FRANCISCO J. MATUS

CROP RESIDUE DECOMPOSITION, RESIDUAL SOIL ORGANIC MATTER AND NITROGEN MINERALIZATION IN ARABLE SOILS WITH CONTRASTING TEXTURES

Promotor:	Dr. L. Brussaard Hoogleraar in de Bodembiologie
Co-promotor:	Dr. A.P. Whitmore Wetenschappelijk onderzoeker bij het DLO-Instituut voor Agrobiologisch en Bodemvruchtbaarheidsonderzoek (AB-DLO)

FRANCISCO J. MATUS

CROP RESIDUE DECOMPOSITION, RESIDUAL SOIL ORGANIC MATTER AND NITROGEN MINERALIZATION IN ARABLE SOILS WITH CONTRASTING TEXTURES

Proefschrift
ter verkrijging van de graad van
doctor in de landbouw- en milieuwetenschappen,
op gezag van de rector magnificus,
dr. C.M. Karssen,
in het openbaar te verdedigen
op dinsdag 29 november 1994
des namiddags te vier uur in de Aula
van de Landbouwuniversiteit te Wageningen

To Daisy

STELLINGEN

1 Once an equilibrium of soil organic matter is achieved in soil, the variation in nitrogen mineralization depends on the inputs of organic matter from individual crops in a crop rotation rather than on the soil characteristics.

* This thesis

2 In contrast to what is generally accepted added labelled plant materials (of low C:N ratio) in cropped and uncropped soils decompose at similar rates.

* This thesis

3 The mechanisms of physical protection of soil organic matter in soils of different textures differ.

* This thesis

4 Similar rates of decomposition of added labelled plant materials in soils of very different textures are possible if initially decomposition is the dominant process rather than physical protection.

* This thesis

5 Chemical composition of plant material rather than soil texture and soil structure is the determining factor in the decomposition of labelled plant residues immediately after incorporation.

* This thesis

6 Regardless of the energy applied to disperse soils, the soil organic matter concentration in silt and clay size fractions is higher in coarse- than in fine-textured soil.

* This thesis

7 Science relies on genius, instrument and method...

* Dr. Carlos Rivera, Lecture of Philosophy of Science and Research Methods, Pontificia Universidad Católica de Chile.

8 ... but it is not irrational to look first at work already done, and to rearrange one's own thoughts rather than experimentally to rearrange nature.

* H.T. Pledge.

9 The neo-liberal concept of a free market in Chile is far from the rationality of sustainable agriculture.

10 Ivan Illich has calculated that the car, taking into account how many hours the owner has to work extra in order to be able to buy and use it, does not go faster than the bike.

* Willem Hoogendijk, The Economic Revolution

11 Our modern society is one in which you should remain consuming goods, otherwise the happiness seems in danger.

12 Democracy without democrats is the desire of the Chilean right-wing party (UDI).

Stellingen behorend bij het proefschrift "Crop residue decomposition, residual soil organic matter and nitrogen mineralization in arable soils with contrasting textures".
F. J. Matus, Wageningen, 29 November 1994.

PUBLICATIONS

The chapters 2 through 7 were or will be published as separate papers, with some modifications.

Chapter 2

Matus, F.J. and J. Rodríguez (1994) A simple model for estimating the contribution of nitrogen mineralization to the nitrogen supply of crops from a stabilized pool of soil organic matter and recent organic input. *Plant and Soil* **162**, 259-271.

Chapter 3

Matus, F.J. (1995) Effect of soil texture, soil structure and cropping on the decomposition of crop residues. I. Residual organic C and microbial biomass C derived from decomposing ^{14}C-labelled wheat straw in a clay and sand soil. *Soil Biology & Biochemistry* (submitted).

Chapter 4

Matus, F.J. (1995) Effects of soil texture, soil structure and cropping on the decomposition of crop residues. II. Residual organic N, microbial biomass N and inorganic N derived from decomposing ^{15}N-labelled wheat and clover in a clay and sand soil. *Soil Biology & Biochemistry* (submitted).

Chapter 5

Matus, F.J. (1995) The distribution of soil organic matter of various aggregate size classes in arable soil. I. Relationships between clay content of aggregates of a sand and a clay soil and carbon mineralization, nitrogen mineralization and microbial biomass carbon. *Soil Biology & Biochemistry* (submitted).

Chapter 6

Matus, F.J. (1995) The distribution of soil organic matter of various aggregate size classes in arable soils. II. Residual organic ^{14}C, residual ^{15}N, microbial biomass ^{14}C and ^{14}C and ^{15}N mineralization rates in a sand and a clay soil. *Soil Biology & Biochemistry* (submitted).

Chapter 7

Whitmore, A.P. and Matus, F.J. (1995) The decomposition of wheat and clover residues in soil: measurements and modelling. *Plant and Soil* (submitted).

CONTENTS

ABSTRACT

To evaluate the significance of cropping, soil texture and soil structure for the decomposition of ^{14}C- and ^{15}N-labelled crop residues, a study was conducted in a sand and a clay soil under laboratory and field conditions. The distribution of residual ^{14}C, residual ^{15}N and microbial biomass ^{14}C of different aggregate size classes and physical protection of soil organic matter as indicated by the rates of ^{14}C and ^{15}N mineralization after soil disaggregation were also studied in the same soils. Soil texture and soil structure were not determining factors in the decomposition of residual labelled soil organic matter, but residue type was important for N mineralization soon after incorporation. Recently formed labelled soil organic matter was less well physically protected than older soil organic matter and adsorption of soil organic matter on to silt and clay particles was the main mechanism of physical protection in sand soil. In clay soil the results were not conclusive as regards the main mechanism of protection of recently formed soil organic matter. In conclusion because soil structure broken up by soil disruption, and soil texture were found to have no effect on the rate of decomposition of recently formed organic matter in soil, we do not need to include these factors when the C and N mineralization from crop residues in arable soils has to be estimated.

ACKNOWLEDGEMENTS

It is difficult to express in a few lines my indebtedness to the many people who have contributed to the results presented here. As important to me as my scientific education was the experience of everyday life in The Netherlands. Both will be valuable assets for the rest of my life.

Completion of my thesis would not have been possible without the guidance, help and support of my promotor Prof. Lijbert Brussaard and co-promotor Dr. Andy Whitmore. I enjoyed the discussions, even into the evening. I am deeply indebted to them.

This study was conducted under the financial support of the Chilean Ministry of Planification and Cooperation (MIDEPLAN) through of the Agencia de Cooperación Internacional (AGCI) and the sandwich PhD Fellowship Programme of Wageningen Agricultural University. I thank Ir. C.W.M. de Ranitz and Mr. C.M.M. Van Heijst from Wageningen Agricultural University for the approval of my grant.

I am grateful to Talca University for supporting my application to the Chilean Ministry and for giving me permission to stay in The Netherlands while keeping available my former position at the Agronomy School, and for supporting me financially. I especially thank Dr. Alvaro Rojas Marín, Rector of Talca University for keeping me informed about the latest developments at the University.

I am indebted to the Research Institute for Agrobiology and Soil Fertility (AB-DLO) where all the research was carried out. Ricardo Catalán, a Chilean colleague, made the first contact with the Institute possible, through Dr. J.J. Neeteson, Head of the Department of Soil Fertility and Dr. J.A. Van Veen, former Head of the Department of Soil Biology. I offer them my deep gratitude. I also thank to the former Director of the Institute Dr. K. Harmsen who kindly welcomed me as a guest worker.

The execution and sampling of my field experiments was made possible by F.H. de Vries, P. Datema, W. E. Willems, H. Schepers, O.A. Slim, J. Greving, E.L. Schreuder, H. Matthijs, H. Van Rein and others. Thank you for your help and friendship. I thank M. Bralts and J. Postmus for organizing the layout of my glasshouse experiments and D. Kerstens for labelling the enormous number of samples and for his patience when I arrived with yet more. I thank the Central Laboratory and in particular Mr. T. Lepelaar and Mr. J. Dijkstra for the ^{15}N analyses.

The isotopic experiment with ^{14}C would have not been possible without the help and tuition of J.W. Dalenberg and the technical assistance of M. Wolters. Thank for your dedication and friendship. I am indebted to J.H. Van Ginkel who contributed the ^{14}C-labelled material. The technical assistance of G. Brouwer, K. Kampen, A. Lahr-Renkema, A. Vos, P.R. Bolhuis and J.S. Zwiers is gratefully acknowledged. I also thank Ir. J. Marinissen who advised me to obtain the soil aggregates.

I am indebted to Dr. P. de Willigen, Ir. J. Hassink and Dr. K.B. Zwart for having made room

in their busy programmes to give priority to reading this thesis. I also thank Dr. K.B. Zwart for his translation of the English summary into Dutch. I thank Dr. L.A. Bouwman for his comments and suggestions on chapter 5, Dr. J.J. Neeteson and Ir. W.P. Wadman for their comments on chapter 2, and Dr. J. Bloem and Ir. J.A. de Vos for his comments on chapter 6. I would like to extend my special appreciation to J. Hassink for his friendship, guidance and support during my stay in The Netherlands.

Several persons helped me during the preparation of my project: Dr. M. Van Noordwijk, Dr. J.J. Neeteson, Ir. E.L.J. Verberne, Ir. J. Hassink and Dr. A.P Whitmore. Thanks you for your contributions.

I thank Ir. P.A.I. Ehlert, J.W. Steenhuizen and S. Wijnia for their kind welcome when I arrived in The Netherlands. I also thank to the Administrative Department of the Institute: J. Pentinga, E.J.M. Van Wijk, W. Boersma, H. Dieperink, H. Sommer and several others for their cooperation, whenever needed.

I especially thank H. Terburg for the English corrections on the manuscript and for his help in the layout of this thesis. I am also indebted to J. Robot and M. Bos-Bouwsema for their help with the Figures. I thank the secretaries: Ms. T. Offringa, A. Gorter, C.A. Bahlmann and A. Mansholt, as well as J. Nijborg and H. Visscher for their help.

I extend grateful thank to my parents, parents-in-law and other members of our families for their concern, support and encouragement during this time.

I express my deepest gratitude to my wife, Daisy, for her support, patience and cheerfulness during the completion of this project.

Francisco J. Matus
Haren, Autumn 1994

CHAPTER 1

GENERAL INTRODUCTION

Soil organic matter

All soils with a history of plant cover contain soil organic matter, derived directly from plant and animal debris or synthesized during biological decomposition. An important step in the comprehension of soil organic matter dynamics was to recognize that only a small part of the total organic carbon and nitrogen in soil is biologically active. Jansson (1958) brought together many of the concepts involved in mineralization-immobilization of nitrogen and confirmed the earlier suggestion of Gainey (1936) that the soil organic matter is composed of an active and a passive phase. The current understanding of soil organic matter has been summarized in comprehensive simulation models of the dynamics of organic carbon and nitrogen in soil (Jenkinson and Rayner, 1977; Paul and Juma, 1981; Van Veen *et al.*, 1985; Parton *et al.*, 1987; Whitmore and Parry, 1988 ; Verberne *et al.*, 1990). All these models contain the concept of active and passive pools of soil organic matter with carbon and nitrogen cycling mainly restricted to the active pool.

Decomposition of plant residues and soil organic matter and mineralization of nitrogen in soil

Decomposition of the active soil organic matter fraction includes physical breakdown of plant residues by animals and abiotic processes. Soil organisms are the living mass of soil organic matter consisting of bacteria, fungi, protozoa, nematodes and other invertebrates which may not all be present within any one ecosystem. A number of factors can modify the course of the decomposition of plant residues in soils. It has long been recognized that residues from young plants decompose more rapidly than those from older plants (Waksman and Tenney, 1928). Differences in the release rate of carbon and nitrogen are often ascribed to the amount of carbon and nitrogen and their ratio in plant residues. However, Herman *et al.* (1977) indicated that apart from the carbon to nitrogen ratio, the lignin to nitrogen and the carbohydrate to nitrogen ratio were good indicators of differences in crop residue decomposition in soil. Chemical compounds in plants such as structural carbohydrates (cellulose, hemicellulose, lignin) and polyphenols (Tian *et al.*, 1992[a]; Tian *et al.*, 1992[b]) as well as the nature of the decomposer community (Tian *et al.*, 1994) show a good correlation with the decomposition rate of plant materials in soil.

The presence of living plants has also been suggested to influence the decomposition of plant residues in soil. Cropping induced less decomposition of residual ^{14}C compared with fallow soils in the field (Führ and Sauerbeck, 1968; Shields and Paul, 1973; Jenkinson, 1977) which was

attributed to less water being available in the cropped soils (Shields and Paul, 1973; Jenkinson, 1977) or restricted soil aeration (Führ and Sauerbeck, 1968). However, during laboratory experiments under controlled humidity and temperature conditions both positive and negative effects have been described (Sallih and Bottner, 1988), while in experiments where the aeration was controlled a reduction in the decomposition was observed (Reid and Goss, 1982). Living roots also led to negative (Huntjens, 1971) or positive (Hart *et al.*, 1979; Jingguo and Bakken, 1989) effects on net nitrogen mineralization. Roots may induce more microbial activity in the rhizosphere by exudation of organic compounds (Helal and Sauerbeck, 1986). It seems that the presence of plants, rather than stimulating nitrogen mineralization, reduces the microbial immobilization of N through effective competition with microbes for mineral nitrogen (Jingguo and Bakken, 1989). It has been suggested that with high nitrogen concentrations in soil, microbes prefer root-derived materials over native soil organic matter and that at low nitrogen concentrations this preference does not occur (Liljeroth *et al.*, 1990; Kuikman *et al.*, 1990).

Soil organic matter protection

Destruction of soil structure, i.e., disruption of soil aggregates, is another factor influencing the decomposition of soil organic matter (Craswell and Waring, 1972; Sørensen, 1983[a]; Nordmeyer and Richter, 1985; Cabrera and Kissel, 1988; Van Gestel *et al.*, 1991; Hassink, 1992). The soil organic matter content of most soils declines when virgin land is cultivated (Van Veen and Paul, 1981; Van der Linden *et al.*, 1987). The loss is most rapid during the first few years, thereafter it diminishes considerably (Van Veen and Paul, 1981). The decline in soil organic matter content after cultivation cannot be attributed entirely to a limited return of the amount of plant residues incorporated in soil. A greater decrease of soil organic matter is also possible as a result of disruption of soil aggregates (Elliott, 1986). Physical protection from microbial degradation arises when soil organic matter is entrapped in small pores within soil aggregates (Elliott and Coleman, 1988) or is physico-chemically adsorbed onto the surface of clay minerals (Edwards and Bremner, 1967; Tisdall and Oades, 1982) or because soil organic matter is encrusted by clay particles (Tisdall and Oades, 1982). The destruction of soil aggregates exposes previously inaccessible organic matter to microbial attack (Rovira and Greacen, 1957; Craswell and Waring, 1972; Hassink, 1992). The increase in carbon and nitrogen mineralization released from the soil after soil structure is disrupted constitutes indirect evidence that soil organic matter is physically protected from biodegradation. Clay soils may protect relatively more soil organic matter than sand soils in small pores (Hassink *et al.*, 1993). Microorganisms are more numerous in inner pores of aggregates than on the external surfaces and in large pores (Vargas and Hattori 1986).

Predation of bacteria by protozoa and nematodes has been observed to increase the carbon and nitrogen mineralization rates in soil (Elliott *et al.*, 1980; Kuikman and Van Veen, 1989;

Rutherford and Juma, 1992). Limitation of the accessibility of pores to soil organisms has been proposed as an important control mechanism of microbial turnover in soil (Elliott *et al.*, 1980; Postma *et al.*, 1989; Heijnen *et al.*, 1991). In spite of all of the above-mentioned effects, however, ^{14}C- and ^{15}N-labelled residues derived from different plant materials decomposing in soils with a range of textures were found to be very similar, especially in the long term (> 1 year), when the rates of decomposition were adjusted to a standard temperature. An example for ^{14}C-labelled residues is presented in Figure 1.

The general objective of this thesis is to evaluate the significance of soil texture and soil structure for the short-term decomposition (within 1 year) of crop residues and soil organic matter and, *mutatis mutandis*, for the physical protection of soil organic matter.

