of null hypothesis reject, a mean comparative test has been made.

### 3.2.2 Monitoring of nutrient mobility in SolResidus

Two method in parallel have been set in order to monitor the nutrient mobility: (1) Yearly investigation of pH, TOC and available values of Ca, Mg, K and P. The aggressiveness of the different analytical method makes it unlikely the existence of a short time variation (ie seasonal variation) of these parameters. And (2), the second approach focus more reactionary compartments and bases itself on water extraction (hot water for carbon). The monitoring was carried out on an monthly basis from October 2011 till October 2012 in eight samplings .

### 3.3 Chemical analysis

At each sampling period, composite samples were taken in each plots of both experiments for three depths: 0-10 cm (A), 10-20 cm (B) et 20-30 cm (C). Before analysis, each sample was dried at 60°C and passed through a sieve at 2mm. For each sampling batch, pH KCl, Total Organic Carbon (TOC) and extractable elements content (P,Mg, Ca and K) were measured systematically. The different methods used to measure soil parameters are listed in table 5. The Pav content was obtained using the Acetate-EDTA extraction [Lakanen and Erviö, 1971].

Table 5: Principles and references of analysis performed on samples

	Principles	Reference
Acido-basic status		
pHH <sub>2</sub> O and pH KCl	Soil :solution ratio 2:5 w:v H <sub>2</sub> O ou KCl 0,1N ; 30min agitation.	NF ISO 10390 Adapted method
Organic status		
Total organic carbon	Sulfo-chromique oxydation in acid medium.	Walkley-Black
(Total Organic Carbon) COT in g.100g-1 Total Nitrogen	Titration by sel de Mohr. Acid Mineralisation in NH <sup>4+</sup> .	ISO 14235 Method Kjeldahl
NT in g.kg-1 Hot water extracted Carbon	Distillation et entraînement à la vapeur, piégage dans H <sub>3</sub> BO <sub>3</sub> , titration. Extrait de 6g dans 60ml, 80°C.	ISO 11261
HWC en mg.kg-1	Oxydation sulfo-chromique en milieu acide. Titrage au sel de Mohr.	
Granulometry	Method par sedimentation after mineralo-organic cement elimination and peptization.	automated method by Robinson's pipette ISO 11277
Extractable elements		
Available P, K, Ca, Mg	extraction by CH <sub>3</sub> COONH <sub>4</sub> + EDTA à pH 4,65 ; 20:100 w:v, 30 min. Assay by atomic absorption or colorimetry (P).	[Lakanen and Erviö, 1971]
P, K, Ca, Mg solubles	Water extraction at 20°C, 20:50 w:v , 30 minutes. Assay by atomic absorption or colorimetry (P)	

#### 3.3.1 Phosphatase activity

To estimate the phosphatase activity, the release of p-nitrophenol from p-nitrophenyl phosphate was measured following exposure to soil in a modified universal buffer (MUB) at pH 6.5 for acid phosphatase activity and at pH 11.0 for alkaline as described by [Tabatabai and Bremner, 1969]. For each sample, 1.0 g of soil was incubated with 1 mL 0.1 M p-nitrophenyl phosphate, 4 mL MUB and 0.2ml of toluene for 60 min at 37°C. After incubation, each sample was filtered quickly with Whattman 2v filter after 1 mL 0.5 M CaCl<sub>2</sub> and 4 mL 0.5 NaOH were added. Samples were then homogenized and the p-nitrophenol formed was measured using a colorimeter at 400 nm.

To evaluate accuracy and analytical variability of the method, a sample was repeated 3 time across every batches. In addition, 2 other samples were repeated in each batch to evaluate the inner variability. The inner variability showed good results with ranging from 1 to 4%. For the acid phosphatase estimation, an analytical variability between batches of 7.4 % has been observed and a variability of 15 % for the alkaline.

The selection of samples to analysed was made following the pedological report [Colinet et al., 2013] that showed no significant interaction in the depth B (10-20 cm) for SolResidus. Thus, only the samples of the depth A (0-10 cm) and C (20-30 cm) were analysed. During the process, samples of the plots 14,15 and 16 of SolResidus were lost reducing the repetition number of these modalities to three instead of four.

#### 3.4 Statistical analysis

Statistical analyses were performed with R software [R Core Team, 2019]. The statistical analyses were systematically applied to assess the effect of tillage management for both experiments and crop residues management for SolResidus on soil measurements, as follows. First, a 2-way ANOVA was performed, including the experiment as fixed factor and the plot position (line and columns of the Latin square) as random factors to check for global differences.

After what, for each experiment, a 2-way ANOVA was performed with modality(RT In and Out, CT In and Out, ST, WP, DSH and SP), depths and their interactions as fixed factors and the plot position (line and columns of the Latin square) as random factors. In case of significative interactions, every depth for the four treatments were intercompared and ranked using a post-hoc test (Student-Newman-Keuls—SNK). Analyses of variance (2-way ANOVA) and SNK tests were performed with the "lme4" package [Bates et al., 2015] and "agricolae" package [de Mendiburu, 2020]. The conditions of application of the ANOVA test (normality of the distribution and homoscedasticity) were systematically checked on the residuals of the ANOVA, using respectively a Shapiro–Wilk test and a Bartlett test.

To study the Pav pool evolution during the year, the same mixed model was used including the date as a random factor. In addition, a student's T-test was used to test for each treatment whether the soil factors of the last sampling year and the first sampling year were significantly different. A PCA analysis was carried out on the 2020 data including the posphatase activity in order to see correlation from the different soil variables using the "FactoMineR" package [Lê et al., 2008].

## 4 Results

### 4.1 Results from 2013 pedological report

The results of the comparative test between plots from cartography of the chemical variability of the experimental parcel of SolResidus of 2013 are shown in ANNEXE B. A moderate but non-negligible variability at site scale has been observed considering analytical variability coefficients around 5 to 10 %. Analysed properties have an increased variability as follows:  $\text{pH} < \text{COT} < \text{Ca} = \text{K} < \text{Mg} < \text{P}$ . The result of this cartography for P is shown at figure 12. In terms of average properties, the experimental field shows imbalance characterized by high P content and low Mg value. This shows the importance to take into account the spatial variability of P in the current analysis.