Outline of this thesis

The starting point of this thesis is a practical model which estimates nitrogen mineralization from the nitrogen input from crops in different agricultural systems (chapter 2). This study suggests that the nature of the crop residues and the soil type do not affect the long-term decomposition (> 1 year). Cropping, soil disruption, soil texture and residue type have been found to determine short-term decomposition of crop residues or soil organic matter (see above). A laboratory and a field study in which ^{14}C-labelled and ^{15}N-labelled with different fibre content were incorporated into two soils of contrasting texture but similar organic carbon contents were conducted to test the extent to which cropping, soil texture and soil disruption are important in determining differences in crop residue decomposition in the short term i.e. 1 year (chapters 3 and 4). The effects of soil texture on the stabilization of residual ^{14}C and ^{15}N were studied in the same experiments using a chemical method to fractionate soil organic matter (only for ^{14}C) and by comparing the slower phases of decomposition of residual ^{14}C and ^{15}N in the two soils. The hypothesis that soil disruption exposes organic materials physically protected against decomposition was further tested in the same experiments. A gentle wet sieving technique was used to quantify the distribution of total carbon and nitrogen, residual ^{14}C and ^{15}N and microbial biomass in each of several aggregate size classes in the two soils (chapters 5 and 6). Different mechanisms of soil organic matter protection were deduced in the two soils and the effect of these mechanisms on the rates of mineralization of carbon, nitrogen, ^{14}C and ^{15}N also inferred. Finally, the usefulness of addressing all these factors determining the rate of decomposition of plant residues in a computer simulation model of soil organic matter turnover was evaluated (chapter 7).

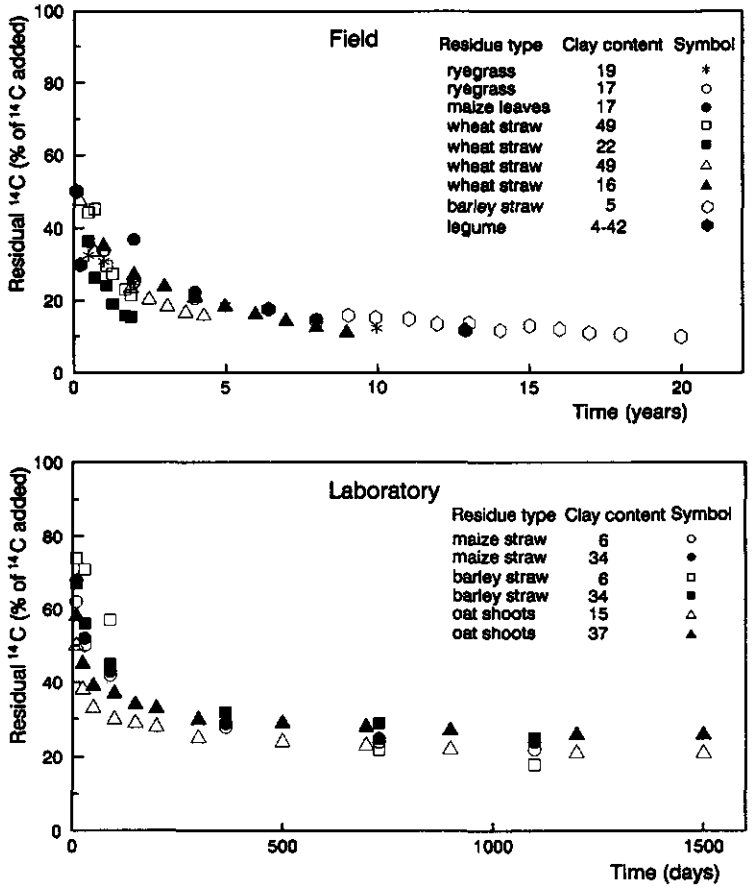

Fig. 1. Residual ^{14}C decomposition from plant residues in soils (Shields and Paul, 1973; Jenkinson, 1977; Jenkinson and Ayanaba, 1977; Sauerbeck and Gonzales, 1977; Sørensen, 1983[b]; Ladd *et al.*, 1985; Sørensen, 1987; Voroney *et al.*, 1989; Ayanaba and Jenkinson 1990; Nowak and Nowak, 1990). Decomposition rates are adjusted to the mean annual air temperature under field conditions (9°C, Rothamsted Experimental Station, Jenkinson and Ayanaba, 1977) or in the laboratory (20°C, Sørensen, 1983[b]).

REFERENCES

Ayanaba, A. and Jenkinson, D.S. (1990) Decomposition of carbon-14 labeled ryegrass and maize under tropical conditions. *Soil Science Society of America Journal* **54**, 112-115.

Cabrera, M.L. and Kissel, D.E. (1988) Potentially mineralizable nitrogen in disturbed and undisturbed soil samples. *Soil Science Society of America Journal* **52**, 1010-1015.

Craswell, E.T and Waring, S.A. (1972) Effect of grinding on the decomposition of soil organic matter-I. The mineralization of organic nitrogen in relation to soil type. *Soil Biology & Biochemistry* **4**, 427-433.

Edwards, A.P.and Bremner, J.M. (1967) Microaggregates in soils. *Journal of soil Science* **18**, 65-73.

Elliott, E.T. (1986) Aggregate structure and carbon, nitrogen and phosphorus in native and cultivated soils. *Soil Science Society of America Journal* **50**, 627-633.

Elliot, E.T. and Coleman, D.C. (1988). Let the soil work for us. *Ecological Bulletins* **39**, 23-32.

Elliott, E.T., Anderson, R.V., Coleman, D.C. and Cole, C.V (1980) Habitable pore space and microbial trophic interactions. *Oikos* **35**, 327-335.

Führ, F. and Sauerbeck, D. (1968) Decomposition of wheat straw in the field as influenced by cropping and rotation. In *Isotopes and Radiation in Soil Organic Matter Studies, pp.* 241-250. Proceedings IAEA/FAO Symposium, Vienna.

Gainey, P.L. (1936) Total nitrogen as a factor influencing nitrate accumulation in soils. *Science* **42**, 157-163.

Hart, P.B.S., Goh, K.M, and Ludecke, T.E. (1979) Nitrogen mineralization in fallow and wheat soils under field and laboratory conditions. *New Zealand Journal of Agricultural Research* **22**, 115-125.

Hassink, J. (1992) Effect of soil texture and structure on carbon and nitrogen mineralization in grassland soils. *Biology and Fertility of Soils* **14**, 126-134.

Hassink, J., Bouwman, L.A., Zwart, K.B., Bloem, J and Brussaard, L (1993) Relationships between soil texture, physical protection of soil organic matter, soil biota, and C and N mineralization in grassland soils. *Geoderma* **57**, 105-128.

Heijnen, C.E., Hok-A-Hin, C.H. and Van Veen, J.A. (1991) Protection of *Rhizobium* by bentonite clay against predation by flagellates in liquid cultures. *FEMS Microbiology Ecology* **85**, 73-80.

Helal, H.M. and Sauerbeck D.R. (1986) Influence of plant roots on the stability of soil organic matter and soil aggregates. In *Trans XIII ISSS Congress* **3**, pp. 776-777 ISSS, Hamburg.

Herman, W.H., McGill, W.B. and Dormaar, J.F. (1977) Effect of initial chemical composition on decomposition of roots of three grass species. *Canadian Journal of Soil Science* **57**, 205-215.

Huntjens, J.L.M. (1971) The influence of living plants on mineralization and immobilization of nitrogen. *Plant and Soil* **35**, 77-94.

Jansson, S.L. (1958) Tracer studies on nitrogen transformation in soil with special attention to mineralization-immobilization relationships. *Annals of the Royal Agricultural College* Sweden **24**, 101-361.

Jenkinson, D.S. (1977) Studies on the decomposition of plant material in soil. V. The effects of plant cover and soil type on the loss of carbon from ^{14}C labelled ryegrass decomposing u nder field conditions. *Journal of Soil Science* **28**, 424-434.

Jenkinson, D.S. and Ayanaba, A. (1977) Decomposition of carbon-14 labeled plant materials under tropical conditions. *Soil Science Society of America Journal* **41**, 912-915.

Jenkinson, D.S. and Rayner, J.H. (1977) The turnover of soil organic matter in some of the Rothamsted classical experiments. *Soil Science* **123**, 298-305.

Jingguo, W. and Bakken, L.R. (1989) Nitrogen mineralization in rhizosphere and non-rhizosphere soil: Effect of spatial distribution of N-rich and N-poor plant residues. In: *Nitrogen in organic wastes applied to soils* (J.A. Hansen and H. Henriksen Eds.), pp. 81-97. Academic Press, London.

Kuikman, P.J. and van Veen, J.A. (1989) The impact of protozoa on the availability of bacterial nitrogen to plants. *Biology and Fertility of Soils* **8**, 13-18.

Kuikman P.J., Liljeroth, E. and Van Veen, J.A. (1990) Interactions between plants and soil micro organisms in the rhizosphere at different soil nutrient conditions and under predation by protozoa. *Congress of Soil Science, 14th*, Volume III, Japan, pp. 43-48.

Ladd J.N., Amato, M. and Oades, J.M. (1985) Decomposition of plant material in Australian soil. III. Residual organic and microbial biomass C and N from isotope-labelled legume materia ls and soil organic matter decomposition under field conditions. *Australian Journal of Soil Research* **23**, 603-611.

Liljeroth, E., Van Veen, J.A. and Miller, H.J. (1990) Assimilate translocation to the rhizosphere of two wheat lines and subsequent utilization by rhizosphere microorganisms at two soil nitrogen concentrations. *Soil Biology & Biochemistry* **22**, 1015-1021.

Nordmeyer, H. and Richter, J. (1985) Incubation experiments on N mineralization in loess and sandy soil. *Plant and Soil* **83**, 433-445.

Nowak, G. and Nowak, J. (1990) Prolonged humification of ^{14}C-labelled oat residues in extensively fertilized soils. *Soil Biology & Biochemistry* **22**, 1089-1096.

Parton, W.J., Schimel, D.S., Cole, C.V. and Ojima, D.S. (1987) Analysis of factors controlling soil organic matter levels in Great Plains grasslands. *Soil Science Society of America Journal* 5 **1**, 1173-1179.

Paul, E.A. and Juma, N.G. (1981) Mineralization and immobilization of soil nitrogen by microorganisms. In. *Terrestrial nitrogen cycles* (F.E. Clark and T. Rosswal Eds). pp 585-601. *Ecological Bulletins* (Stockholm) **33**.

Postma, J, Walter, S. and van Veen, J.A. (1989) Influence of different initial soil moisture contents on the distribution and population dynamics of introduced *Rhizobium leguminosarum* biovar *trifolii*. *Soil Biology & Biochemistry* **21**, 437-442.

Reid, J.B. and Goss, M.J. (1982) Suppression of decomposition of ^{14}C-labelled plant roots in the presence of living roots of maize and perennial ryegrass. *Journal of Soil Science* **33**, 387-395.

Rovira, A.D. and Greacen, E.L. (1957) The effect of aggregate disruption on the activity of microorganisms in the soil. *Australian Journal Agricultural Research* **8**, 659-673.

Rutherford, P.M. and Juma, N.G. (1992) Influence of texture on habitable pore space and bacterial-protozoan populations in soil. *Biology and Fertility of Soils* **12**, 221-227.

Sallih, Z. and Bottner, P. (1988) Effect of wheat (*Triticum aestivum*) roots on mineralization rates of soil organic matter. *Biology and Fertility of Soils* **7**, 67-70.

Sauerbeck, D.R. and Gonzalez, M.A. (1977) Field decomposition of carbon-14-labelled plant residues in various soil of the Federal Republic of Germany and Costa Rica. *Soil Organic Matter Studies*. International Atomic Energy Agency, Vienna, **1**, 159-170.

Shields, J.A. and Paul, E.A. (1973) Decomposition of ^{14}C-labelled plant material under field conditions. *Canadian Journal of Soil Science* **53**, 297-306.

Sørensen, L.H. (1983a) The influence of stress treatments on the microbial biomass and the rate of decomposition of humified matter in soils containing different amounts of clay. *Plant and Soil* **75**, 107-119.

Sørensen, L.H. (1983b) Size and persistence of the microbial biomass formed during the humification of glucose, hemicellulose, cellulose, and straw in soils containing different amounts of clay. *Plant and Soil* **75**, 121-130.

Sørensen, L.H. (1987) Organic matter and microbial biomass in a soil incubated in the field for 20 years with ^{14}C-labelled barley straw. *Soil Biology & Biochemistry* **19**, 39-42.

Tian, G., Kang, B.T. and Brussaard, L. (1992[a]) Biological effect of plant residues with contrasting chemical compositions under humid tropical conditions - decomposition and nutrient release. *Soil Biology & Biochemistry* **24**, 1051-1060.

Tian, G., Kang, B.T., and Brussaard, L. (1992[b]) Effect of chemical composition on N, Ca, and Mg release during incubation of leaves from selected agroforestry and follow plant species. *Biogeochemistry* **16**, 103-119.

Tian, G., Brussaard, L. and Kang, B.T. (in press) Effect of earthworms and millipedes on breakdown of plant residues with contrasting chemical compositions under humid tropical conditions. *Soil Biology & Biochemistry*.

Tisdall, J.M. and Oades, J.M. (1982) Organic matter and water-stable aggregates in soils. *Journal of Soil Science* **33**, 141-163.

Van der Linden, A.M.A., Van Veen, J.A. and Frissel, M.J. (1987) Modeling soil organic matter levels after long-term application of crop residues and farmyard and green manures. *Plant and Soil*, **101**, 21-28.

Van Gestel, M., Ladd, J.N. and Amato, M.(1991) Carbon and nitrogen mineralization from two soils of contrasting texture and microaggregates stability: influence of sequential fumigation, drying and storage. *Soil Biology & Biochemistry* **23**, 313-322.

Van Veen, J.A. and Paul, E.A. (1981) Organic carbon dynamics in grassland soils. I. Background information and computer simulation. *Canadian Journal of Soil Science* **61**, 185-201.

Van Veen, J.A., Ladd, J.N. and Amato, M. (1985) Turnover of carbon and nitrogen through the microbial biomass in a sandy loam and a clay soil incubated with [^{14}C(U)]glucose and [^{15}N](NH_4)SO_4 under different moisture regimes. *Soil Biology & Biochemistry* **17**, 747-756

Vargas, R. and Hattori, T (1986) Protozoan predation of bacterial cells in soil aggregates. *FEMS Microbial Ecology*. **38**, 233-242.

Verberne, E.L.J., Hassink, J. De Willigen, P., Groot, J.J.R., and Van Veen, J.A. (1990). Modelling organic matter dynamics in different soils. *Netherlands Journal of Agricultural Science* **38**, 221-238.

Voroney, R.P., Paul, E.A. and Anderson, D.W. (1989) Decomposition of wheat straw and stabilization of microbial products. *Canadian Journal of Soil Science* **69**, 63-77.

Waksman, S.A. and Tenney, F.G. (1928) The composition of natural organic materials and their decomposition in the soil: II. Influence of age of plant upon the rapidity and nature of its decomposition- rye plants. *Soil Science* **26**, 155-171.

Whitmore, A.P. and Parry, L.C. (1988) Computer simulation model of the behaviour of nitrogen in soil and crop in the Broadbalk continuous wheat experiment. In *Nitrogen efficiency in agricultural soils* (D.S. Jenkinson and K.A. Smith, Eds), pp. 418-432. Elsevier, London.

CHAPTER 2

A SIMPLE MODEL FOR ESTIMATING THE CONTRIBUTION OF NITROGEN MINERALIZATION TO THE NITROGEN SUPPLY OF CROPS FROM A STABILIZED POOL OF SOIL ORGANIC MATTER AND RECENT ORGANIC INPUT

F.J. Matus[1] and J. Rodríguez[2]

[1] *DLO Research Institute for Agrobiology and Soil Fertility, P.O. Box 129, 9750 AC Haren, The Netherlands*

[2] *División de Suelos del Departamento de Ciencias Vegetales, Pontificia Universidad Católica de Chile, Casilla 114 D, Santiago, Chile*

Received 5 August 1993. Accepted 27 December 1993

Key words: mineralization rate, nitrogen, stabilized organic matter, crop rotation

Summary - A simple model was developed to estimate the contribution of nitrogen (N) mineralization to the N supply of crops. In this model the soil organic matter is divided into active and passive pools. Annual soil mineralization of N is derived from the active pool. The active pool comprises stabilized and labile soil organic N. The stabilized N is built up from accumulated inputs of fresh organic N during a crop rotation but the labile N is a fraction of total N added, which mineralizes faster than the stabilized N. The passive pool is considered to have no participation in the mineralization process. Mineralization rates of labile and stabilized soil organic N from different crop residues decomposing in soil were derived from the literature and were described by the first-order rate equation $dN/dt = -K*N$, where N is the mineralizable organic N from crop residues and K is a constant. The data were grouped K_1 by short-term (0-1 year) and K_2 by long-term (0-10 years) incubation. Because the range of variation in K_2 was smaller than in K_1 we felt justified in using an average value to derive N mineralization from the stabilized pool. The use of a constant rate of K_1 was avoided so net N mineralization during the first year after addition is derived directly from the labile N in the crop residues. The model was applied to four Chilean agro-ecosystems, using daily averages of soil temperature and moisture. The N losses by leaching were also calculated. The N mineralization varied between 30 and 130 kg N ha^{-1} yr^{-1} depending on organic N inputs. Nitrogen losses by leaching in a poorly structured soil were estimated to be about 10% of total N mineralized. The model could explain the large differences in N mineralization as measured by the potential N mineralization at the four sites studied. However, when grassland was present in the crop rotation, the model underestimated the results obtained

from potential mineralization.