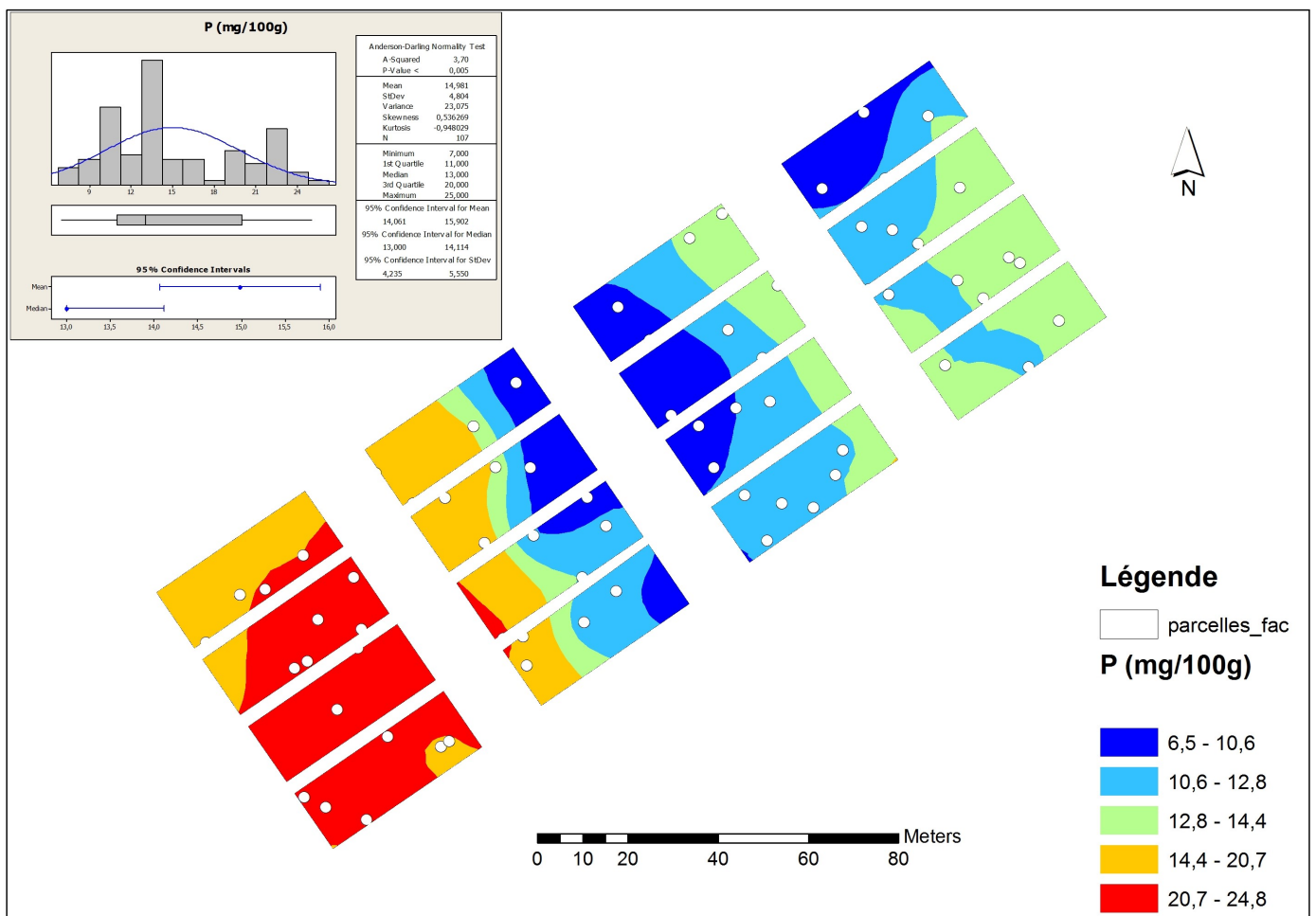


Figure 12: Map of P (mg/100g) within surface layer (0-30cm) for SolResidus experimental field realised by ordinary kriging interpolation on 2012 samples.

The results of the analysis on nutrient mobility of 2013 can be found in ANNEXE C. The results on the Pav have shown a significant effect of the interaction between modality and depth on the layers A and C but not on the B. For ploughed soils, layers A(0-10 cm), B(10-20cm) and C (20-30cm) did not show significant differences. In reduce tillage, P quantities were lightly

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higher on surface than the two other depths. P quantity were higher in plots 1, 2, 3 and 4 independent of the residue management (importation or exportation). The inter-plot variability of the soluble P was important.

Table 6: Soils characteristics for SolResidus and SolCouvert by modalities and depths measured in 2020

Modality*	pH KCl	StdDev	TOC	StdDev	Ca	StdDev	Mg	StdDev	K	StdDev
			g.100g	dry soil <sup>-1</sup>	mg.100g	dry soil <sup>-1</sup>	mg.100g	dry soil <sup>-1</sup>	mg.100g	dry soil <sup>-1</sup>
TCS In	6.94	0.33	1.32	0.26	274	49	7.4	1.5	16.5	7.0
A	7.16	0.34	1.63	0.11	312	58	7.7	1.5	24.5	4.8
B	7.00	0.27	1.28	0.06	271	29	7.5	1.5	15.0	3.0
C	6.65	0.13	1.06	0.04	239	34	7.0	1.8	10.1	1.2
TCS Out	6.93	0.27	1.25	0.25	266	47	7.0	1.2	14.0	6.1
A	7.09	0.30	1.54	0.10	297	54	7.5	1.0	21.1	4.0
B	6.96	0.25	1.20	0.15	264	43	6.7	1.1	11.9	3.5
C	6.73	0.17	1.02	0.10	236	30	6.8	1.7	8.9	1.4
Lab In	6.97	0.23	1.23	0.08	271	25	7.3	0.6	12.6	2.7
A	7.00	0.20	1.21	0.08	272	15	7.1	0.4	11.1	1.5
B	6.92	0.27	1.23	0.11	267	17	7.3	0.8	11.2	1.4
C	7.01	0.29	1.25	0.06	274	41	7.6	0.7	15.5	2.6
Lab Out	6.94	0.20	1.23	0.12	266	39	6.8	1.4	12.1	3.9
A	6.88	0.25	1.15	0.10	265	49	6.6	1.4	9.8	2.9
B	6.97	0.21	1.23	0.12	271	33	6.8	1.5	11.6	3.2
C	6.98	0.18	1.31	0.11	263	44	7.2	1.7	15.0	4.2
DSH	7.07	0.35	1.16	0.19	299	59	8.5	1.4	16.4	5.1
A	7.18	0.57	1.38	0.10	314	83	8.9	1.4	22.3	3.8
B	7.11	0.26	1.14	0.06	287	50	8.3	1.3	13.2	0.8
C	6.91	0.14	0.96	0.05	296	54	8.3	1.8	13.6	3.0
ST	7.18	0.34	1.25	0.28	309	52	8.5	1.7	19.5	6.3
A	7.39	0.23	1.55	0.17	332	57	9.1	1.5	26.9	2.7
B	7.28	0.25	1.21	0.17	303	49	8.8	1.7	17.7	4.1
C	6.88	0.34	1.00	0.11	292	54	7.7	1.9	14.1	2.7
SP	7.20	0.28	1.19	0.07	298	40	8.0	1.4	12.7	3.3
A	7.29	0.33	1.19	0.08	304	39	7.7	1.5	11.3	1.5
B	7.27	0.30	1.19	0.08	322	41	7.9	1.7	13.4	2.1
C	7.05	0.22	1.18	0.07	267	25	8.5	1.3	13.3	5.6
WP	7.15	0.31	1.10	0.05	272	51	7.4	1.4	11.5	2.6
A	7.17	0.40	1.11	0.04	280	54	7.5	1.7	11.1	0.5
B	7.16	0.30	1.10	0.06	285	61	7.5	1.7	13.3	2.0
C	7.13	0.32	1.10	0.05	250	46	7.2	1.0	10.0	3.4

\*Values for each soil management system within each soil parameter are the overall mean of three depths (n = 12).

Values for each Depth in soil management system within each soil parameter are the overall mean of four repetitions (n = 4).

### 4.2 P status in 2020

The analysis was performed on each experiment separately in order to avoid bias from natural spatial variability between each other. A significant difference between tillage modalities has not been shown in neither experiment in Pav content for the first 30 soil's cm (table 7). In contrast, the interaction between depth and modality was significant for both experiments. For SoilResidus, the first layer (0-10cm) showed consistently higher values in the RT modalities compared to the ploughed ones (fig 13 (b)). The highest value of P is observed in the RT treatment when residues were reinstituted (  $13.8 \pm 3.8$  mg.100g dry soil<sup>-1</sup> ). In the second

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(10-20 cm) and third layers (20-30cm), values did not differ significantly between modalities but the lowest was observed in the RT management with exportation of crop residues.

The trend of higher P content on surface for RT managements was also observed in the SolCouvert experiment (fig 13(a)). The highest values are found on the top layer in the strip till management system ( $12.1 \pm 2.8$  mg.100g dry soil<sup>-1</sup>) and in DSH ( $10.2 \pm 0.5$  mg.100g dry soil<sup>-1</sup>). But, in contrast with SolResidus, the lowest value is observed in the third layer of the deep tillage management systems with value of 7.0 and 7.7 mg.100g dry soil<sup>-1</sup> in SP and WP respectively (table 8).

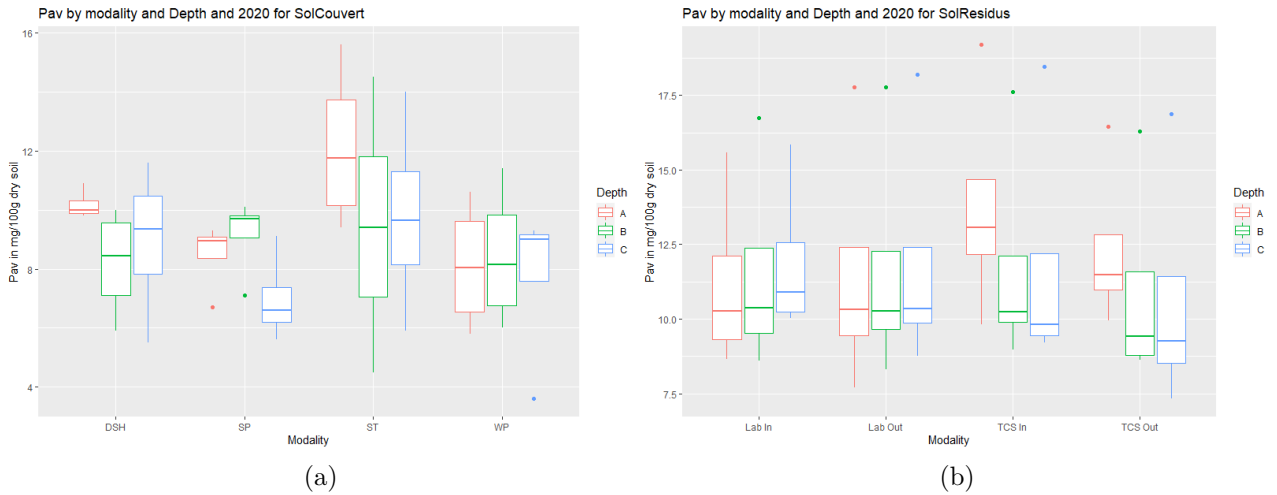


Figure 13: Pav content by modality and depth for (a) SolCouvert and (b) SolResidus measured in 2020. A, 0-10 cm, B, 20-30 cm, C,20-30 cm

Alkaline Phosphatase activity showed consistently lower values than the acid ones apart for the top layer of RT treatments of SolResidus (table 8). The pattern of stratification in RT was also observed in this case (fig 14 and 15). In both experiment, CT showed similar values on the whole horizon whereas RT treatment showed in average higher values in surface and lower in depth. For SolResidus, The values of acid phosphatase ranged from  $150 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  for the deepest depth in RT treatments to  $240 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  for the top layer. The CT treatments gave values close to  $180 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  for both depths (fig 14 (a)). Alkaline phosphatase activities ranged from  $230 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  in the top layer to  $130 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  for 20-30 cm depth in RT treatments whereas CT treatments gave values averaging around  $160 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  (fig 15 (b)).