INTRODUCTION

In Chile, N fertilizer recommendations for arable crops are based on measurement of the amount of soil mineral N in early spring. Such investigations, provide little information because (i) the soil mineral N is measured in the top layer only (0-20 cm) as a starting point to establish different response curves with increasing rates of application of N fertilizer, and (ii) there is little differentiation between agro-ecological zones. Since 1977, estimates of N-mineralization in alluvial and volcanic ash derived soil (allophanic soil) have been made in Chile. Annual N mineralization was calculated from potentially mineralizable N after Stanford & Smith (1972) and used as input to a N balance-sheet equation to obtain the optimum N application rate (Oyanedel and Rodríguez, 1977; Rodríguez and Silva, 1984[a]). The weak point of this approach has been the estimation of the potentially mineralizable N. Potentially mineralizable N may be overestimated by drying and sieving the samples before incubation (Cabrera and Kissel, 1988) or by the bias introduced into the estimation of the parameters as the time of incubation increases (Dendooven, 1990).

Nitrogen mineralization during a growing season comes from soil organic matter and recent organic inputs. Attempts to relate N mineralization to the total soil organic N content failed, especially where allophanic soils are involved, because a large part of soil organic matter is bound to the mineral part of soil (Zunino *et al.*, 1982).

We have developed a practical model that predicts the release of N from a range of soils in different agricultural zones with large variation in fresh organic inputs, weather conditions and soil types; the model avoids the use of the potentially mineralizable N of Stanford and Smith (1972).

In this paper we describe the model, and its application to four Chilean agro-ecosystems.

METHODS

Model description

Fig. 1 shows the fate of crop residues decomposing in soil. The N input in crop residues is divided into resistant (fN_{Ro}) and labile ($fN_L = 1\text{-}fN_{Ro}$) N fractions which enter the active pool of soil organic N. The fractions fN_L and fN_{Ro} contribute to the formation of the labile (N_l) and stabilized (N_s) pool of soil organic N, respectively. By definition, the labile N mineralizes rapidly within the first year after the addition and N_s mineralizes slowly. Annual N decomposition from the resistant N input, N_{Ro} (kg N ha^{-1} yr^{-1}), may be described by the equation:

$$dN_{Ro} / dt = -K_2 * N_{Ro} \quad (1)$$

where N_{Ro} is $fN_{Ro}*N_{RS}$, N_{RS} fresh organic N inputs (kg N ha^{-1} yr^{-1}), K_2 is the rate constant of N mineralization ($years^{-1}$) and t time (years).

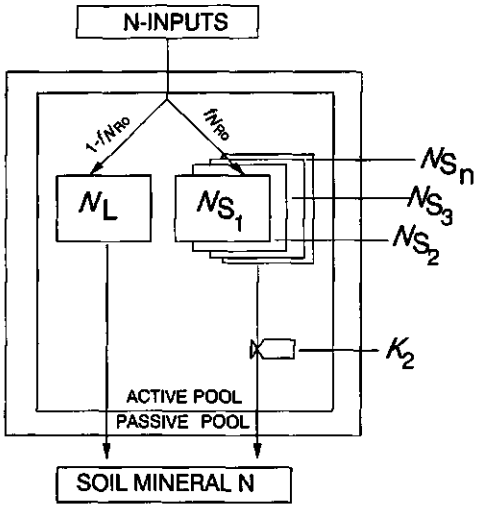

Fig. 1. Fate of N input from crop residues applied to the soil: N_L, N labile; N_{S1} .. N_{Sn} stabilized pool of soil organic N accumulated from the resistant N fraction fN_{Ro} from recent organic inputs, after n years of crop rotation; K_2 rates of N mineralization from N_S.

The N_S pool is accumulated from N_{Ro} and comprises organic materials resistant to decomposition (e.g. lignin), and presumably physically protected soil organic matter localized in soil aggregates which are not penetrated by microflora and fauna (Van Veen *et al.*, 1985). The accumulation rate and size of the N_S pool depend on K_2 and N inputs from crop residues during a crop rotation. In this study the passive pool in Fig. 1 is assumed not to increase or decrease during a growing season. However in the long term, particularly in grassland soil, the release of N from the passive pool may be significant. In the model the N losses by denitrification and N gain by mineralization from the passive pool, were not considered, so the N quantities gained or lost were regarded to be roughly in balance.

Estimation of rate constants (K_1, K_2) and N fractions (fN_{Ro}, fN_L)

Tables 1 and 2 show the rates, K_1 and K_2 and the resistant N fraction, fN_{Ro}, respectively. The data

Table 1. Rates of N mineralization K_1 and K_2 from several ^{15}N-labelled organic materials decomposing in soil in short-term (< 52 weeks) or long-term (0-10 years) incubation

Decomp. period	Lab. or field	Plant materials	C:N	Incub. temp (°C)	k adjusted 16°C (yr^{-1})	References
K_1, undisturbed soils (weeks)						
0- 2	field	mustard	15.1	7.0[A]	18.09	1
0- 2	lab.	lentil	24.0	21.0	1.41	2
0- 4	field	legume	15.3	16.2	1.85	3
0-12	lab.	vigna	18.5	24.3	0.79	4
0-12	lab	medic	18.5	24.3	0.65	4
0-43	field	white clover leaflets	11.0	11.0	2.67	5
0-43	field	red clover leaflets	11.0	11.0	2.16	5
0-43	field	timothy leaflets	16.0	11.0	2.11	5
0-43	field	timothy roots	29.0	11.0	1.25	5
0-43	field	field beans stems+petioles	28.0	11.0	1.22	5
0-43	field	field beans roots	28.0	11.0	0.37	5
0-52	lab.	barley root	35.7	22.5	0.71	6
All rates K_1 (mean)				2.8		
K_2, cultivated soils (years)						
0- 4	field	wheat straw	20.4	12.0[B]	1.66	7
4-10	field	wheat straw	20.4	12.0[B]	0.14	7
K_2, undisturbed soils						
0.6- 2	field	wheat straw	73.1	14.4	0.14	8
1 -2.7	field	mustard	15.0	7.0[A]	0.52	1
1 -2.7	field	ryegrass	33.7	7.0[A]	0.50	1
0.1- 4	field	legume	13.1	16.0	0.24	9
1 - 4	field	legume	15.3	16.2	0.07	3
1 - 8	field	legume	15.3	16.2	0.06	10
1 - 5	lab.	barley root	17.1	22.5	0.03	6
1 - 5	lab.	barley top	35.7	22.5	0.04	6
All rates K_2 (mean of undisturbed soils)				0.2		

lab.: laboratory; decomp.: decomposition

A: assumed temperature; B: mean temperature only considered in months with temperatures over 0°C

1 Jensen (1992); 2 Janzen and Kucey (1988); 3 Ladd *et al.* (1981[b]); 4 Fox *et al.* (1990); 5 Müller *et al.* (1988); 6 Broadbent and Nakashima (1974); 7 Voroney *et al.* (1989); 8 Amato *et al.* (1987); 9 Amato *et al.* (1984); 10 Ladd et al. (1985)

Table 2. Resistant N fraction (fN_{Ro}) from several ^{15}N-labelled crop residues decomposing in soil

Crop residues	C:N ratio	Resistant fN_{Ro} [A]	Reference
mature wheat straw	73.1	0.91	8 **
field beans	27.5	0.82[B]	11
subterranean clover	20.9	0.77[B]	11
red clover	21.0	0.74[B]	11
barley-root	35.7	0.72	6 **
timothy grass	18.9	0.69[B]	11
rye grass	33.7	0.68	1 **
white clover	19.3	0.65[B]	11
barley-top	17.1	0.62	6 **
legume	15.3	0.58	10 **
white mustard	15.1	0.53	1 **
young wheat straw	20.4	0.47	7 **
mean		0.68	

**: references, see Table 1
11: Müller and Sundman (1988)
A: values derived from intercept on y axis from Eq. (4) fitted to the long-term (≥1 year) data points
B: soil organic ^{15}N remaining 10 months after addition

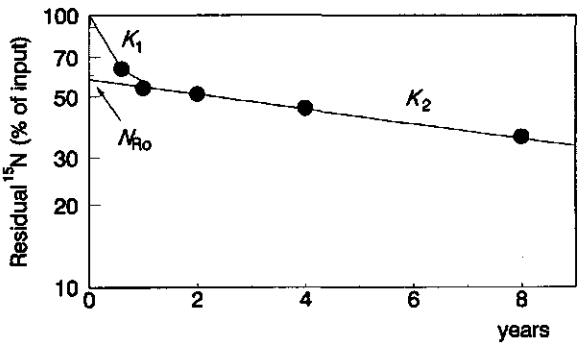

Fig. 2. Fit by first-order equations on the rapid and slow mineralization phase from long-term incubation experiments of ^{15}N-labelled legume residues decomposing in undisturbed soil under field conditions (Ladd *et al.*, 1981[b]; 1985). Note logarithmic scale on *Y* axis.

were derived from published N mineralization experiments using ^{15}N-labelled materials (Broadbent and Nakashima, 1974; Ladd *et al.*, 1981[b]; Amato *et al.*, 1984; Ladd *et al.*, 1985; Amato *et al.*, 1987; Janzen and Kucey, 1988; Müller *et al.*, 1988; Müller and Sundman, 1988; Voroney *et al.*, 1989;

Fox *et al.*, 1990; Jensen, 1992). Nitrogen mineralization was assumed to proceed by two independent reactions following to first-order kinetics. Table 1 shows two groups of rates: K_1 for short-(0-1 year) and K_2 for long-term (0-10 years) incubations. The rates K_2 were obtained from the long-term data (i.e. > 1 year). The residual ^{15}N was plotted on a logarithmic scale against time in order to use the linear regression technique (Stanford and Smith, 1972). An example of long-term N mineralization (Ladd *et al.*, 1981[b]; 1985) is shown in Fig. 2. The slope of the regression line provides the rate K_2 and its intercept on the *Y*-axis, the resistant N fraction fN_{Ro}. The labile N fraction, fN_l, is calculated as $100\text{-}fN_{Ro}$, and mineralizes rapidly with rate K_1. The rates K_1 were derived from a regression, using a one pool from the residual organic ^{15}N found in the short-term (i.e.< 1 year) (Fig. 2). The straight line obtained in short-term and long-term data points, indicated, whether a one- or two-pool model was appropriate to describe the N mineralization process (Voroney *et al.*, 1989).

Effect of soil temperature and moisture content

The N mineralization heavily depends on environmental factors such as soil temperature and moisture content. The effect of temperature on N mineralization is given by:

$$K_2(T_a) = K_2(T_o) * \exp(0.0616 * (T_a - T_o)) \qquad (2)$$

where $K_2(T_a)$ is the constant decay rate K_2 adjusted to the temperature T_a ($5 \leq T_a \leq 35°C$) and $K_2(T_o)$ is the rate K_2 at temperature T_o ($5 \leq T_o \leq 35°C$). It follows from (2) if T_a is $\leq T_o$ the ratio $K_2(T_a)/K_2(T_o)$ is ≤ 1. Eq.(2) was derived from first-order rates K estimated at 5, 15, 25 and 35°C by Stanford *et al.* (1973). The K values obtained between 5 and 35 °C were plotted on a semilog scale against temperature. Eq.(2) is the regression line obtained from plotted data with $R^2 = 0.99$ and slope 0.0616. From (2) it follows that a 10 °C change in temperature leads to a change of $K_2(T_a)$ by a factor of approximately two (Fig. 3).

Cavalli and Rodriguez (1975) indicated that the relationship between soil moisture and N mineralization is linear between field capacity (0.33 kPa) and permanent wilting point (15 kPa). Thus

$$K_2(W_a) = K_2(W_o) * (1.11 * (W_a / W_o) - 0.138) \qquad (3)$$

where $K_2(W_a)$ is the rate constant K_2 adjusted to the soil moisture W_a (g g^{-1}) and $K_2(W_o)$ is the rate K_2 at soil moisture W_o (g g^{-1}). The variation in soil moisture was calculated on a daily basis from precipitation, class A pan evaporation and from the physical soil parameters such as: bulk density, soil moisture at field capacity and permanent wilting point.

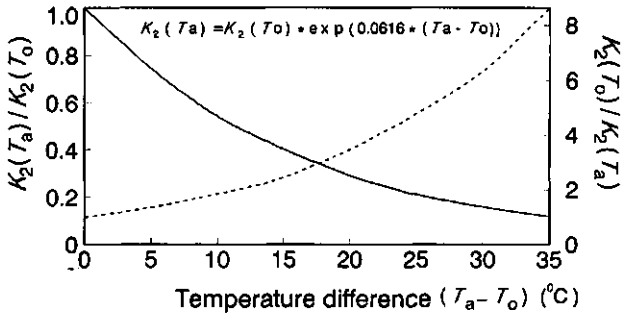

Fig. 3. Soil temperature function; $K_2(T_o)$ = constant decay rate of N mineralization K_2 at temperature T_o; $K_2(T_a)$ = K_2 adjusted by temperature T_a.

Table 1 shows the N mineralization rates adjusted to 16°C assuming that the rate of mineralization doubles for every 10°C increase in temperature.

Estimation of stabilized pool of soil organic N

The integrated form of equation (1) is:

$$\ln N_{Ro} = \ln N_{Ro_t} - K_2 * t \tag{4}$$

Eq. (4) is the logarithmic form of:

$$N_{Ro} = N_{Ro_t} * \exp(-K_2 * t) \tag{5}$$

where N_{Ro} is the resistant N input. After n years of crop rotation, the size of the stabilized pool of soil organic N, N_S can be calculated for each resistant N input from t = 1 as:

$$\begin{aligned} N_{S_n} &= N_{Ro_t} * \exp(-K_2)^n \\ &+ N_{Ro_{t+1}} * \exp(-K_2)^{n-t} \\ &+ N_{Ro_{t+2}} * \exp(-K_2)^{n-(t+1)} \\ &+ \ldots + N_{Ro_n} * \exp(-K_2)^t \end{aligned}$$

$$= \sum_{t=1}^{n} N_{Ro_t} * \exp(-K_2)^{n-t+1} \tag{6}$$

from (6) it follows that the equilibrium of N_S (when $n \to \infty$) after several years of crop rotation may

be approximated by:

$$N_{S_-} = \frac{\frac{\sum_{t=1}^{n} N_{Ro_t}}{n}}{1-\exp(-K_2)} - \frac{\sum_{t=1}^{n} N_{Ro_t}}{n} = \frac{\frac{\sum_{t=1}^{n} N_{Ro_t}}{n}}{\exp(K_2)-1} \quad (7)$$

and (7) is approximated by:

$$N_{RS_-} = \frac{\sum_{t=1}^{n} N_{Ro_t}}{n * K_2} \quad (8)$$

The time when equilibrium is reached (t_{eq}), is defined as 95% of N_S at steady state:

$$t_{eq} = \frac{-\ln 0.05}{K_2} \quad (9)$$

Nitrogen mineralization in the soil

The total N mineralization in soil is calculated from the N_L and N_S pools. The total soil mineral N in year n (N_{min_n}, kg N ha^{-1} yr^{-1}) is given by:

$$N_{min_n} = N_{L_n} + N_{S_n} * (1-\exp(-K_2 * t)) \quad (10)$$

where N_L is N_{Rs}*(1-fN_{Ro}). From Table 1 it appears that K_1 is so large that all N_L is mineralized within a year.