In SolCouvert, as for the values of Pav, phosphatase activity was consistently lower than in SolResidus. The values of acid phosphatase ranged from  $150 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  for the deepest depth in RT treatments (DSH and ST) to  $200 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  for the top layer. The deep tillage treatments gave value averaging for both layers  $160 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  and  $150 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  for SP and WP respectively (fig 14 (b)). Alkaline phosphatase activities ranged from  $190 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  in the top layer to  $90 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  for 20-30 cm depth in RT treatments whereas CT treatments gave value averaging around  $140 \mu\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  (fig 15 (b)).

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Table 7: Significance of the effects of experimental factors and their interactions on Pav pool, acid and alkaline phosphatase enzyme activity, as resulting from analysis of variance (ANOVA).

Variable	Field	Modality		Depth		Block		Depth*Modality	
		SR	SC	SR	SC	SR	SC	SR	SC
Pav	***	NS	NS	*	*	***	***	**	*
Acid Phosphatase activity	*	NS	NS	***	**	NS	NS	**	*
Alcaline phosphatase activity	**	NS	**	***	***	NS	***	**	***

Signif. codes (p-value): 0 '\*\*\*\*' 0.001 '\*\*\*' 0.01 '\*\*' 0.05 '\*' 0.1 '.' 'NS' 1. SR = SolResidus, SC : SolCouvert

Table 8: Data of Pav, alkaline and acid phosphatase enzyme activity in 2020 for SolCouvert and SolResidus by modality and depth.

Modality*	Pav mg.100g	StdDev dry soil <sup>-1</sup>	Acid phosphatase activity µg p-Nitrophénol .g of dry soil <sup>-1</sup> .h <sup>-1</sup>	StdDev	Alkaline phosphatase activity µg p-Nitrophénol .g of dry soil <sup>-1</sup> .h <sup>-1</sup>	StdDev
Lab In	11.5	2.9	188	18	170	4
A	11.2 b	3.1	188 b	26	173 b	4
B	11.5 b	3.6				
C	11.9 b	2.7	188 b	10	167 b	5
Lab Out	11.7	3.9	181	12	163	24
A	11.5 b	4.3	182 b	12	155 b	36
B	11.7 b	4.2				
C	11.9 b	4.3	181 b	16	167 b	18
TCS In	12.5	3.8	190	45	182	59
A	13.8 a	3.9	230 a	15	231 a	31
B	11.8 b	3.9				
C	11.8 b	4.4	149 c	11	131 b	13
TCS Out	11.3	3.4	189	42	178	61
A	12.3 b	2.8	227 a	14	228 a	30
B	10.9 b	3.6				
C	10.7 b	4.2	152 c	11	129 b	34
ST	10.5	3.4	168	34	147 A	64
A	12.1 A	2.8	193 AB	24	191 A	50
B	9.5 BC	4.3				
C	9.8 BC	3.4	143 C	21	103 D	42
DSH	9.1	1.9	176	41	130 B	44
A	10.2 B	0.5	199 A	13	165 B	23
B	8.2 BC	1.9				
C	9.0 BC	2.6	152 BC	48	94 D	22
SP	8.2	1.6	163	22	144 A	24
A	8.5 BC	1.2	164 ABC	18	145 C	25
B	9.2 BC	1.4				
C	7.0 C	1.5	162 ABC	29	142 C	26
WP	8.1	2.3	151	18	129 B	21
A	8.1 BC	2.2	153 BC	22	131 C	21
B	8.4 BC	2.4				
C	7.7 BC	2.8	149 BC	17	128 C	24

\*Values for each soil management system within each soil parameter are the overall mean of three depths (n = 12).

Values for each Depth in soil management system within each soil parameter are the overall mean of four repetitions (n = 4).

Means followed by the same letter in column represent groups from SNK test for SolResidus experiment, and in capital letter for solCouvert.



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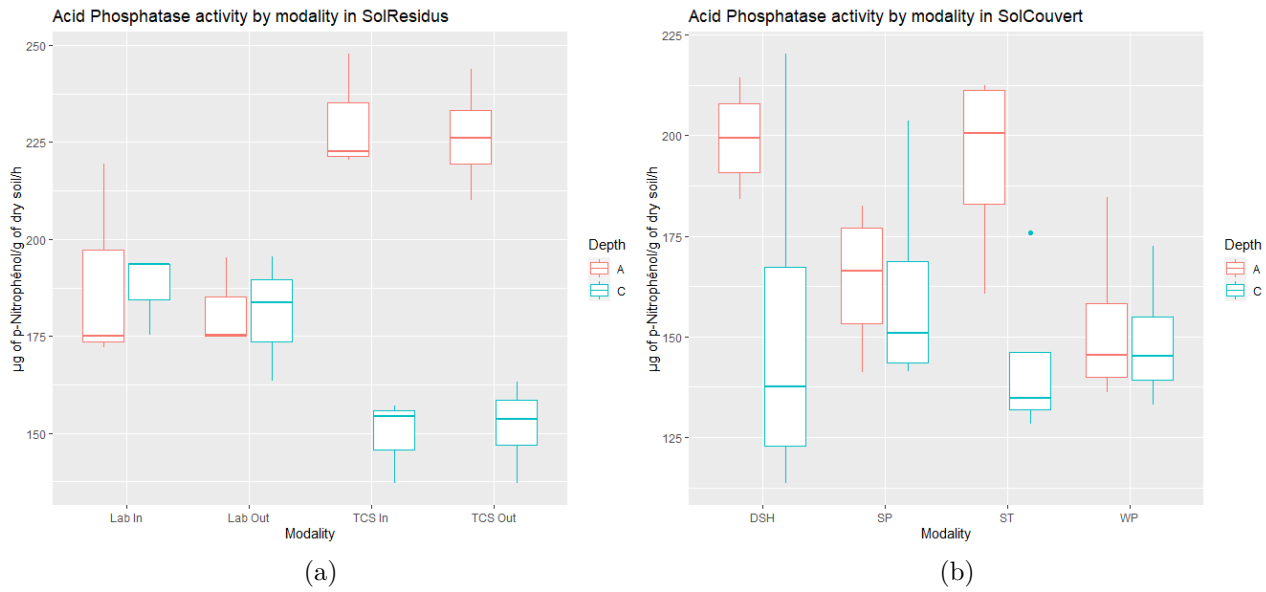


Figure 14: Acid phosphatase activity for (a) SolCouvert and (b) SolResidus measured on 2020. A, 0-10 cm, C, 20-30 cm.

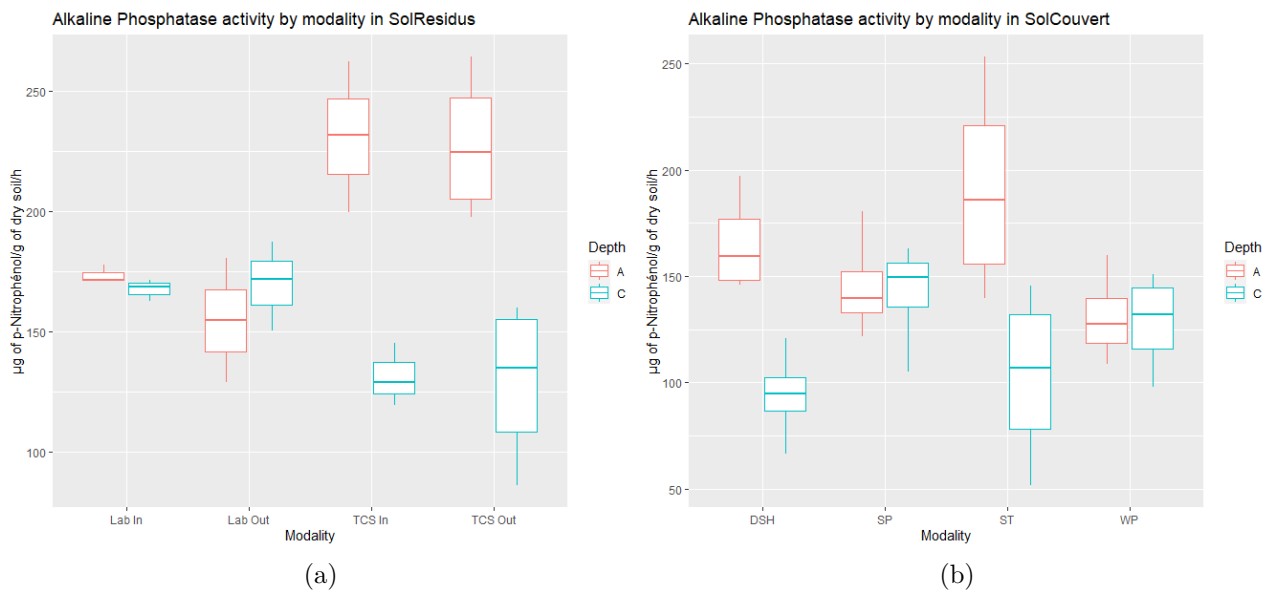


Figure 15: Alkaline phosphatase activity forfor (a) SolCouvert and (b) SolResidus measured on 2020. A, 0-10 cm, C, 20-30 cm.

### 4.3 P evolution

Figure 16 and 17 show the evolution of  $P_{av}$  through time for SolResidus and SolCouvert respectively. In order to analyse the evolution of the  $P_{av}$  pool, the amount of P measured at the start of the experiment and in 2020 were systematically compared for each modality. The year compared was 2013 for SolCouvert and 2011 for SolResidus. The results are shown in table 9. Both experiments are subject to an exhaustion strategy which mean no fertilizer were

#### 4. RESULTS

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applied apart of Nitrate forms. Only sugar lime was sprayed late 2016 that contained some phosphorous which is visible in the following data (fig 16 and 17).

All modalities showed a decrease in Pav content but trends were only significant for CT treatments in both experiments (table 9). The highest drop is found in the Winter Ploughing treatment in SolCouvert, with a diminution of 4.2 mg.100g dry soil<sup>-1</sup>. All RT treatments showed a decrease in Pav content but not significantly. The lowest decrease correspond to the loosening and shallow tillage system (DSH) with an overall drop of only 0.1 mg.100g dry soil<sup>-1</sup>.

A singularity in the data is visible for the year 2014 in the solResidus experiment were a decrease in Pav is observed succeeding a Faba Bean Crop (fig 16). This decrease seems to have been partly compensated in the following measurements. In SolCouvert, the first measurements of 2013 presented high value compared to the following years. An increase, mainly in the top layer seems to have impacted the RT treatments (DSH and ST).

Table 9: Variation in Pav content ( mg.100g dry soil<sup>-1</sup>) by modality since the start of each experiment and 2020 and before and after 2012-2013 and 2014-2015 cover crops for SolResidus with signification

Modality	Global P variation	p-value	2013-2014 P variation	p-value	2014-2015 P variation	p-value
Lab In	-1.00**	0.00155	0.5	0.068	-0.2	0.44
Lab Out	-1.6***	3.88e-5	0.42	0.108	-2.7	0.154
TCS In	-0.6	0.405	0.4	0.0595	- 0.2	0.459
TCS Out	-0.8	0.241	0.3	0.226	-0.3	0.24
WP	-4.2 ***	2e-12				
SP	-2.4 ***	6.14e-5				
ST	-1.5	0.0867				
DSH	-0.1	0.677				

Signif. codes (p-value): 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '?' 0.1 'NS' 1

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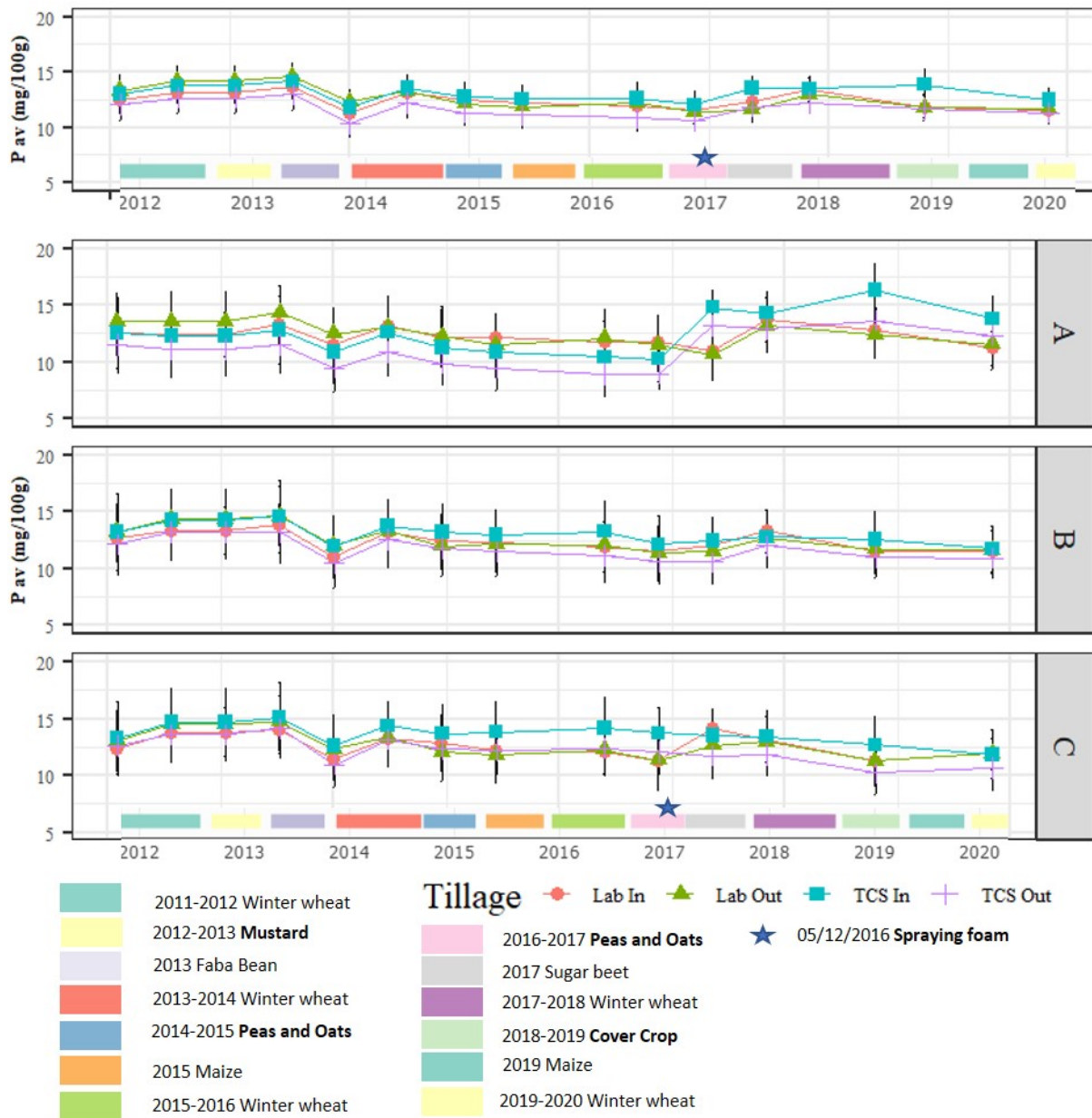


Figure 16: Evolution of the  $P_{av}$  pools by modality and depth for SolResidus experiment. Lab, ploughing, TCS, reduced tillage, In/Out, residues management. A, 0-10 cm, B, 20-30 cm, C, 20-30 cm.