Nitrate leaching

The nitrate movement in soil was calculated according to a modified equation of Burns (1980):

$$L = A * (P/(P+(W_a * b_d)))^z \quad (11)$$

where L is the amount of N leached (kg N ha^{-1}) below the rooting depth z (cm), A the amount of N-NO_3^- (kg N ha^{-1}) present to depth z, W_a soil moisture content (g g^{-1}), b_d soil bulk density (g cm^{-3}) and P percolation below depth z. The percolation can be estimated as function of the surplus of precipitation (P_p, cm) over class A pan evaporation (E_V, cm) and the amount of water (h, cm) present between field capacity and permanent wilting point up to depth z:

$$P = P_P - E_V , \qquad if \qquad P_P - E_V > h \tag{12}$$

$$P = 0 , \qquad if \qquad P_P - E_V \leq h \tag{13}$$

Field sites used to evaluate the model

The model was evaluated in four Chilean agro-ecosystems: Osorno, Temuco, Cauquenes and Rancagua. These sites are representative of agriculture in Southern and Central Chile. Tables 3 and 4 show some soil characteristics and the N inputs from crop rotations at each site. Cauquenes is a dryland area and the N input varies, depending on the amount and distribution of rainfall in winter. The soil of Cauquenes is highly eroded with a shallow rooting depth. Osorno and Temuco are soils derived from volcanic ash. In the last two sites the amount and distribution of rainfall within the year result in intermediate levels of productivity. Rancagua is an irrigated area with well structured soils which allow the highest grain yields in the country to be obtained.

The crop rotations in Osorno and Temuco have been approximately the same for more than 80 years and in Cauquenes and Rancagua for more than 100 years and reflect the time since colonization. The yields of maize and wheat have increased in Rancagua during the last 25 years as a result of the use of fertilizer and the introduction of new cultivars; the low input of N (Table 4) from wheat management is caused by residues being burned after harvest. In Osorno, although the mixed grassland (clover + ryegrass) can be maintained for between 6 and 20 years, pasture of 8 years duration is usual. The crop rotation and annual N inputs were estimated by Sierra and Rodríguez (1986), Matus and Rodríguez (1989), and Rodríguez (1990). The N inputs were obtained from the dry matter yield and by measurement of the N concentrations in above- and below-ground (0-20 cm) residues after harvest. The input of N from roots and dead plant material that enter the soil during a growing season were also considered (Matus and Rodríguez, 1989). From Osorno, Temuco and Rancagua, a total of 29 soil samples (0-20 cm) were taken and the potentially mineralizable soil N was measured by the method of Stanford and Smith (1972) (Rodríguez and Silva, 1984[b]; Rodríguez and Sierra, 1987). In the Cauquenes soil mineral N was estimated by the uptake of wheat in control field experiments without added N (García, 1973).

RESULTS

Accumulation of stabilized pool of soil organic N

The two components required to build up the N_s pool are the constant decay rate K_2 (Table 1) and

Table 3. Precipitation, air temperatures and soil characteristics of the top soil layer (0-20 cm) for the four Chilean agro-ecosystems

	Osorno	Temuco	Cauquenes	Rancagua
Soil properties				
Soil Order (Soil Taxonomy)	Andisols	Ultisols	Alfisols	Inceptisols
Clay content (%)	23.0	44.1	19.0	26.5
Allophane (%)	21.0	0.0	0.0	0.0
pH (1:2.5, water)	5.3	5.1	6.0	6.8
soil org. C (t ha^{-1})	145.0	66.2	18.0	34.2
soil org. N (t ha^{-1})	12.8	6.3	1.8	2.9
C:N ratio	11.3	10.5	10.0	11.8
Physical parameters				
Bulk density (g cm^{-3})	0.8	1.0	1.2	1.2
Water holding capacity (%)	22.0	9.0	8.0	10.0
Temperature (°C)				
Sept.-March (spring-summer)	12.6	13.2	18.0	17.3
April-Aug. (autumn-winter)	8.0	8.6	11.4	11.9
Annual air temperature	10.7	11.2	15.3	15.1
Precipitation (mm)				
Sept.-March (spring-summer)	380.0	352.0	100.1	78.4
April-Aug. (autumn-winter)	810.0	862.0	532.5	516.1
Annual precipitation	1190.0	1214.0	632.6	594.5

Table 4. Nitrogen inputs from crop residues in a crop rotation at four Chilean agro-ecosystems

Agro-eco-system	Crop rotation	N input (N_{RS}) (kg N ha yr^{-1})
Osorno	sugar beet	50
	wheat	39
	grassland (ryegrass + clover, 6 or 20 years)	150
Temuco	rapeseed	72
	wheat	30
	clover (2 years)	150
Cauquenes	wheat	19
	grass (5 years)	36
Rancagua	maize	98
	wheat	43

the resistant N fraction, fN_{Ro} (Table 2). Table 1 shows that (i) the constant decay rates K_2 in the long-term (0-10 years incubations) vary less than K_1 in the short-term (0-52 weeks) incubations, (ii) K_1 and K_2 decrease as the time of incubation increases and (iii) K_2 diminishes little in

incubations longer than three years. The rates K_2 in undisturbed soil were about 7 times lower than K_1 (the highest value of K_1, was excluded) and the range of variation between the highest and lowest K_2 values from undisturbed soil incubations between 1 to 8 years was 2 times compared to 50 times for the range of variation of K_1. The relatively low variation in K_2 for several organic inputs decomposing in different soil and weather conditions has already been observed by Kolenbrander (1974). Experiments with ^{15}N- and ^{14}C-labelled plant materials have confirmed this finding (Ladd *et al.*, 1985; Voroney *et al.*, 1989). The constant rates K_2 for N may be compared with those for carbon (C) in long-term incubation (Jenkinson, 1981). Table 5 shows the

Table 5. Decay rates K_2 of ^{14}C-labelled organic materials decomposing in soil in long-term incubation

Decomp. period (years)	Incubation lab. or field	Plant materials	Incub. temp. (°C)	K Incub. temp. (yr^{-1})	K 16°C (yr^{-1})	Reference
1-10	lab.	wheat straw	9.0	0.12	0.18	16
1-10	field	rye grass	9.1	0.09	0.14	12
1- 3	lab.	glucose	20.0	0.17	0.13	14
0- 6	lab.	glucose	20.0	0.14	0.11	13
1- 3	lab.	hemicellulose	20.0	0.13	0.10	14
2-10	field	wheat straw	12.0[A]	0.08	0.10	7 **
1- 3	lab.	barley straw	20.0	0.11	0.09	14
12-20	field	barley straw	7.0	0.05	0.09	15
1- 3	lab.	cellulose	20.0	0.10	0.08	14
1- 4	field	legume	16.2	0.08	0.08	3 **
1- 3	lab.	maize straw	20.0	0.10	0.08	14
0- 6	lab.	cellulose	20.0	0.09	0.07	13
mean					0.10	

lab.: laboratory; decomp.: decomposition
**: references, see Table 1
12: Jenkinson (1977); 13: Sørensen (1972); 14:Sørensen (1983); 15: Sørensen (1987); 16: Sauerbeck and Gonzalez (1977)
A: mean temperatures only considered in months with temperatures over 0°C

K_2 values adjusted to 16°C from several organic materials. These figures vary between 0.07 and 0.18 yr^{-1} and agree well with the observed data of N mineralization from Table 1.

Table 6 shows an example of the calculation of the N_s pool in a crop rotation of maize-wheat in Rancagua. Annual average values of K_2 were obtained after adjusting for soil temperature (Eq. 2) and moisture (Eq. 3) on daily basis. Annual N inputs were obtained from Table 4 and fN_{Ro} was taken as the value 70% as obtained from Table 2. The accumulation curve started from the beginning of land cultivation. After several years of crop ratation an equilibrium of N_s pool, estimated by Eq. 8, is reached. The time of equilibrium, t_{eq}, was 81 years as calculated

by Eq. 9.

Fig. 4 shows an accumulation curve of the N_s pool for Rancagua and Osorno. Both agro-ecosystems approach equilibrium. In Osorno grassland contributed most to the size of N_s pool;

Table 6. Example of accumulation (Janssen, 1984) of N_s pool (rounded figures) in Rancagua as calculated for addition of 98 and 43 kg N ha^{-1} $year^{-1}$ (N_{RS}) from crop residues in a crop rotation of maize and wheat. The fN_{Ro} was taken 0.7 and K_2, 0.037 yr^{-1}.

Crop residues added in the rotation	Years after addition				
	1	2	3	4..	..81
maize	66	64	62	59..	..3[A]
wheat		29	28	27..	..2[B]
maize			66	64..	..4
wheat				29..	..2
.	.	.	.	.	.
.	.	.	.	.	.
total	66	93	156	179..	..1334

[A] = 98*0.7*exp(-0.037*81)
[B] = 43*0.7*exp(-0.037*80)

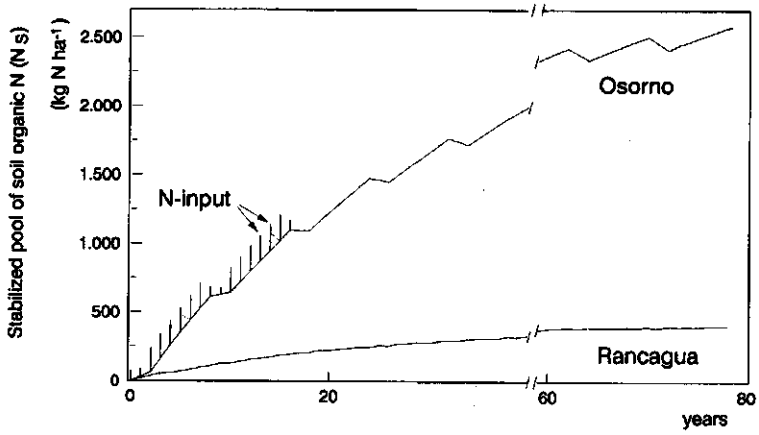

Fig. 4. Accumulation of stabilized pool of soil organic N, N_s, as shown for two crop rotations in the agro-ecosystems of Osorno and Rancagua.

however, the slower rate K_2 in this sites means that the equilibrium is reached later as the crop rotation continues. Rancagua has been cultivated longer than Osorno and its higher mean annual soil temperature (Table 3) raises the value of decomposition (K_2) and thus equilibrium is

Table 7. Stabilized pool of soil organic N (N_S), rates K_2 and time of equilibrium (t_{eq}) at four Chilean agro-ecosystems as estimated by the model

Agro-eco-systems	Crop rotation	Constant rate K_2[A] (yr^{-1})	Equilib. time t_{eq} (years)	Stabilized organic N (N_S) 80 or 100 yr. (kg N ha^{-1})	Stabilized organic N (N_S) steady-state
Osorno		0.031	97		
	sugar beet-			2302	2755
	wheat-			2416	2699
	6 years grassland			2571	2806
Osorno		0.031	97		
	sugar beet-			2725	3120
	wheat-			2744	3053
	20 years grassland			2974	3182
Temuco		0.034	88		
	rapeseed-			1894	2064
	wheat-			1850	1996
	2 years clover			1928	2064
Cauquenes		0.038	79		
	wheat-			569	583
	5 years native grass			578	590
Rancagua		0.037	81		
	maize-			1294	1327
	wheat			1277	1303

A: annual average; adjusted by soil temperature and moisture

reached more quickly.

Table 7 shows the N_S pools and t_{eq} at the four field sites as calculated by the model. The N_S pools were predicted for 80 years of crop rotation in Osorno and Temuco and 100 years for Cauquenes and Rancagua. The N_S pools at equilibrium varied between 600 and 3000 kg N ha^{-1} and were greatest where the N inputs came from grassland. The constant rates K_2 ranged from 0.031 to 0.038 yr^{-1}; this variation was also mainly caused by differences in temperature amongst sites (Table 3). The predicted time of equilibrium has already been reached at Cauquenes and Rancagua but Osorno and Temuco have not yet achieved this level.

Annual net N-mineralization

Table 8 shows the contribution of annual net N mineralization to the N supply of crops as

Table 8. Contribution of net N mineralization to the N supply of crops as estimated by the model at four Chilean agro-ecosystem.

Agro-eco-systems	Crop rotation	Preceding crop	Mineralization N_L[A]	Mineralization N_S[B]	N-leaching	N-supply
			(kg N ha^{-1})			
Osorno	sugar beet-wheat-6 years grassland	grassland[C]	45	71	0	116
		sugar beet	14	75	0	89
Osorno	sugar beet-wheat-20 years grassland	grassland[C]	45	85	0	130
		sugar beet	14	85	0	99
Temuco	rape seed-wheat-2 years clover	clover[C]	46	64	< 2	110
		rape seed	22	63	< 2	85
Cauquenes	wheat-5 years native grass	grass[C]	10	22	3	29
Rancagua	maize-wheat	wheat	19	47	< 2	66
		maize	12	48	< 2	60

A: N_L = labile N
B: N_S = stabilized pool of soil organic N
C: ploughed in the last year of grassland

calculated by the model. Nitrogen supply was estimated from the amount of N minerlaized according to Eq. (10) minus the nitrate leaching obtained with Eq. (11). For example, in Osorno the N supply for sugar beet was calculated from the mineralization of N_L and N_S for the last year of grassland minus the N leached below the rooting depth of 20 cm. The N supply ranged over-all sites from 29 to 130 kg N ha^{-1} yr^{-1}. The most important contribution of N_L and N_S to the total soil mineral N was in the agro-ecosystem of Osorno. The lowest N supply and the largest N leaching were predicted in the eroded soils of Cauquenes.

Model evaluation

Fig. 5 shows the plot of net N mineralization estimated by the model and the potential N mineralization estimated by the method of Stanford and Smith (1972). The total soil mineral N calculated by the model was less than predicted as shown by the 1:1 line. The under-prediction was worse in the agro-ecosystems where pasture was present. The quantity and quality of organic

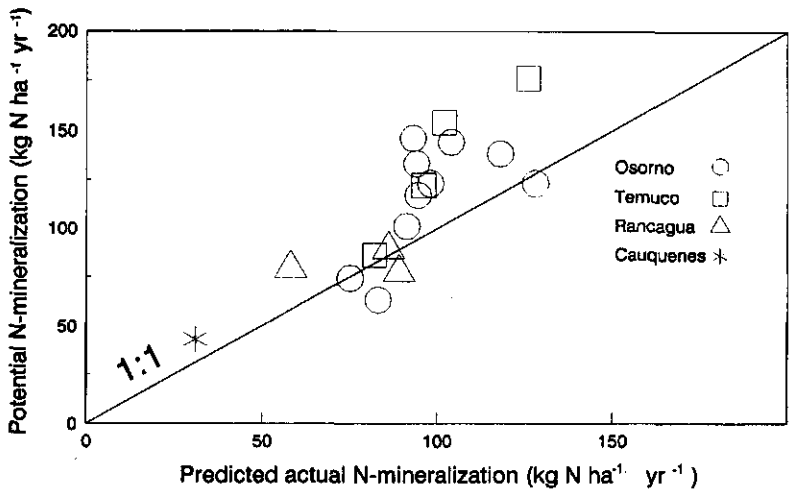

Fig. 5. Plot 1:1 of soil mineral N as predicted by the model at the four sites studied and potential N mineralization estimated by the method of Stanford and Smith (1972).

input in grassland soils may be the cause of an increase in N mineralization in the field (Nordmeyer and Richter, 1985).

DISCUSSION

Model parameter estimation: rate constant K_2 and resistant N fraction fN_{Ro}

Plant materials decay with different rates depending on their C:N ratio and lignin content. In the early stages of decomposition a rapid N mineralization (high *K* value) is expected in crop residues with low C:N ratio and low lignin content (Parton *et al.*, 1987). As the time of incubation increases, lower rates are observed (Hunt, 1977) (Table 1). The decay rate K_1 represents the easily decomposable fractions (e.g. amino-sugars, proteins, hemicellulose, cellulose and microbial products) followed by a slow constant decay rate K_2, which was assumed to reflect supply from the stabilized N_s pool.

It can be argued that the use of empirical values of K_2 is not possible because they decrease as the incubation time increases (Janssen, 1984). The turnover time of soil organic matter has been considered as the resultant of a series of first-order kinetic reactions from intermediate pools (Van Veen *et al.*, 1981). An oversimplification may be made by assuming that K_2 represents the N mineralization of a homogeneous stabilized pool of soil organic N. Evidence for the existence of an N_s pool can be found indirectly from radiocarbon dating and the fractionation of soil

organic matter. Complete turnover of apparently very humic organic material (passive pool) requires between 600 to 1400 years. These figures are 50-100 times lower than the mineralization of young soil organic matter (active pool) derived from recent organic input (Fig. 1). The turnover rates for young soil organic matter have been found to range between 0.047 and 0.069 yr^{-1} (Balesdent *et al.*, 1988; Balesdent *et al.*, 1987; Schwartz *et al.*, 1986), close to the rates K_2 between 1 and 8 years of incubation studies (Table 1).