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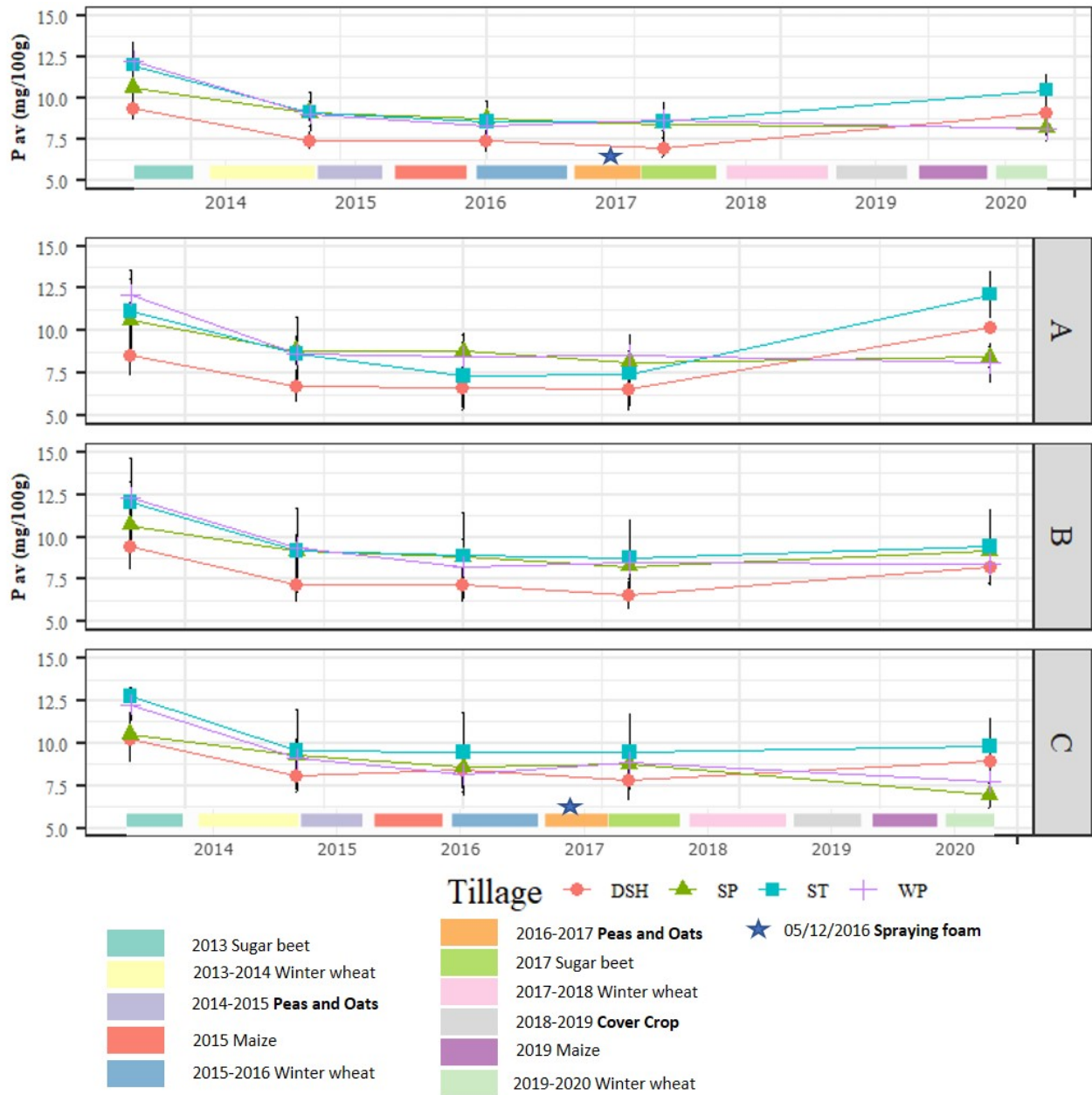


Figure 17: Evolution of the P<sub>av</sub> pools by modality and depth for SolCouvert experiment. DSH, loosening and shallow tillage, WP, winter ploughing, ST, strip till, SP, spring ploughing. A, 0-10 cm, B, 20-30 cm, C, 20-30 cm.

A principal component analysis was performed to study the interaction between soil parameters and phosphorous related characteristics (fig 18). Soils data correspond to the top and bottom layers (A and C) and the tillage modalities (RT and CT). The two principal components allowed to explain 50% and 20% of the variance respectively. Depths for RT treatments were easily differentiated. It seems that RT favoured Pav, COT, phosphatase activity and K for the top layer, while influencing negatively those variables for the bottom layer. For CT treatments, differentiations between groups were less evident.

## 5 Discussion

For both experiments, none of the modalities have shown a significant differences in Pav content on average on 2020 samples in the first 30 cm of soil but nutrients stratification occurred significantly for all RT systems. In addition, the diminution of Pav has been lower for all RT systems since the start of each experiment. The main difference occurred in the top layer (0-10cm) with a enrichment in nutrients, TOC and a higher phosphatase activity.

In the bottom layer, the foreseen impoverishment in Pav for RT treatments has not been observed although the smallest value in Pav for SolResidus corresponds to the 20-30 cm layer of the reduced tillage with residues exportation modality (TCS Out). In contrast, a significant impoverishment was observed in CT treatments in SolCouvert with Spring Ploughing giving the lowest value. An explanation could be the enhancement of mesofauna in RT treatment that can carry nutrients upwards from deeper in the soil profile as [Lemtiri et al., 2018] showed a negative impact of CT on those communities.

Residue treatments had low impact on the Pav in SolResidus. [Hiel et al., 2018] explained it by the low P content of the crop residues that were consistently lower than the theoretical threshold of 2-3 mg/g of P. Below this value, immobilisation by microbial biomass occurs and mineralisation is hampered [Damon et al., 2014]. Nonetheless, an effect seems to have appeared. The curve of the TOC evolution are consistently higher for the residue incorporation modalities (ANNEXE D) and the effect seems to be stronger in the RT treatment where the top layer presented significantly higher Pav content than the others.

Overall, in RT management systems, the distance among groups along the first component showed that the 0-10 cm soil layer has characteristics to the cycle of P that differ from others (fig 18 (b)). This is a similar results that [Tiecher et al., 2012] showed in his study of organic P under RT systems. As noted in his study, the present results show that the importance of biological reactions linked to P cycle increases in the soil surface layer under RT compared to CT.

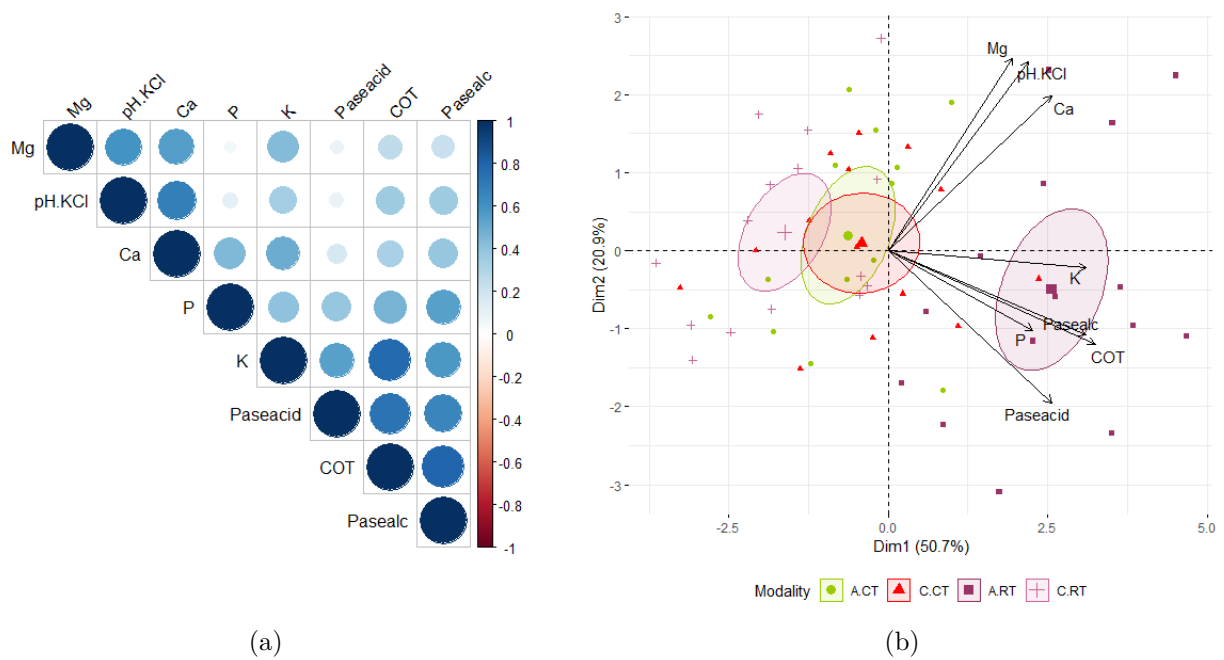


Figure 18: a: Correlation matrices between soil variable; b: PCA analysis, confidence ellipse level in normal probability = 0.95, RT = Reduce Tillage, CT = Conventional tillage, A = 0-10 cm depth soil layer and C = 20-30cm depth soil layer.

In this regard, soil phosphatase strongly controls the biotic pathway of soil P cycle but, in this case, showed relatively small correlation between Pav and Pase acid (0,35) and Pase alkaline (0,50) (fig 18(a)). This goes along with [Margalef et al., 2017] which showed little correlation between Pav (from Bray and Mehlich method) and phosphatase activity. In contrast Porg seems to be a better indicator for phosphatase activity [Margalef et al., 2017] but was not measured here. This indicates that the mechanisms influencing the availability of P are not all measured by the [Lakanen and Erviö, 1971] method. Pav is the result from the balance between plant and microbial sink, and the source from inorganic compounds and organic matter, controlled partly by phosphatase activity. Thus, Pav can be seen as an instantaneous picture of the immediately available P whereas phosphatase relates only the source terms [Margalef et al., 2017].

Values of phosphatase found in both experimental fields were in the same range as the ones reported by [Roldán et al., 2005] and [Eivazi and Tabatabai, 1977] but consistently lower than [Tiecher et al., 2012] observed in highly weathered tropical soil. This can partly be explained by the methodology used. The first used dried samples as here while Tiecher used fresh ones. In general, alkaline phosphatase activity is often more important in alkaline soils which was not observed here. In this regards [Eivazi and Tabatabai, 1977] showed that, air dried samples decrease alkaline phosphatase activity. Still, patterns differed according to modality with a higher content in phosphatase in the soil surface layer in RT compared to CT.

Phosphatase is mainly produced by fungi and plants [Margalef et al., 2017, Dakora and Phillips, 2002]. In addition, organic matter can act as a protection for soil enzymes by forming complexes with humic compounds keeping them in active forms [Quiquampoix and Mousin, 2005]. The interaction between TOC and phosphatase can explain the high correlation among those variables (fig 18(a)). Thus, the higher phosphatase activity in the top layer under RT

compared to CT and the increased activity in soil layer 0.10-0.30 m under CT can be attributed to two mechanisms : (1) the lower soil disturbance in the RT that favour edaphic biota development [Tiecher et al., 2012] and (2) the priming effect in CT [Faucon et al., 2015].

Cover crops can enhanced phosphatase activity [Hallama et al., 2019] but this parameter could not be verified in this case due to the lack of comparative measurements. In order to validate or invalidate this hypothesis, further analysis are required. A way to go might be the use of a comparative experimental field under similar tillage systems without the implementation of cover crop. Nonetheless, an increase (not significant) of the Pav content was observed after a cover crop composed of mustard between 2012 and 2013 in SolResidus (table 9). In contrast, a decrease, not significant either, was observed after a peas and oats cover crop in winter 2014-2015.

It is hard to assert a real impact of those cover crops on the Pav content for two main reasons : (1) the sampling date did not coincided with the sowing day of the following crop, that might have already influenced the P content and (2) analytical measurement variability. In spite of that, leaving crops residues on the field seems to have had a positive effect by reducing the decrease and improving the increase of Pav in the soil compared to the residues exportation for both tillage systems.

the drop in Pav content after the faba bean crop, independent of modalities, could not be attributed to analytical error. Faba bean (*Vicia faba* L.) is a carboxylate-exuding legume known to be a plant that enhances phosphorus solubilisation in the soil and facilitates uptake by the following crops [Rose et al., 2010, Zhang et al., 2016, Zhou et al., 2009]. The mechanisms by which faba bean enhance phosphorus efficiency are of two types : (1)P uptake enhancement by roots traits, i.e. modified root architecture, mycorrhizal symbiosis and formation of root hair [Richardson et al., 2011] and (2) increased P mobilisation from non-labile pools by roots exudation of citrate, known for its high P solubilisation capacity [Shen et al., 2011], malate and various compounds. [Rose et al., 2010], after an analysis on 50 different genotypes on acidic and alkaline soils, showed that P uptake efficiency was strongly correlated with root traits in acidic soils but not in alkaline, where faba bean showed a higher malate exudation.

They also observed a decrease of the Pi pools extracted by NaOH and NaHCO<sub>3</sub> after growing faba bean that didn't impact P uptake and yield of the following winter wheat crop in the alkiline soil and even enhanced it for the acidic one. The same trend was observed here [Hiel et al., 2018]. Therefore, the drop in Pav content following the faba bean crop of 2013 with little impact on the following winter wheat could be explained by the following scenario : (1), the citrate and malate exuded solubilized P from non available Pi pools, (2) P utpake by the faba bean crop combined with microbial community enhancement that immobilized P in organic forms that are not measured by the [Lakanen and Erviö, 1971] resulted in a drop of Pav and (3), during winter wheat growing season, an enhanced phosphatase activity from microbial community releasing P from Porg pools assuring crop needs.

This hypothesis of changes in soil microbial community occuring in faba bean crop resluting in similar or higher P uptake by the following crop was first made by [Nuruzzaman et al., 2005] and followed by [Rose et al., 2010]. the conclusion of [Aschi et al., 2017] goes in the

same direction by showing that including faba bean in a crop rotation promotes soil microbial communities by providing available carbon and nitrogen as well as suitable soil pH.

If the faba bean crop impacted similarly all modalities, the fertilization event of late 2016 resulted in more distinctive impacts. Sugar lime is constituted partly of P, mainly in organic forms<sup>5</sup>, and thus can be considered a P fertilizer. P fertilization efficiency is often very low for the first year, but when taken in a long period, almost all P input are returned to crop [Johnston et al., 2014]. In this case the fertilization event resulted in a global increase of P<sub>av</sub> for all modalities but with a higher impact in the RT systems (figure 16 and 17) on the surface layer. This, linked with the higher phosphatase activity of top layer in RT, could indicate an improvement of fertilization efficiency when fertilizer composed of P<sub>org</sub> are used (i.e. manures, limes, mud from STEP).

Although RT treatments seem to have scored higher in the phosphorous point of view, [Hiel et al., 2018] showed in her study an overall decrease of 3.5% in yield for RT treatment in Sol-Residus. The main reason was the decrease in germination rates attributed to higher pathogens in the top layer in RT treatment. This result goes in the same direction as [Pittelkow et al., 2014]. His study on the impact of conservative agriculture showed that no till systems potential contribution in intensive farming systems is often limited and often impact negatively yields. But when combined with others conservation agriculture principles such as residue management and cover crops, those negative effects can be minimized and even give greater results than conventional systems [Pittelkow et al., 2014]. The best results were observed in drier climate. In addition, it has been shown that RT systems can improve risks of P loss [Rodrigues et al., 2016, Cambouris et al., 2017]. Even if erosion is often reduce in RT, accumulation of reactive P on the top soil can result in higher environmental P loss.

This points out the fact that decision-making at the farming level is multi-factorial dependent and needs to take numerous implications into account. Nonetheless, the current data tend to show that RT favour biotic pathways to assimilate phosphorus on surface, where most of the fertilizer are applied, compared to CT. This combined with the findings of [Bruwier, 2019] that showed an important sollicitation of the P<sub>org</sub> pool by crops in an exhaustion scenario in similar pedo-climatic conditions, indicates that conservation agriculture practices could represent promising leads for a reduction of mineral P fertilizer consumption in cultivated Cutanic Luvisol of Belgium.

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<sup>5</sup>[Thomas, 2012] reported a content of 9.1 kg/t of organic P<sub>2</sub>O<sub>5</sub>



## 6 Conclusion

The faculty of Gembloux Agro-Biotech started a decade ago the SolResidus and SolCouvert experiments in order to test the long term impacts of conservative agricultural practices and compare them with the conventional ones in an intensive production system. Among agricultural issues phosphorus management is a key feature in order to achieve sustainability in the current food production systems in Belgium. In this sense, those long term agronomic experiments offered an incredible opportunity to evaluate impacts of such practices on the P cycle.

In this case, the practices tested focus on reduced tillage, cover crop implementation and residue management. Soils parameters collected on the two experimental fields since their beginning were analysed in order to distinguish the impacts of those practices on the P cycle. In addition, acid and alkaline phosphatase activities were measured on the 2020 samples in order to investigate the biological pathways of P mobilisation that appeared to be greatly solicited in those types of soils. The current study followed the paper of [Hiel et al., 2018] which studied the dynamic of nutrients for the SolResidus experiment in an integrative approach including plants and yield data up until 2016.

We hypothesised that under RT systems, a stratification of nutrients in surface layers of the soil, including available P, will occur in the reduced tillage modalities. Even with an important natural phosphorus spatial variability for both experiments, the analysis of the data showed that it was the case with, on average, a higher  $P_{av}$  content in the top layer (0-10 cm) of every RT modalities in the samples collected in 2020 compared to CT. In contrasts, the 20-30 cm layer impoverishment in  $P_{av}$  content was not homogeneous between CT and RT modalities indicating a potential enhancement of mesofauna activity under the RT systems.