Table 2 shows a positive and significant ($P<0.05$) correlation ($r = 0.70$) between fN_{Ro} and C:N ratios of different plant materials. The mature crop residues showed the largest fN_{Ro} values, suggesting a greater resistance to decomposition as indicated by their high C:N ratios as well.

Stabilized pool of soil organic N

Table 7 shows that the grassland soil, generally accumulated more soil organic N in the stabilized pool. Cauquenes, even when grassland was present in the crop rotation, exhibited the lowest accumulation in N_s pool because of the quantity and presumably the quality of the N input, incorporated every year (Table 4). No large differences in the size of the N_s pool calculated from short or long periods of grassland in Osorno were predicted. Averaged over the crop rotation, 20 years of grassland resulted in a N_s pool 16% greater than 6 years of grassland. There were little differences between sites and managements in times of equilibrium (Table 7). The values ranged from 79 years in Cauquenes to 97 years in Osorno. The time of equilibrium were obtained according to Eq. 9 with the rates K_2 corrected for soil temperature and moisture. Neither soil moisture nor soil temperature at each site were limiting factors to the decomposition in spring. A small range of soil moisture and temperature amongst sites were observed, this little differences may have accounted for similar time of equilibrium . It seems probable that the agro-ecosystem approaches equilibrium (Fig. 4) (Martel and Paul, 1974) and that the N_s pool originates from the accumulation of humified fraction (N_{Ro}) or stabilized soil matter (Balesdent *et al.*, 1988) or because of the accumulation of "young soil organic matter" (Janssen, 1984) with faster turnover rates than a passive pool.

Soil disruption by cultivation is another factor that can affect the size of N_s pool. The empirical parameters used in the model were obtained from undisturbed soil, but soil tillage enhances the rate K_2 of N mineralization. Evidence of this has been observed in short-term incubation experiments (Nordmeyer and Richter, 1985; Gregorich *et al.*, 1989) and in disturbed grassland soils with an increasing number of years of cultivation (Lathwell and Bouldin, 1981). Soil tillage can not have a long-term effect on N mineralization, as outputs from the N_s pool may equal the inputs in the long run. In the first years, however, (and for the duration of most agronomic experiments), N mineralization will be increased if starting from a relatively large N_s pool of undisturbed soil, e.g. grassland soils.

Annual net N mineralization and model evaluation

Table 8 shows substantial differences in the calculated net N mineralization at the four sites. In agro-ecosystems with several years of grassland, greater N mineralization is expected. Leaching losses in winter months predicted by the model were smaller than 2.4 kg N ha^{-1} yr^{-1}, except in the poorly structured soil of Cauquenes where the N losses were between 10 and 13% of the total amount mineralized. About 60-90% of the N mineralized originated from the N_S pool. The remaining N came from the labile N pool, derived from the N input from the immediately preceding crop. In the literature, an average of 17% of the inputs from ^{15}N-labelled plant material is accounted for as N losses by denitrification and leaching (Müller and Sundman, 1988; Müller, 1988; 1987; Wagger *et al.*, 1985; Ladd *et al.*, 1981[a]; 1983). If this figure is subtracted from the mineralization N_L in Table 8, the total soil mineral N (N_L+N_S) must be decreased by 2-7%. These losses are small and therefore were not considered in the annual estimation. However, in Temuco the denitrification losses may have been greater than the other sites because of its higher rainfall and heavy clay soils (Table 3).

Annual N mineralization in Osorno was about 0.7-1.0% from the total soil organic N compared to 1.7-2.3% in the other agro-ecosystems. In this soil the higher soil organic matter content may have a large passive pool compared with an elevated amount of humic compounds. The allophanic soils have a high anion-adsorption capacity favoring the stabilization of organic polymers and microbial substances formed during decomposition (Zunino *et al.*, 1982). In general the model output corresponded well with the potential N mineralization (Fig. 5). A double exponential model has been used to describe N mineralization in disturbed soils according to first-order kinetics (Cabrera and Kissel, 1988) but the results are variable when the parameter values are compared from different incubation times (Dendooven, 1990).

CONCLUSIONS

The purpose of our model was to simulate a range of N mineralization in different field sites where little information was available. The estimated release of N by mineralization was close to the soil mineral N observed at the four sites studied and any differences were explained on the basis of N input from crop rotations. The model estimates a dynamic equilibrium after several cycles of crop rotation. When this equilibrium is achieved the variation in the N_S pool depends on the N input from individual crops. The model suggests that the contribution to the N supply of the crop is determined more by cropping history reflected in the accumulation of a stabilized pool of soil organic matter. The model has the advantage that it is simple, however it does not consider the short-term dynamics of N mineralization within a growing season (e.g N immobilization), and must therefore be used on an annual basis.

Acknowledgements - We are grateful to Drs. A.P. Whitmore and L. Brussaard for their contribution to the discussion and valuable comments on the manuscript. We thank H. Terburg for the english corrections in the manuscript.

REFERENCES

Amato M, Jackson, R. B., Butler, J. H. A. and Ladd, J. N. (1984) Decomposition of plant material in Australian soils. II Residual organic ^{14}C and ^{15}N from legume plant parts decomposing under field and laboratory conditions. Aust. J. Soil Res. 22, 331-341.

Amato M, Ladd J N, Ellington A, Ford G, Mahoney J E, Taylor A C and Walsgott D 1987 Decomposition of plant material in Australian soils. IV. Decomposition in situ of ^{14}C- and ^{15}N-labelled legume and wheat materials in a range of Southern Australian soils. Aust. J. Soil Res. 25, 95-105.

Balesdent J, Mariotti A and Guillet B 1987 Natural ^{13}C abundance as a tracer for studies of soil organic matter dynamics. Soil Biol. Biochem. 19, 25-30.

Balesdent J, Wagner G H and Mariotti A 1988 Soil organic matter turnover in long-term field experiments as revealed by carbon-13 natural abundance. Soil Sci. Soc. Am. J. 52, 118-124.

Broadbent F E and Nakashima T 1974 Mineralization of carbon and nitrogen in soil amended with carbon-13 and nitrogen-15 labeled plant material. Soil Sci. Soc. Am. Proc. 38, 313-315.

Burns I G 1980 A simple model for predicting the effects of winter leaching of residual nitrate on the nitrogen fertilizer need of spring crops. J. Soil Sci. 31, 187-202.

Cabrera M L and Kissel D E 1988. Evaluation of a method to predict nitrogen mineralized from soil organic matter under field conditions. Soil Sci. Soc. Am. J. 52, 1027-1031.

Cavalli I and Rodríguez J 1975 Efecto del contenido de humedad en la mineralizacion de nitrogeno en nueve suelos de la provincia de Santiago. Ciencia e Inv. Agr. 2, 101-112.

Dendooven L 1990 Nitrogen mineralization and nitrogen cycling. Doctoral thesis, Katholieke Universiteit te Leuven, Belgium, 180 p.

García R 1973 Productividad y manejo de los suelos chilenos. Programa de las Naciones Unidas para el Desarrollo. FAO.

Fox R H, Myers R J K and Vallis I 1990. The nitrogen mineralization rate of legume residues in soil as influenced by their polyphenol, lignin, and nitrogen contents. Plant and Soil 129, 251-259.

Gregorich E G, Kachanoski R G and Voroney R P 1989 Carbon mineralization in soil size fractions after various amounts of aggregate disruption. J. Soil. Sci. 40, 649-659.

Hunt H W 1977 A simulation model for decomposition in grassland. Ecology 58, 469-484.

Janssen B H 1984 A simple method for calculating decomposition and accumulation of "young" soil organic matter. Plant and Soil 76, 297-304.

Janzen H H and Kucey R M N 1988 C, N, and S mineralization of crop residues as influenced by crop species and nutrient regime. Plant and Soil 106, 35-41.

Jenkinson D S 1977 Studies on the decomposition of plant material in soil. V. The effects of plant cover and soil type on the loss of carbon from ^{14}C labelled ryegrass decomposing under field conditions. J. Soil Sci. 28, 424-434.

Jenkinson D S 1981 The fate of plant and animal residues in soil, In The Chemistry of Soil Processes. Eds. D.J. Greenland and M.H.B. Hayes. pp. 505-561.

Jensen E S 1992 The release and fate of nitrogen from catch-crop materials decomposing under field conditions. J. Soil Sci. 43, 335-345.

Kolenbrander G J 1974 Efficiency of organic manure in increasing soil organic matter content.Trans. 10th Int. Congr. Soil Science, Moscow, 1974, 2, pp. 129-136.

Ladd J N, Oades J M and Amato M 1981a Distribution and recovery of nitrogen from legume residues decomposing in soil sown to wheat in the field. Soil Biol. Biochem. 13, 251-256.

Ladd J N, Oades J M and Amato M 1981b Microbial biomass formed from ^{14}C, ^{15}N- labelled plant material decomposing in soils in the field. Soil Biol. Biochem. 13, 119-126.

Ladd J N, Oades J M and Amato M 1983. Utilization by wheat crops of nitrogen from legume residues decomposing in soils in the field. Soil Biol. Biochem. 15, 231-238.

Ladd J N, Amato M and Oades J M 1985 Decomposition of plant material in Australian soil. III. Residual organic and microbial biomass C and N from isotope-labelled legume materials and soil organic matter decomposition under field conditions. Aust. J. Soil Res. 23, 603-611.

Lathwell D J and Bouldin D R. Soil organic matter and soil nitrogen behavior in cropped soils. Trop. Agric.(Trinidad) 58: 341-348.

Martel Y A and Paul E A 1974. Effects of cultivation on the organic matter of grassland soils as determined by fractionation and radiocarbon dating. Can. J. Soil Sci. 54, 419-426.

Matus F J and Rodríguez J 1989. Modelo simple para estimar el suministro de N en el suelo. Ciencia e Inv. Agr. 16: 33-45.

Müller M M 1987 Leaching of subterranean clover-derived N from loam soil. Plant and soil 102, 185-191.

Müller M M 1988 The fate of clover-derived nitrogen (^{15}N) during decomposition under field conditions: effect of soil type. Plant and Soil 105, 141-147. Müller M M and Sundman V 1988 The fate of nitrogen (^{15}N) released from different plant materials during decomposition under field conditions. Plant and Soil 105, 133-139.

Müller M M, Sundman V, Soininvaara O and Meriläinen A 1988 Effect of chemical composition on the release of nitrogen from agricultural plant materials decomposing in soil under field conditions. Biol. Fertil. Soils 6, 78-83.

Nordmeyer H and Richter J 1985 Incubation experiments on N mineralization in loess and sandy soil. Plant and Soil 83, 433-445.

Oyanedel C and Rodríguez J 1977. Estimación de la mineralizacion del N en el suelo. Ciencia e Inv. Agr. 4, 33-44.

Parton J W, Schimel D S, Cole C V and Ojima D S 1987 Analysis of factors controlling soil organic matter levels in Great Plains grasslands. Soil. Sci. Soc. Am. J. 51, 1173-1179.

Rodríguez J 1990 La fertilización de los cultivos un método racional. División de Suelos del Departamento de Ciencias Vegetales, Pontificia Universidad Católica de Chile, 406 p.

Rodríguez J and Sierra C 1987 Efecto del historial de cultivo en el pool de N lábil y resistente del suelo. Ciencia e Inv. Agr. 14, 63-70.

Rodríguez J and Silva H 1984a Estimación de la mineralizacion de N y requerimientos de N de los cultivos en suelos con distinto manejo. Ciencia e Inv. Agr. 11, 73-80.

Rodríguez J and Silva H 1984b Nitrogeno potencialmente mineralizable en andisoles. Ciencia e Inv. Agr. 11, 81-88.

Sauerbeck D R and Gonzalez M A 1977 Field decomposition of carbon-14-labelled plant residues in various soil of the Federal Republic of Germany and Costa Rica. Soil Organic Matter Studies. Inter. Atomic Energy Agency, Vienna, Vol. 1, p. 159-170.

Schwartz D, Mariotti A, Lanfranchi R and Guillet B 1986 $^{13}C/^{12}C$ ratios of soil organic matter as indicators of vegetation changes in the Congo. Geoderma 39, 97-103.

Sierra C and Rodríguez J 1986 Efecto del manejo del suelo en el suministro de N. Ciencia e Inv. Agr. 13, 231-240.

Sørensen L H 1972 Role of amino acid metabolites in the formation of soil organic matter. Soil Biol. Biochem. 4, 245-255.

Sørensen L H 1983 Size and persistence of the microbial biomass formed during the humification of glucose, hemicellulose, cellulose, and straw in soils containing different amounts of clay. Plant and Soil 75, 121-130.

Sørensen L H 1987 Organic matter and microbial biomass in a soil incubated in the field for 20 years with ^{14}C-labelled barley straw. Soil Biol. Biochem. 19, 39-42.

Stanford G and Smith S J 1972 Nitrogen mineralization potentials of soil. Soil Sci. Soc. Am. Proc. 36, 465-472.

Stanford G, Frere J H and Schwaninger D H 1973 Temperature coefficient of soil nitrogen mineralization. Soil Sci. 115, 321-323.

Veen J A van, Ladd J N and Amato M 1985 Turnover of carbon and nitrogen through the microbial biomass in a sandy loam and a clay soil incubated with [$^{14}C(U)$]glucose and [^{15}N]$(NH_4)SO_4$ under different moisture regimes. Soil Biol. Biochem. 17, 747-756.

Van Veen J A, McGill W B, Hunt H W, Frissel M J and Cole C V 1981 Simulation models of the terrestrial nitrogen cycle. In F.E. Clark & T.Rosswall (Eds). Terrestrial nitrogen cycle. Processes ecosystems strategies and management impacts. Ecol. Bull. Stockholm 33, 35-48, Sweden.

Voroney R P, Paul E A and Anderson D W 1989. Decomposition of wheat straw and stabilization of microbial products. Can. J. Soil Sci. 69, 63-77.

Wagger M G, Kissel D E and Smith S J 1985. Mineralization of nitrogen from nitrogen-15 labelled crop residues under field conditions. Soil Sci. Soc. Am. J. 49, 1220-1226.

Zunino H, Borie F, Aguilera S, Martin J P and Haider K 1982. Decomposition of ^{14}C-labelled glucose, plant and microbial products and phenols in volcanic ash-derived soils of Chile. Soil Biol. Biochem. 14, 37-43.

CHAPTER 3

EFFECT OF SOIL TEXTURE, SOIL STRUCTURE AND CROPPING ON THE DECOMPOSITION OF CROP RESIDUES. I. RESIDUAL ORGANIC C AND MICROBIAL BIOMASS C DERIVED FROM DECOMPOSING ^{14}C-LABELLED WHEAT STRAW IN A CLAY AND SAND SOIL

F.J. Matus

DLO Research Institute for Agrobiology and Soil Fertility, P.O. Box 129, 9750 AC Haren, The Netherlands

Summary - The hypothesis was tested that soil texture, soil structure and cropping, play major roles in the dynamics of carbon in soil. Two soils with similar total carbon contents and crop management histories, but contrasting textures were amended with ^{14}C-labelled wheat residues. The soils were incubated in pots in a greenhouse for 266 days. Seven treatments either without or with addition of nitrogen (N) fertilization (150 kg ha^{-1}) were included: (1) cropping plus N, (2) cropping without N, (3) bare soil followed by cropping plus N, (4) bare soil followed by cropping without N, (5) bare plus N (6) mixed soils plus N and (7) bare followed by mixing plus N. No effect of cropping on residual ^{14}C, microbial biomass ^{14}C or residual ^{14}C in humus as measured by acid hydrolyzation was observed. Only in the clay soil, cropping with N-fertilization tended to reduce the loss of labelled C in comparison with bare soil, but this effect disappeared after 70 days of decomposition. Living roots of wheat plants did not suppress or stimulate the decomposition of labelled residues in either soil. Apparently soil microbes preferred the labelled residue rich in N (2.4%) over root-released carbon. Mixing did not influence the decline of residual ^{14}C, microbial biomass ^{14}C and residual ^{14}C in humus forms. This suggests that recently formed soil organic matter was not exposed to further degradation after hand-mixing of both clay and sand soils. No effect of soil texture was observed on residual ^{14}C, microbial biomass ^{14}C and ^{14}C in humus forms. The amounts of humified ^{14}C (^{14}C associated with the clay and silt particles) were similar in both soils. However, the ^{14}C concentration of the clay and silt particles was nine times higher in the sand soil than in the clay soil. This suggests that silt and clay particles of coarse-textured soils are more concentrated with newly produced soil organic matter than that of fine textured soils.