The phosphatase activity data suggest that the biological pathways is enhanced in the top soil layer under RT treatments compared to CT. This could explained the higher P mobilisation on this layer of the Porg contained in the sugar lime sprayed in late 2016 in the top layer of RT treatments compared to CT.

Residue management appeared to have less impact on the availability of P than tillage modalities. Nonetheless the data suggest a beneficial trend on the  $P_{av}$  pools in the modality where the residues were left on the field. The cover crop implication could not be investigated in depth due to the lack of comparative measurements, but the literature suggests that its implementation in crop rotation can have beneficial influences, mainly by improving the biological pathways of P mobilisation.

This study had a pedological specific approach which mainly focused in the nutrient dynamics on the soil dimension. Its results suggest that conservative agricultural practices offer promising leads to the reduction of mineral P fertilizer consumption. However, when guidelines for farmer are to be written, other dimensions such as environmental impacts, yield and human and material costs need to be taken into account.

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## A Results from measured proprieties on sounding samples

Table 10: Synthesis of measured proprieties on sounding samples: Minimum/Maximum and relative amplitude and Variation coefficient (CV) / (N=16)

Horiz.	pHKCl	COT g.kg-1	A %	L	S	P	K	Mg mg.100g-1	Ca
(Ap) (0-30)	6,3 / 7,0	10,4 / 13,8	13,8 / 18,9	75,3 / 80,8	4,7 / 7,2	8,2 / 22,8	13,3 / 21,0	4,6 / 10,5	206 / 321
% *	10	28	32	7	44	108	47	72	45
CV	3	8	11	2	12	32	13	28	15
BT (40-60)	6,2 / 6,7	3,1 / 8,8	15,2 / 27,9	68,0 / 79,8	3,5 / 5,8	0,5 / 5,9	9,4 / 17,1	5,3 / 13,7	176 / 298
% *	8	106	59	16	40	210	58	97	52
CV	2	28	17	5	13	58	19	28	18
C (110-120)	6,2 / 6,6	1,3 / 5,6	17,7 / 25,6	71,0 / 78,0	2,5 / 5,3	0,5 / 1,1	7,2 / 12,1	4,7 / 11,7	233 / 291
% *	8	286	34	10	73	162	50	114	22
CV	2	77	8	2	24	41	14	27	6

\*Amplitude rapportée à la moyenne

## B Results of nutrient's spatial variability characterisation

Table 11: Statistical synthesis of fertility indicators of SolRésidus experimental field.

(N=107)	pHKCl	COT	P	K	Ca	Mg
		g.kg-1	mg.100g-1			
Moyenne	6,79	12,7	14,9	16,2	256	8,3
Ecart-type	0,19	1,2	4,9	2,7	37	1,7
Coef. Var.	2,8%	9,4%	32,9%	16,7%	14,4%	20,5%
Min.	6,40	9,4	6,5	10,5	205	4,6
Max	7,30	16,0	24,8	22,2	369	11,8
Range	0,90	6,5	18,3	11,7	164	7,2
P25	6,70	12,2	11,3	14,2	227	6,9
P50	6,80	12,8	13,2	16,4	243	8,6
P75	6,90	13,4	19,8	18,4	286	9,7
EIQ	0,20	1,3	8,5	4,2	59	2,8
dissymétrie	-0,10	-0,12	0,54	-0,21	0,87	-0,34
Aplatissement	-0,26	0,45	-0,93	-0,71	-0,04	-0,66
geostatistical parameters						
Model*	— Sphérique —					
Scope	75-120m	65-170m	65-110m	65-195m	120-194m	85-135m
C / (C <sub>0</sub> + C)	65%	38%	97%	64%	92%	87%

\*spheric model correspond to a law of type  $\gamma(h) = C_0 + C ((3h/2\alpha) - 1/2(h/\alpha)^3)$

where  $\gamma(h)$  = semi-variance for pairs of points separated of a distance h

C<sub>0</sub> = semi-variance for h=0 ; C la semi-variance spatially dependant ;  $\alpha$  = the scope.

The ratio C/(C<sub>0</sub>+C) correspond to the spatially structured semi-variance proportion



Table 12: Results of comparison tests on means from plots of SolR siduals experimental field

Placette*	pHKCl	COT	P	K	Ca	Mg
1	ab	c	a	a	a	bc
2	b	ab	a	a	a	ab
3	b	ab	a	b	a	b
4	ab	a	a	bc	a	b
5	d	f	bc	b	b	c
6	d	ef	bc	c	bc	c
7	bc	bc	b	d	c	c
8	bc	b	b	de	c	d
9	c	e	bc	c	bc	ab
10	cd	e	bc	d	c	b
11	c	d	bc	f	cd	c
12	bc	bc	bc	f	c	d
13	bc	d	c	d	c	a
14	ab	bc	bc	ef	c	b
15	a	bc	bc	g	bc	b
16	a	c	b	e	c	ab

\*Plots with different letters for a the same variable are considered statistically different

## C Results from nutrient mobility monitoring

Table 13: Effect of management\*depth on soil characteristics according to studied modality. Signification of measured elements by depth.

Elements	Observed effect / Signification		
	Depth A (0 – 10 cm)	Depth B (10 – 20 cm)	Depth C (20 – 30 cm)
Conductivity	ns	ns	***
Humidity	***	***	***
Phosphorus	***	ns	***
Potassium	**	***	ns
Magnesium	ns	*	***
Calcium	*	*	**

\*\*\* p < 0,001; \*\* p < 0,01; \* p < 0,05; ns = non significant

## D TOC evolution

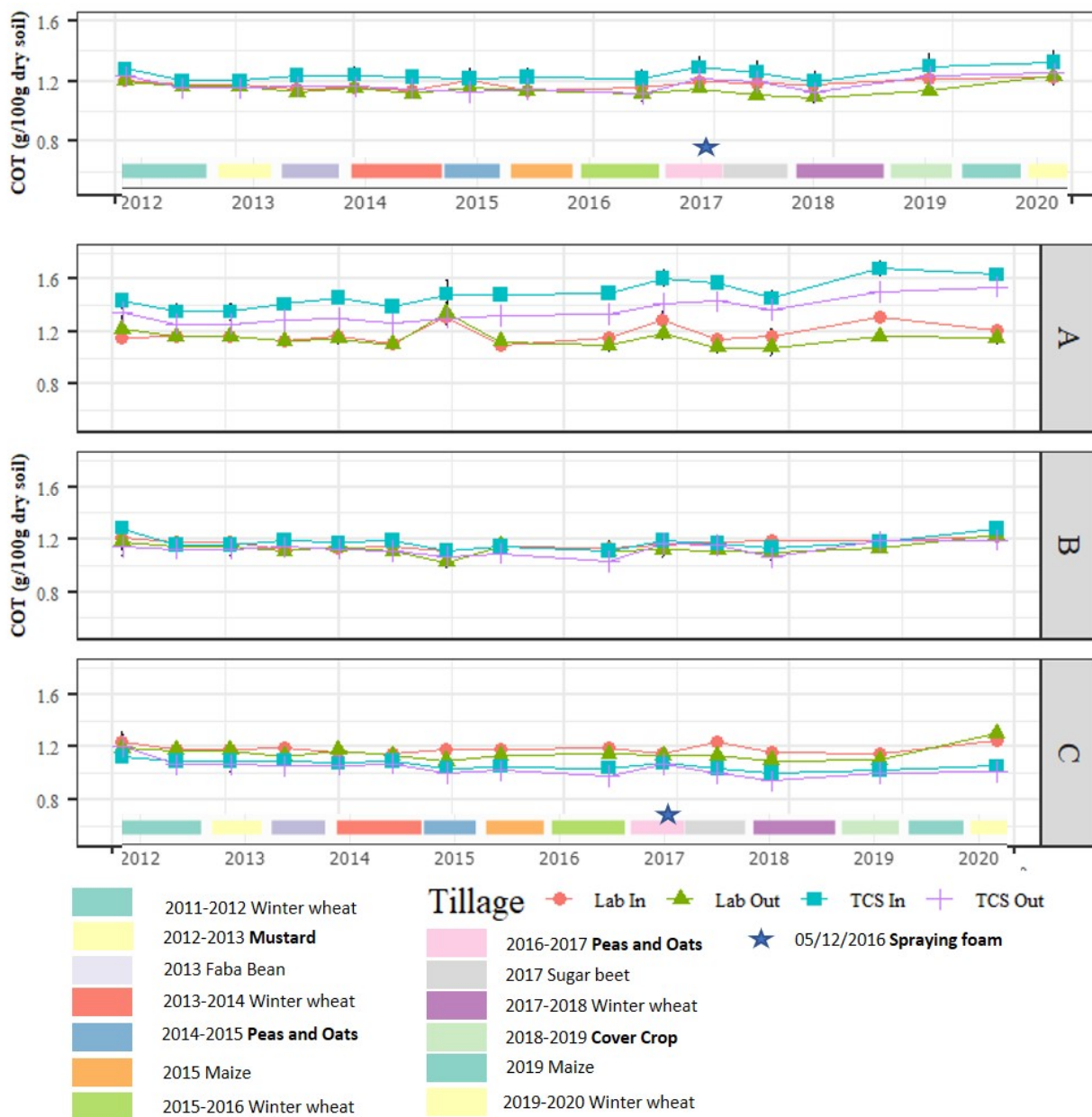


Figure 19: Evolution of the P av pools by modality and depth for SolResidus experiment