INTRODUCTION

Cropping has been suggested to influence the decomposition of soil organic matter. The presence of living roots induced less decomposition of residual ^{14}C compared with bare soils in a number of field experiments (Führ and Sauerbeck, 1968; Shields and Paul, 1973; Jenkinson, 1977). This was

attributed to lower microbial activity resulting from restricted aeration (Führ and Sauerbeck, 1968) or to less water being available in the planted soils (Shields and Paul, 1973; Jenkinson, 1977). In other studies where the aeration and soil moisture content were controlled both positive and negative effects have been described (Sallih and Bottner, 1988). Mechanical disruption of soil aggregates (Rovira and Greacen, 1957; Crasswell and Waring, 1972; Hassink, 1992) or drying and rewetting (Sørensen, 1983[a]; van Gestel *et al.*, 1991) have been described to increase the decomposition rate of soil organic matter. This phenomenon can be attributed to the release of physically protected organic matter from soil aggregates (Elliott, 1986). Hassink *et al.*, (1993) studied the effect of soil texture on pore accessibility to bacteria and their predators and found that bacterial biomass was positively correlated with the pores 0.2-1.2 μm in diameters in a range of grassland soils. Gregorich *et al.* (1991) studying ten soils reported that the microbial biomass derived from ^{14}C-labelled glucose was positively correlated with the clay content after 90 days of incubation, but the residual organic ^{14}C was similar in all soils. The similar rates of decomposition of different ^{14}C-labelled plant residues under field (Shields and Paul, 1973; Jenkinson, 1977; Sauerbeck and Gonzalez, 1977; Sørensen, 1987) and laboratory conditions (Sørensen, 1983[b]; Nowak and Nowak, 1990) for different soils suggests that the decomposition of the plant residues is independent of the clay content of the soils. Amato and Ladd (1992), however, found a positive correlation between residual ^{14}C derived from legumes and clay content of soil. There are no studies so far in which the relationship between soil texture, soil structure and cropping on the decomposition of labelled residues have been investigated at the mean time in soils of different textures. One of the useful techniques in this type of study is the application of labelled ^{14}C organic materials to distinguish between "fresh" and "old" organic matter decomposition in soil. Using this technique in the present study the hypothesis was tested that the interaction between soil texture, soil structure and root-induced microbial activity can modify the carbon dynamics in soil. The specific aims of this study were to investigate the effect of cropping and soil disruption on (1) the rate and extent of decomposition of ^{14}C-labelled wheat (*Triticum aestivum* L.) incorporated in a clay and sand soil, and (2) the stabilization of the residual organic ^{14}C and newly synthesized microbial biomass in a clay and sand soil.

MATERIALS AND METHODS

Soils

A clay and a sand soil were used; some properties of the soil are given in Table 1. Both soils have a long agricultural history and were obtained from farmers' fields located in Groningen, the Northeast province of The Netherlands. The soils have been cropped in a rotation with cereals, sugar beet and beans in combination with bare-fallow periods since 1986. The upper (0-15 cm)

layer of each soil was collected in November 1990, mixed separately, sieved through a 0.5 cm mesh size, and stored field moist at 5°C.

Table 1. Soil properties

Soil properties		Clay	Sand
Chemical			
Total C (%)		1.76	1.62
Total N (%)		0.19	0.11
C:N		9.30	14.70
$N\text{-}NH_4^+$ (mg kg^{-1})		2.10	0.40
$N\text{-}NO_3^-$ (mg kg^{-1})		14.50	13.50
P (mg l^{-1})		12.20	21.80
K (mg kg^{-1})		273.90	107.90
Mg (mg kg^{-1})		nd	116.00
$CaCO_3$ (%)		0.30	0.60
pH-KCl		6.98	7.22
Physical			
Particle size distribution (g kg^{-1})			
Clay	(< 2 µm) (%)	481	47
Fine silt	(2-20 µm)	289	34
Coarse silt	(20-50 µm)	174	265
Fine sand	(50-250 µm)	38	535
Coarse sand	(250-2000 µm)	18	119
Water holding capacity (%)			
-0.2 kPa		48	24
-25.0 kPa		40	21
-1555.0 kPa		22	5
Bulk density (g cm^{-3})		1.15	1.30

nd = not determined

Incorporation of ^{14}C labelled and unlabelled wheat straw

In February 1992 ^{14}C-labelled wheat straw (particle size < 2 mm; specific activity: 2.69 MBq g^{-1} C; C:N 17; N, 2.37%) was incorporated into the soils at a rate of 0.12 mg g^{-1} soil (on dry matter basis). Since the quantity of the labelled residue was small in comparison with the amount usually added in the pot experiments (1-5 mg straw g^{-1} soil), an additional amount of 3.2 mg g^{-1} soil of unlabelled wheat straw with comparable chemical composition (C:N 16; N, 2.18 %) was mixed with the labelled residue.

Treatments

Seven treatments: cropping, soil mixing, bare, a combination of bare and mixing, a combination of bare and cropping and absence or supply of nitrogen were used in an experiment carried out for 266 days (Table 2). Seventy two four-liter pots for the cropping treatment and 60 two-liter

Table 2. Treatments in incubation studies

Period[1]			Number
1	2	Nitrogen[2]	Pots[3]
Cropped-N	Cropped-N	no	24
Cropped+N	Cropped+N	yes	24
Bare-N	Cropped-N	no	12
Bare+N	Cropped+N	yes	12
Bare+N	Bare+N	yes	24
Bare+N	Mixing+N	yes	12
Mixing+N	Mixing+N	yes	24
Total			132

[1] Period 1 = February-May; Period 2 = August-November.
[2] yes = 150 kg N ha^{-1} in each period; no = no N fertilization.
[3] Either six or three samplings, two soils, two replications.

pots for the bare and mixing treatments were prepared. In the cropping treatment spring wheat (*Triticum aestivum* L.) was used. In February 1992, after addition of labelled residue, four pre-germinated wheat plants were transplanted (referred to as sowing) to each pot and thinned to two plants per pot one week later. Wheat plants were harvested in May after 90 days (first period). The second period started in August 1992; wheat was harvested 100 days later. To assess the effect of cropping or mixing during the last phase of decomposition, the soils that during the first period had been conserved under bare condition were mixed or cropped during the second period. The mixing treatment consisted of disruption of the soil aggregates to < 0.5 cm in diameter by hand. Mixing was performed six times: 13, 42, and 74 days after sowing in the first period, and 0, 30 and 70 days after sowing in the second period. On day 0, both mixing and sowing coincided. Soils in the bare treatments were conserved undisturbed, and the small amount of spontaneous vegetation growing in the pots was cut and left on top of the soil within the pots. Each treatment was replicated twice in a completely randomized design.

Mineral fertilization

In each period, three nutrient solutions for four- or two-liter pots containing, respectively

0.09 g or 0.04 g P, 0.22 g or 0.11 g K and 0.12 g or 0.06 g Mg were added. For four- or two liter pots that received N-fertilization, 0.33 g or 0.15 g N was applied on three occasions; 25% of the total application, 10 days before sowing and 25% and 50%, 30 and 60 days after sowing, respectively.

Incubation, watering and samplings

All pots were placed in a greenhouse and the environment was controlled as follows: 16 hours light, (516 W m^{-2}) at 21 ± 4°C and 8 hours dark at 16 ± 2°C; the water holding capacity of the soil (WHC) was kept at 75%, i.e. 0.18 and 0.36 g water g^{-1} dry soil in sand and clay soil respectively. All pots were watered to make up the weight loss between consecutive waterings. A total of 103 waterings at intervals of 1 or 2-days were required. Between the first harvest and the second time of sowing, each pot was covered with a plastic plate to avoid losses of water. Samplings were conducted at 0, 30, 50, 90, 195, 216 and 266 days after the labelled residue was incorporated. Sampling on days 90 and 266 coincided with harvest. At each sampling and for each treatment, the soil of two replicate pots were used. Shoots (leaves, stems and ears at ripening) of wheat plants were cut off at the level of the soil surface and roots, picked by hand. Shoots and roots were cleaned and dried at 60°C (for 48 hours). Moist soil from each pot was thoroughly mixed and stored at 5°C for analysis.

Analytical procedures

At each sampling date the following measurements were made: (1) total C, (2) C present in the microbial biomass, and (3) C present in the non-hydrolyzable fraction of soil organic matter.

Total C and residual ^{14}C. Total C was measured using a wet oxidation method. A duplicate subsample (1 g) was placed on the bottom of a screw glass tube (3.6 cm diameter, approximately 250 cm^3) with 5-ml of a concentrated acid-dichromate solution, consisting of 117.7 g H_2SO_4, 78.4 g H_3PO_4 and 160 g $K_2Cr_2O_7$. Each tube contained a glass vial with 10 ml 0.5 M NaOH and was immediately sealed (teflon screw lid) and left standing for 30 min at room temperature. Thereafter, the tubes were heated for 2 hours at 160°C and left overnight. After precipitating the carbonate with excess $BaCl_2$ (0.75 M) the total amount of carbon was determined in a 5-ml aliquot of the NaOH trapping solution and titrated with a standard solution of 0.5 M HCl to end point pH 8.3 (Dalal, 1979). The labelled carbon trapped as CO_2 was measured in a 1-ml aliquot of the NaOH diluted with 1-ml of demineralized water plus 10 ml of a scintillation cocktail (Insta-Gel, Packard Instruments Company) in a low potassium glass scintillation vial. Samples were counted in a liquid scintillation counter, Rackbeta II 1215, Wallac, programmed according to a previously

prepared quenching curve. One or two hours counting were needed to reach a standard deviation less than 2%.

Microbial biomass C. Microbial biomass, was assessed by direct determination of water soluble C in the soil solution after fumigation (Van Ginkel *et al.*, 1994). Fifty grams of soil was adjusted to 65% WHC and fumigated in moist, ethanol-free, chloroform vapor at room temperature for 24 hours. After removing the chloroform by repeated evacuations, the soil was placed in a PVC centrifuge tube of similar design to that described by Van Ginkel *et al.* (1994) and centrifuged for 1 hour at 20,000 *g*. Another 50 g of non-fumigated subsample, adjusted to 65% WHC, was similarly extracted. The radioactivity of ^{14}C in the extract was determined in a 1-ml sample plus 10 ml of a scintillation cocktail and counted in a program set with a quenching curve prepared as above. Unlabelled soluble C was measured using a dry oxidation method. A duplicate 30 µl sample was injected in an analyzer for total organic C, TOC-500, Shimadzu, (oxygen consumption 150 ml min^{-1}) equipped with a furnace to reach 680°C and an infra-red CO_2 analyzer. Microbial biomass was obtained from the difference between the amount of C in soil solution from fumigated soils and the amount of C from non-fumigated soils (FE_c). The factor K_c relating the flush of C (fumigated minus unfumigated samples) with the amount of microbial biomass was 0.186 (Van Ginkel *et al.*, 1994).

Non-hydrolyzable ^{14}C. Soil organic matter fractionated by acid hydrolyzation removes non-humified carbohydrates, proteinaceous compounds, fungal structures and dead microbial tissues (Choudhri and Stevenson, 1957; Shields *et al.*, 1973). Here, the expression non-hydrolyzable C was used to refer to the C remaining in soil after acid hydrolyzation. Either 3 g of clay or sand soil (on dry matter basis) were boiled under reflux at 100 °C for 16 hours in 1.5 N sulphuric acid (Sørensen, 1981). The soil suspension was filtered under vacuum and the soil was rinsed with demineralized water (3 x 10 ml) and dried at 70 °C. Total C and the radioactivity were analyzed as described above.

Statistical analysis

Analysis of variance (ANOVA; Genstat 5.0), and Student's t-test were used to analyze the differences of means between treatments. The level of probability at which significant differences were compared was set at the 5%.

RESULTS

Residual ^{14}C

The residual ^{14}C (^{14}C remaining in the soil) declined in two phases: a rapid decay during the first 30 days after incorporation and a slow phase between 30 and 266 days of decomposition. About 105% of total ^{14}C added was recovered on day 0 (Fig. 1).

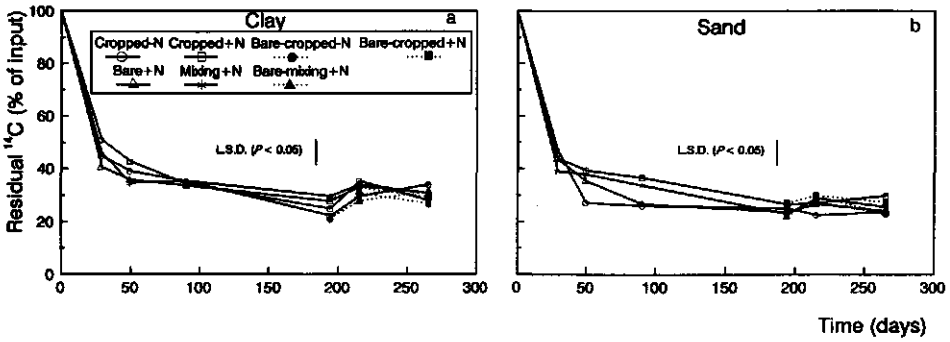

Fig. 1. Residual ^{14}C (% of input) in (a) clay soil and in (b) sand soil for the seven treatments studied (mixing times on days: 13, 42, 74, 171, 201 and 241).

Cropping and mixing. After 30 days of decomposition, between 40% and 55% of applied ^{14}C was still found in the soil. Cropping with N-fertilization, on day 30, tended to reduce the decomposition compared with bare soils in the experiment with clay soil (Fig. 1a), but this effect was not observed in the sand soil (Fig. 1b). On day 50 cropped sand soils with N fertilization tended to retain more residual ^{14}C than cropping without N-fertilization (Fig. 1b), however this effect was not observed in the clay soil (Fig. 1a).

Mixing did not affect the decline of residual ^{14}C, neither in the rapid decline nor in the slow phase of decomposition (Fig. 1a and b).

Soil texture. No effect of soil texture was found on residual ^{14}C (cf. Fig. 1a and b). The decay rates of residual ^{14}C between days 30 and 266, assuming first order kinetics, are shown in Table 3. The decay rates were slightly slower in the clay, (0.0015 day^{-1}; $T_{1/2}$ = 577 days) than in the sand soil (0.0020 day^{-1}; $T_{1/2}$ = 385 days), but the difference was not significant.

Microbial biomass ^{14}C

Cropping and mixing. There was no effect of cropping on labelled microbial biomass (Fig. 2a and b). After 30 days of incubation in sand soil (Fig. 2b), microbial biomass ^{14}C in the cropped soils tended to be higher than in the bare soil. In the clay soils (Fig. 2a) this tendency was not found. As incubation progressed, ^{14}C present in microbial biomass was found to be similar for all treatments.

Table 3. First order decomposition rate constants (day^{-1}) of residual ^{14}C between 30 and 266 days of incubation in clay and sand soil

	Clay	Sand
Cropped-N	0.0016	0.0020
Cropped+N	0.0022	0.0019
Bare-cropped-N	0.0018	0.0022
Bare-cropped+N	0.0012	0.0014
Bare+N	0.0010	0.0021
Mixing+N	0.0012	0.0020
Bare-mixing+N	0.0015	0.0021
Mean	0.0015	0.0020

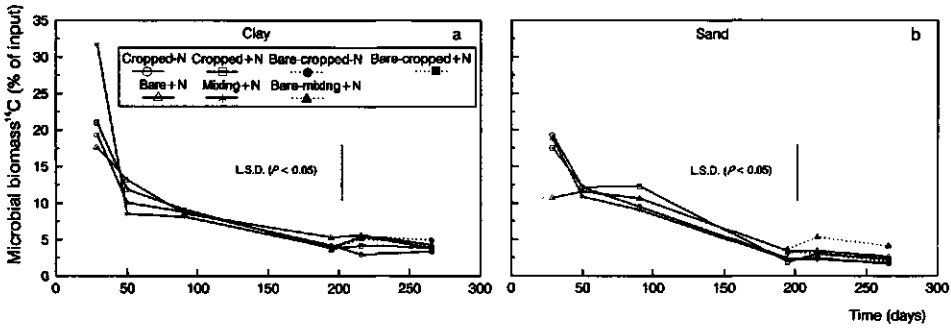

Fig. 2. Microbial biomass-^{14}C (% of input) in (a) clay and (b) sand.

At the beginning of the incubation, mixing, increased the amount of microbial biomass ^{14}C only in the clay soil (Fig. 2a). After 266 days, no significant differences between mixing, cropping and bare treatments were established in spite of the six mixings that had been conducted.

Soil texture. Soil texture hardly influenced microbial biomass ^{14}C. The overall average of

treatments, about 22% of the input of ^{14}C in the clay and 19% in the sand soil were found in the microbial biomass after 30 days of incubation (Fig. 2a and b). These values declined at nearly the same rates during the followings days of decomposition.

Non-hydrolyzable ^{14}C

Cropping and mixing. The amounts of residual organic ^{14}C left after acid hydrolyzation are presented in Fig. 3. In the clay soil, non-hydrolyzable ^{14}C in cropping plus N-fertilization was significantly ($P < 0.05$) higher than in bare soils 30 and 50 days of decomposition (Fig 3a). This difference was not found in the sand soil (Fig. 3b). During the following samplings, cropping hardly affected the amount of non-hydrolyzable ^{14}C in both soils, except in one treatment in one particular time in the clay soil (Fig. 3a).

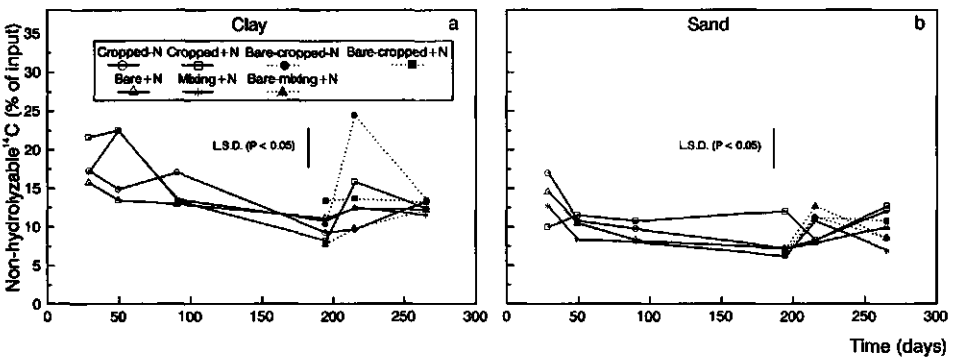

Fig. 3. Residual ^{14}C (% of input) in (a) clay and in (b) sand soil after acid hydrolyzation.

There was no effect of mixing on the amount of non-hydrolyzable ^{14}C throughout the six samplings, except in the clay soil on day 50 where significantly higher values than in the bare soils were observed (Fig 3a). In general the higher values found in the clay soils were attributable to a higher sampling error (as noted by the large standard error of the data), because in sand soil no such increase in the non-hydrolyzable ^{14}C (Fig. 3b) was found and in clay soil this increase in the residual ^{14}C was not observed (Fig. 1a).

In general, the results for non-hydrolyzable ^{14}C were also consistent with the results for ^{14}C measured in the acid solution (after hydrolyzation) in cropped, mixed and bare soils (data not shown).

Soil texture. The amount of ^{14}C in the non-hydrolyzable organic forms was slightly less in the sand than in the clay soil. As an overall average of the treatments and sampling times, 14% of added

^{14}C were present in non-hydrolyzable forms in the clay and 10% in the sand soils.

DISCUSSION

General

In recent studies the microbial biomass-C has been positively correlated with the clay content of soil (Amato and Ladd, 1992; Gregorich *et al.*, 1991). These results must be treated with caution, since microbial biomass has also been correlated with the soil organic matter content, which is itself often correlated with clay content (Amato and Ladd, 1992). In the present investigation soils were used that had been under a similar crop rotation for at least 5 years and had nearly similar total C contents. Therefore effects of soil texture will not be confounded with the effect of soil organic matter. Conclusions about the behaviour of the microbial biomass in relation to clay content may therefore be safely drawn from the results.

Effect of cropping

No effect of cropping on residual ^{14}C, microbial biomass ^{14}C and non-hydrolyzable ^{14}C was observed. In clay soils, cropping with N- fertilization tended to reduce the loss of labelled C in the residue during the initial stages of decomposition (Fig 1a). This effect was, however, more apparent than real, since the differences disappeared after 70 days of decomposition and the same effect was not observed in the sand soil (Fig 1b). Several studies under field (e.g. Führ and Sauerbeck, 1968) or laboratory conditions (e.g. Reid and Goss, 1982; Bek, 1994) have reported a reduction in the decomposition of labelled crop residues. However, Sallih and Bottner (1988) described that roots in cropped soils induced faster decomposition of labelled residue than bare soils after 100 days of incubation. The labelled crop residues used in this study containing 4.2% N had a low C:N ratio (17). The results support the observation that cropping has little influence on the mineralization of labelled residues rich in N (Jingguo and Bakken, 1989). During the first 30 days of decomposition in the clay soil and the first 50 days in the sand soil, labelled residues tended to decompose less in N-fertilized cropped soils than in non-fertilized cropped soils (Fig. 1a and b). At high levels of N in soils microbes prefer root derived materials over native soil organic matter or crop residues; at low N levels this preference does not seem to occur (Liljeroth *et al.*, 1990; Kuikman *et al.*, 1990). Roots may induce more microbial activity in the rhizosphere by exudation of organic compounds (Helal and Sauerbeck, 1986). However, such an effect on the microbial biomass was not evident (Fig. 2).

Effect of mixing

No effects of mixing of the soil on residual organic ^{14}C, microbial biomass ^{14}C and non-hydrolyzable ^{14}C were observed, except on microbial biomass ^{14}C on the first sampling in the clay soil (Fig. 2a). Moreover, mixing did not have any effect on unlabelled total C and unlabelled microbial biomass C (data not shown). Matus (1995) found that recently added labelled soil organic matter within macro-aggregates (> 250 μm) cannot be easily exposed to biodegradation by soil disaggregation. It is possible that tillage of virgin soil causes disintegration of macro-aggregates into micro-aggregates (< 250 μm) and that micro-aggregates are relatively unaffected (Elliott, 1986). It seems that after long-term land tillage, the macro-aggregates remaining in the soil are resistant to disintegration by further cultivation. The soils used in this study have been in arable use for a long time. This suggests that mixing was not sufficiently disruptive to break up the macro- aggregates left after cultivation.

Effect of soil texture on residual ^{14}C, microbial biomass ^{14}C and non-hydrolyzable ^{14}C

The decline of residual ^{14}C (Fig. 1), microbial biomass ^{14}C (Fig. 2) and hydrolyzable ^{14}C (Fig. 3) was similar in both soils throughout the incubation, except non-hydrolyzable ^{14}C for the treatments cropping plus N-fertilization and mixing in the clay soil. Table 4 present the half-life values of the residual ^{14}C of the soils studied here (Table 3) and other soils with different textures incubated under similar controlled conditions, but with different organic materials added.

Table 4. The half-lives (years) of residual ^{14}C derived from several organic materials decomposing in a range of soils with different contents of clay and silt as compared with the half-life obtained in this study for different incubation periods

Number of soils used	Clay+Silt (< 20 μm) %	Half-life (years) Period of incubation 30-90 days	90 - ≤ 700 days	Reference
4	8-75	0.2-0.5	2	Sørensen (1981)
4	11-77	0.4-0.6	2	Sørensen (1983*)
2	8-77	0.3-0.5	2-3	This study
2	29-50	n.d.	2-4	Sallih and Bottner (1988)

n.d. = not determined

The half-lives, calculated in different periods of decomposition, ranged between 0.2 and 0.5 years during the 30-90 day period and from 2 to 4 years during the 90-700 day period. The half-life

values indicate that there are no large differences in decomposition rates for different organic materials or soil textures. Non-hydrolyzable ^{14}C i.e. soil organic matter remaining after acid hydrolyzation, was assumed to be a humified fraction, associated with the silt and clay primary particles. The curves of non-hydrolyzable ^{14}C presented in Fig 3, were considered to be essentially similar between treatments and soils. On the average of the six samplings and treatments, it can be calculated that non-hydrolyzable ^{14}C per unit of clay and silt particles was nine times more concentrated in the sand soil than in the clay soil. Such a large difference is hard to reconcile with the similar rates of decomposition of labelled residue found in Fig 1 and Table 4. Hassink *et al.* (1995) suggest that the accumulation of organic matter in soil depends on the degree of saturation of its protective sites. A possible explanation for the observed similar rates of decomposition between soils is that if the protective sites of the soils investigated here are already occupied so less soil organic matter can be stabilized physically compared with soils that have protective sites available (Hassink *et al.*, 1995). Fresh organic materials would decompose at similar rates in both soils because they are not physically protected against decomposition.

Acknowledgements - I thank H. Van Ginkel for the supply of labelled plant material, J.W. Dalenberg for technical support, M. Wolters for her assistance in the greenhouse. I also thank Ir. J. Hassink and Dr. K.B. Zwart for their useful comments and discussions on the manuscript. I especially thank Dr. L. Brussaard and Dr. A.P. Whitmore for their support, critical comments and discussions on the manuscript. Part of this work was supported by grant N° P806 under the sandwich PhD Fellowship Programme of Wageningen Agricultural University and by a grant under the Presidential Fellowship Programme of the Agencia de Cooperación Internacional (AGCI) del Ministerio de Planificación y Cooperación de Chile (MIDEPLAN).

REFERENCES

Amato, M. and Ladd, J.N. (1992) Decomposition of ^{14}C-labelled glucose and legume material in soils: properties influencing the accumulation of organic residue C and microbial biomass C. *Soil Biology & Biochemistry* **24**, 455-464.

Bek, S. (1994) Mineralization of ^{14}C-labelled unripe straw in soil with and without rape (*Brassica napus* L). *Biology and Fertility of Soils* **17**, 21-26.

Choudhri, M.B. and Stevenson, F.J. (1957) Chemical and physicochemical properties of soil humic colloids 3. Extraction of organic matter from soils. *Soil Science Society of America , Proceedings* **21**, 508-513.

Craswell, E.T. and Waring, S.A. (1972) Effect of grinding on the decomposition of soil organic matter -II. Oxygen uptake and nitrogen mineralization in virgin and cultivated soils. *Soil Biology & Biochemistry* **4**, 435-442.

Dalal, R.C. (1979) Simple procedure for the determination of total carbon and its radioactivity in soils and plant materials. *Analyst* **101**, 151-154.

Elliot, E.T. (1986) Hierarchic aggregate structure and organic C, N, and P in native and cultivated soils. *Soil Science Society of America Journal* **50**, 627-633.

Führ, F. and Sauerbeck, D. (1968) Decomposition of wheat straw in the field as influenced by cropping and rotation. In *Isotopes and Radiation in Soil Organic Matter Studies.* pp. 241-250, Proceedings IAEA/FAO Symposium, Vienna.

Gregorich, E.G., Voroney, R.P. and Kachanoski, R.G. (1991) Turnover of carbon through the microbial biomass in soils with different textures. *Soil Biology & Biochemistry* **23**, 799-805.

Hassink, J. (1992) Effects of soil texture and structure on carbon and nitrogen mineralization in grassland soils. *Biology and Fertility of Soils* **14**, 126-134.

Hassink, J., Bouwman, L.A., Zwart K.B. and Brussaard, L. (1993) Relationships between habitable pore space, soil biota and mineralization rates in grassland soils. *Soil Biology & Biochemistry* **25**, 47-55.

Hassink, J., Matus, F.J., Chenu, C. and Dalenberg, J.W. (1995) Interaction between soil biota, soil organic matter and soil structure. *Advances in Agroecology* (submitted).

Helal, H.M. and Sauerbeck D.R. (1986) Influence of plant roots on the stability of soil organic matter and soil aggregates. In *Trans XIII ISSS Congress* **3**, pp. 776-777 ISSS, Hamburg.

Jenkinson, D.S. (1977) Studies on the decomposition of plant material in soil. V. The effects of plant cover and soil type on the loss of carbon from ^{14}C labelled ryegrass decomposing under field conditions. *Journal of Soil Science* **28**, 424-434.

Jingguo, W. and Bakken, L.R. (1989) Nitrogen mineralization in rhizosphere and non-rhizosphere soil: Effect of spatial distribution of N-rich and N-poor plant residues. In: *Nitrogen in organic wastes applied to soils* (J.A. Hansen, J.A. and H. Henriksen, Eds.), Academic Press, London, pp. 81-97

Kuikman P.J., Liljeroth, E. and van Veen, J.A. (1990) Interactions between plants and soil microorganisms in the rhizosphere at different soil nutrient conditions and under predation by protozoa. *Congress of Soil Science, 14th*, Volume III, Japan. pp. 43-48.

Liljeroth, E., van Veen, J.A. and Miller, H.J. (1990) Assimilate translocation to the rhizosphere of two wheat lines and subsequent utilization by rhizosphere microorganisms at two soil nitrogen concentrations. *Soil Biology & Biochemistry* **22**, 1015-1021.

Matus, F.J. (1995) The distribution of soil organic matter of various aggregate size classes in arable soils. II. Residual organic ^{14}C, residual ^{15}N, microbial biomass ^{14}C and ^{14}C and ^{15}N mineralization rates in a sand and a clay soil. *Soil Biology & Biochemistry* (submitted).

Nowak, G. and Nowak, J. (1990) Prolonged humification of ^{14}C-labelled oat residues in extensively fertilized soils. *Soil Biology & Biochemistry* **22**, 1089-1096.

Reid, J.B. and Goss, M.J. (1982) Suppression of decomposition of ^{14}C-labelled plant roots in the presence of living roots of maize and perennial ryegrass. *Journal of Soil Science* **33**, 387-395.

Rovira, A.D. and Greacen, E.L. (1957) The effect of aggregate disruption on the activity of microorganisms in the soil. *Australian Journal of Agricultural Research* **8**, 659-673.

Sallih, Z. and Bottner, P. (1988) Effect of wheat (*Triticum aestivum*) roots on mineralization rates of soil organic matter. *Biology and Fertility of Soils* **7**, 67-70.

Sauerbeck, D.R. and Gonzalez, M.A. (1977) Field decomposition of carbon-14-labelled plant residues in various soil of the Federal Republic of Germany and Costa Rica. *Soil Organic Matter Studies.* International Atomic Energy Agency, Vienna, **1**, 159-170.

Shields, J.A. and Paul, E.A. (1973) Decomposition of ^{14}C-labelled plant material under field conditions. *Canadian Journal of Soil Science* **53**, 297-306.

Shields, J.A., Paul, E.A. and Lowe, W.E. (1973) Turnover of microbial tissue in soil under field conditions. *Soil Biology & Biochemistry* **7**, 753-764.

Sørensen, L.H. (1981) Carbon-nitrogen relationships during the humification of cellulose in soils containing different amounts of clay. *Soil Biology & Biochemistry* **13**, 331-321.

Sørensen, L.H. (1983a) The influence of stress treatments on the microbial biomass and the rate of decomposition of humified matter in soils containing different amounts of clay. *Plant and Soil* **75**, 107-119.

Sørensen, L.H. (1983b) Size and persistence of the microbial biomass formed during the humification of glucose, hemicellulose, cellulose, and straw in soils containing different amounts of clay. *Plant and Soil 75, 121-130.*

Sørensen, L.H. (1987) Organic matter and microbial biomass in a soil incubated in the field for 20 years with ^{14}C-labelled barley straw. *Soil Biology & Biochemistry* **19**, 39-42.

Van Gestel, M., Ladd, J.N. and Amato, M.(1991) Carbon and nitrogen mineralization from two soils of contrasting texture and microaggregates stability: influence of sequential fumigation, drying and storage. *Soil Biology & Biochemistry* **23**, 313-322.

Van Ginkel, J.H., Merckx, R. and van Veen, J.A. (1994) Microbial biomass method based on soluble carbon in the soil solution. *Soil Biology & Biochemistry* **26**, 417-419

CHAPTER 4

EFFECTS OF SOIL TEXTURE, SOIL STRUCTURE AND CROPPING ON THE DECOMPOSITION OF CROP RESIDUES. II. RESIDUAL ORGANIC N, MICROBIAL BIOMASS N AND INORGANIC N DERIVED FROM DECOMPOSING ^{15}N-LABELLED WHEAT AND CLOVER IN A CLAY AND SAND SOIL

F.J. Matus

DLO Research Institute for Agrobiology and Soil Fertility, P.O. Box 129, 9750 AC Haren, The Netherlands

Summary - This study is about the effect of cropping and soil disruption on the rate of ^{15}N release from ^{15}N-labelled wheat (*Triticum aestivum* L.) and clover (*Trifolium repens* L.) and the incorporation of this ^{15}N in the microbial biomass and the subsequent stabilization of ^{15}N in a sand and in a clay soil. The plant materials were added to soils in cylinders in spring 1991 and were allowed to decompose for about 16 months under field conditions. For cropping treatments, spring wheat was used, and for soil disruption, the top soil layer (0-15 cm) was mixed at six separate occasions.