D. TOC EVOLUTION

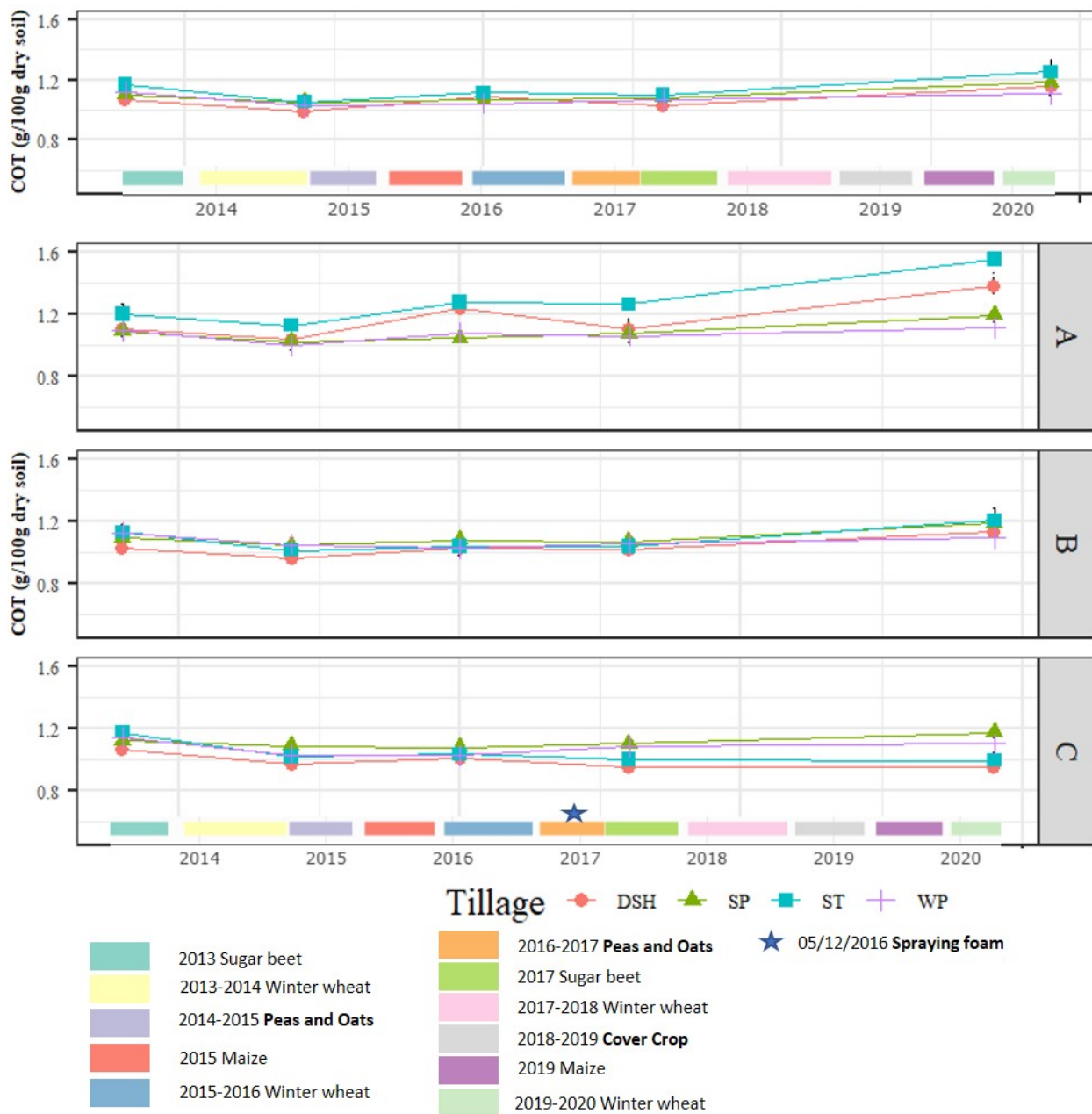


Figure 20: Evolution of the P av pools by modality and depth for SolCouvert experiment

## E P evolution by modality and plots

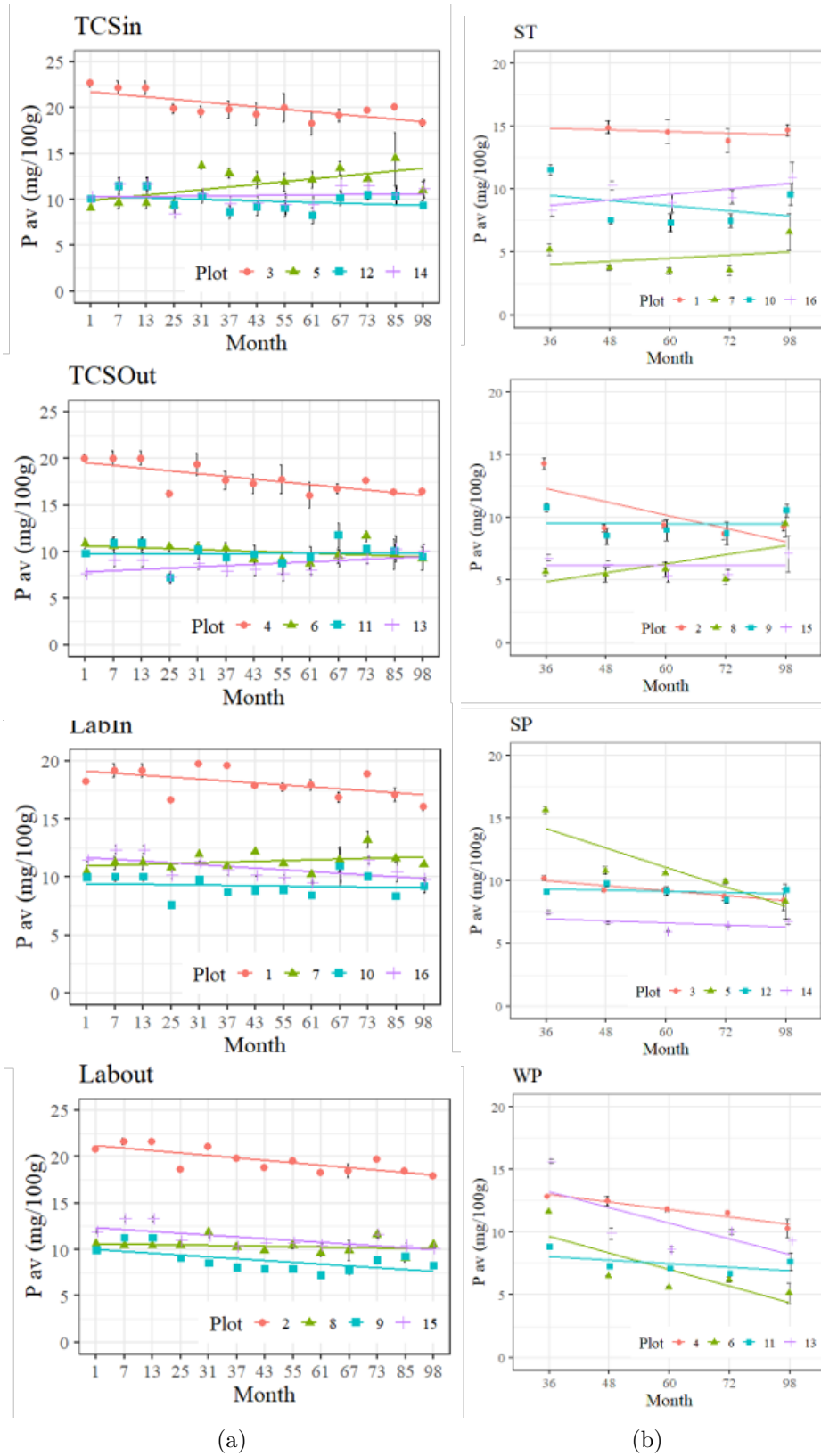


Figure 21: Pav content by Modality and plot for (a) SolRésidus and (b) SolCouvert