During the first 48 days of decomposition the residual organic-^{15}N derived from clover materials, containing 4.2% N, declined faster than that from wheat materials, containing 3.2% N. The difference was explained by the lower content of structural carbohydrates (cellulose, hemicellulose and lignin) found in clover than in wheat. After 16 months of decomposition about 25% of the N from clover and 33% of the N from wheat in both sand and clay soil were recovered in organic residues. The total amount of N unaccounted for was slightly larger in sand than in clay and larger in clover than in wheat. Apparently, much of the labelled N derived from the two residues was lost by leaching after 67 days of decomposition due to a heavy rainfall. The labelled ^{15}N found in shoots and roots of the spring wheat crop growing on the cylinders was similar in wheat and clover: 10% and 14%, respectively, in 1991, and about 1% for the two crops in 1992.

Cropping did not affect the organic-^{15}N remaining in soil or microbial biomass-^{15}N. The concentration of inorganic-^{15}N derived from clover and wheat were similar in cropped soils and no differences between mixing and bare treatments were observed. Apparently soil microbes in the rhizosphere preferred the ^{15}N labelled residues above root-derived material, probably because both clover and wheat had C:N ratios well below the critical level. There was no effect of mixing on organic-^{15}N, microbial biomass-^{15}N and inorganic-^{15}N. This supports the hypothesis that recent soil organic matter that is protected within soil aggregates cannot easily be exposed by soil disruption.

Soon after incorporation, residue-^{15}N was transformed to forms of stable soil organic matter. The mineralization rates of N estimated between 48 and 477 days of decomposition were similar in clay and sand soil. These mineralization rates, as well as published rates derived from several ^{15}N-labelled crop residues decomposing in a wider range of soils and published rates derived from the changes in total N during cultivation of tropical soils, fitted a general inverse relationship with the time of decomposition or cultivation. Similar decomposition rates rise the question if the protective sites in the investigated soils are saturated with soil organic matter so that recently formed organic matter cannot be physically protected.

INTRODUCTION

Controversy still exists about the effect of cropping on the rate of soil organic matter decomposition. The presence of living roots has been suggested to reduce the decomposition of ^{14}C-labelled residues (Führ and Sauerbeck, 1968; Bek, 1994). The opposite effect, however, has also been found (Sallih and Bottner, 1988). Living roots were found to lead to positive (Hart *et al.*, 1979) or negative (Huntjens, 1971) effects on net nitrogen (N) mineralization. Soil tillage is another factor affecting the decomposition of soil organic matter. Several years of cropping arable land have led to lower N mineralization rates than found in grassland soils. This may be either a consequence of a lower input of organic matter or a higher decrease of soil organic matter as a result of disruption of soil aggregate structure (Elliott, 1986) or both. Disruption of aggregates exposes previously inaccessible organic materials to microbial attack (Rovira and Greacen, 1957; Craswell and Waring, 1972; Hassink, 1992). Despite the above-mentioned effects, only small differences in the rates of soil organic matter decomposition were found in a range of soils (Matus and Rodríguez, 1994). Similar and stable mineralization rates of ^{14}C- and ^{15}N-labelled residues decomposing under both field (Ladd *et al.*, 1981) or laboratory conditions (Sørensen, 1983) have been found in a range of soils. Amato and Ladd (1992), however found that the rates of mineralization varied between soils.

The aim of a previous paper (Matus, 1995[a]) was to investigate the effect of cropping, soil disruption and soil texture on ^{14}C-labelled wheat straw decomposition and the subsequent stabilization of newly formed soil organic matter in a clay or a sand soil. It was concluded that cropping, soil texture and soil disruption had little effect on residual ^{14}C decomposition in soil. The specific aims of the study reported here were to investigate (1) the effect of cropping, N fertilization and soil disruption on the rate of ^{15}N release from wheat (*Triticum aestivum* L.) and white clover (*Trifolium repens* L.) incorporated in a clay and sand soil, and (2) the stabilization of the residual ^{15}N and newly synthesized microbial biomass in clay and sand soil.

MATERIALS AND METHODS

Field site and soils

This study was conducted at the DLO Research Institute for Agrobiology and Soil Fertility (AB-DLO) in the northern part of The Netherlands (53°13'N lat., 6°35'E long.) over a period including the spring-summer seasons 1991 and 1992. The upper (0-15 cm) and lower layer (15-50 cm) from clay and sand soil in long-term arable use were used in the experiment. Each layer was mixed separately, sieved to 5 mm, and stored field moist at 5°C until used. Some chemical and physical properties of the topsoils are given in Table 1.

Table 1. Soil properties

Soil properties		Clay	Sand
Chemical			
Total C (%)		1.76	1.62
Total N (%)		0.19	0.11
C:N		9.30	14.70
$N\text{-}NH_4^+$ (mg kg^{-1})		2.10	0.40
$N\text{-}NO_3^-$ (mg kg^{-1})		14.50	13.50
P (mg l^{-1})		12.20	21.80
K (mg kg^{-1})		273.90	107.90
Mg (mg kg^{-1})		nd	116.00
$CaCO_3$ (%)		0.30	0.60
pH-KCl		6.98	7.22
Physical			
Particle size distribution (g kg^{-1})			
Clay	(< 2 μm) (%)	481	47
Fine silt	(2-20 μm)	289	34
Coarse silt	(20-50 μm)	174	265
Fine sand	(50-250 μm)	38	535
Coarse sand	(250-2000 μm)	18	119
Water holding capacity (%)			
-0.2 kPa		48	24
-25.0 kPa		40	21
-1555.0 kPa		22	5
Bulk density (g cm^{-3})		1.15	1.30

nd = not determined

Precipitation and soil temperature

Precipitation and soil temperature were registered during the experimental period. Soil temperature was measured automatically at 7.5 cm and 15 cm depth.

Preparation of labelled ^{15}N-plant material

The plant material used as organic N sources were prepared by growing wheat (*Triticum aestivum* L.) and white clover (*Trifolium repens* L.) in a greenhouse. In January 1991, 200 pots were filled with 15 kg sand soil and watered automatically with a nutrient solution containing (g per pot) 2.1 g P as $Ca(H_2PO_4)_2.H_2O$, 2.8 g K as K_2SO_4, 1.0 g Mg as $MgSO_4.7H_2O$, 0.03 g Cu as $CuSO_4.5H_2O$, 0.2 g Fe as $FeEDDHA_{(Fe\ 138)}$, 0.05 g Zn as $ZnSO_4.7H_2O$, 0.3 g Mn as $MnSO_4.7H_2O$, 0.005 g Mo as $Na_2MoO_4.2H_2O$ and 0.05 g B as $Na_2B_4O_7.10H_2O$. White clover and spring wheat were sown in 100 pots each at a sowing rate of 0.5 g and 4 g per pot, respectively. Labelled N, 10.4 g per pot of $(^{15}NH_4)_2SO_4$ (10.2 atom% excess) was supplied 7 times at 4-day intervals (Amato *et al.*, 1987). Plants, shoot and roots were harvested 11 weeks after sowing and roots were washed free of soil. All materials were dried for 48 h at 60°C and ground (particle size < 2 mm); the chemical properties are given in Table 2.

Table 2. Chemical properties of plant materials incorporated in April 1991

Plant material	Enrichment ^{15}N (atom %)	C	N	Cellulose	Hemicellulose	Lignin	Cellular content[1]	Water soluble C[2]	Ash
				(% dry matter)					
Wheat shoot	8.523	35.1	3.6	22.1	14.5	3.7	46.2	11.1	13.4
Wheat root	7.310	32.8	1.8	32.4	27.3	4.0	21.1	2.7	15.2
Clover shoot	7.588	33.3	4.4	19.0	1.5	3.3	62.7	11.3	14.4
Clover root	6.669	32.0	3.5	27.3	4.1	6.9	45.3	5.3	16.4

[1] Obtained by difference
[2] Extracted in $CaCl_2$

Soil treatments, ^{15}N-labelled crop residue incorporation

^{15}N labelled clover or wheat were incorporated in clay or sand soil with or without N fertilization (150 kg ha^{-1}). The soils were confined in PVC cylinders, 20 cm in diameter and 30 or 50 cm in length. The soil within the cylinders was either kept bare, mixed or cropped. The 30-cm cylinders were used for the bare and mixing treatment and the 50-cm cylinders for the cropping treatment to allow satisfactory root growth. Due to practical limitations, clover plus N for bare and mixing was not included as treatment. Three replicate cylinders were used per treatment. Each cylinder

was closed at the bottom with a nylon mesh. In April 1991 the cylinders were filled with a weighed portion of the subsoil (15-50 cm) from clay or sand up to 18 cm below the rim of each cylinder. Other weighed portions of topsoil (0-15 cm) from clay or sand were thoroughly mixed with ^{15}N-labelled clover or wheat residues before placement into the cylinders and gently compressed. The amount of crop residue added per cylinder was a mixture of 12 g shoot and 3 g root material (on dry matter basis). The C:N ratio of the mixture was 11 for wheat and 8 for clover (Table 2). The cylinders were transferred to the field and placed in 60 concrete microplots of 1 x 1 x 1 m, previously filled with similar subsoil of clay or sand as used in the cylinders. The cylinders were randomized, arranged in a grid pattern and the remaining empty portions of the microplots were filled with similar topsoil of clay or sand as used in the cylinders. For the crop treatments spring wheat (*Triticum aestivum* L.) was used. On 29 April, 1991 and on 15 April 1992, microplots and cylinders were sown at a rate of 10 seeds per cylinder (102 kg ha^{-1}). Spring wheat was harvested in September 1991 and August 1992. The mixing treatment consisted of disruption of the soil aggregates to < 0.5 cm by hand in the top soil (0-15 cm). Mixing was performed six times: 25, 67, and 103 days after sowing in 1991 and 0, 41 and 104 days after sowing in 1992. On day 0, mixing and sowing of spring wheat coincided. Soils in the bare treatments were conserved undisturbed, and the small amount of spontaneous vegetation growing in the cylinders was cut and left on top of the soil within the cylinders. The cylinders were protected from splashing of the soil caused by heavy rainfall with a plastic shield around the internal edge of the cylinders.

Mineral fertilization

In April 1991 and 1992 the treatments were amended with a solution containing 0.31 g N as NH_4NO_3 per cylinder, equivalent to 100 kg N ha^{-1}. All cylinders were amended with a base nutrient solution containing per cylinder: 0.12 g P and 0.31 g K as K_2HPO_4 and 0.10 g Mg as $MgSO_4.7H_2O$, equivalent to 40 (P), 100 (K) and 80 (Mg) kg ha^{-1}. A second N fertilization containing 0.16 g N $cylinder^{-1}$ as NH_4NO_3 in solution, equivalent to 50 kg N ha^{-1}, was applied after 52 days of sowing.

Sampling

Soil was sampled six times during the experimental period. Samples were taken 48, 67, 135, 393, 420 and 477 days after ^{15}N-labelled residue incorporation. Sampling on days 135 and 477 coincided with the harvest of the spring wheat. At each sampling and for each treatment three cylinders were removed. The top layer of soil (0-15 cm) of each cylinder was carefully separated from the bottom layer of soil (15-27 cm of the bare and mixing treatments and 15-47 cm of the cropping treatment). Each layer was mixed separately, sieved through a 0.005-m screen and a subsample of about 2 kg was taken and stored at 5°C for analysis. At each sampling, the holes left by the

removed cylinders were filled with clay or sand soil similar to the soil used in the cylinders. In the cropping treatment, shoots and roots were harvested. Roots were collected from two replicate cylinders. The cylinders were longitudinally cut in half and the soil from each portion was weighed. Roots and soil from one portion were separated by repeated washing through a 2-mm sieve. The other portion was mixed, sieved and sampled for analysis. Shoots and roots were dried at 60°C for 48 h and stored for analysis.

Analytical procedures

Total N and mineral N. Total N (including inorganic NO_3^--N plus NH_4^+-N) was determined by oxidation with sulphuric acid and salicylic acid and the N distilled according to Deijs (1961). Inorganic N in soil was measured colorimetrically after extraction with 1 M KCl solution for 1 h (soil:water, 1:2.5) using a Technicon autoanalyser (Traacs 800).

Total organic C, total N and carbohydrate composition in plant materials. Total organic-C was determined by a concentrated acid dichromate-oxidizable organic matter solution according to Mebius (1960). Structural carbohydrate content in the labelled ^{15}N plant material, cellulose, hemicellulose and lignin, were determined as neutral detergent fibre (NDF). Cellulose and lignin were determined as acid detergent fibre (ADF) and lignin as acid detergent lignin (ADL) (Van Soest, 1967). Water soluble C was determined in filtrate after extraction in $CaCl_2$ for 1 hour. The organic C rendered extractable was measured by dry oxidation in a TOC-500, Shimadzu analyzer.

Microbial biomass N. At each sampling time, microbial biomass-N was measured in the top-soil (0-15 cm) by the fumigation-extraction method (Brookes *et al.*, 1985). Total N in the extract was analyzed after Kjeldahl digestion; ^{15}N was determined in the same way as described for inorganic-N (see below). The amounts of ^{15}N in the microbial biomass were calculated as the difference between the amount of ^{15}N present in fumigated extracts minus the amount of ^{15}N in non-fumigated extracts. The conversion factor used for N (k_n) was 0.54 (Brookes *et al.*, 1985).

^{15}N determination. The ^{15}N concentration in the soil and plant materials was determined using a Carlo Erba NA 1500 nitrogen analyzer linked to a SIRA 10 V. G. Isogas micromass spectrometer. The ^{15}N in inorganic-N extracts was determined after steam-distillation with $FeSO_4$ and Ag_2SO_4. The distillate was collected in dilute boric acid, neutralized with sulphuric acid and taken to dryness in a hot block. The NH_4^+-N was converted to N_2 by reaction with alkaline lithium hypobromite at the inlet to the mass spectrometer and analyzed for the isotopic ratio of ^{15}N:^{14}N. All ^{15}N determinations were corrected for soil background, 0.373 atom%. Nitrogen in the soils was expressed on an oven-dry basis (105°C).

Calculation methods

Residual organic-^{15}N was calculated as the difference of total-^{15}N minus inorganic-^{15}N (NH_4^+-N and NO_3^--N) in soil and the total-^{15}N of roots. Inorganic-^{15}N concentration from cropped soils was calculated as the sum of the inorganic-^{15}N concentration in soil plus total-^{15}N of shoots and roots (assumed to have originated from inorganic-^{15}N taken up by crop).

Statistical analysis

Analysis of variance (ANOVA; Genstat 5.0), and Student's t-test were used to analyze the differences of means between treatments. The probability at which significant differences were compared was set at the 5%.

RESULTS

Total recovery of ^{15}N

^{15}N recovered in different pools of soil N. The total recovery of ^{15}N derived from clover and wheat was similar in all treatments. Table 3 presents an example of the total amounts of ^{15}N recovered from cropped soil and its distribution in different N pools in soil in 1991 and 1992. An important proportion of labelled N derived from the two residues was retained in both soils as non-biomass. The proportion of ^{15}N in microbial biomass was found to be similar in both soils, and also for clover and wheat residues. The uptake of ^{15}N by the wheat and clover plants at harvest was small: about 10% and 14% for wheat and clover in 1991, respectively and about 1% for both plant materials in 1992.

^{15}N unaccounted for. The ^{15}N unaccounted for increased in time and was higher for the clover than for the wheat residue, whereas significant differences between sand and clay soils were established only occasionally, with more ^{15}N unaccounted for in sand than in clay (Table 4). The largest differences between treatments were observed after 48 days of decomposition for wheat, and for clover, after 393 days.

Nitrogen transported to 15-27 cm or 15-47 cm layers. Inorganic-^{15}N and organic-^{15}N derived from clover and wheat residues were transported deeper than 15 cm. Between 5% and 25% of residue-^{15}N derived from clover was found as inorganic-N in both soils, and between 1% and 8% as organic-N, after 48 days of decomposition (Table 5).

Table 3. Total recovery of ^{15}N (% of input ^{15}N) derived from clover and wheat residue for non N-fertilized cropping in sand (S) and clay (C) soils in 1991 and 1992 in the field

Sampling Time (days)	Extrac. Inorg.^{15}N (0-47 cm)		Non-biomass-^{15}N (0-47 cm)		Microbial biomass-^{15}N (0-15 cm)		Crop[1]		Total	
	S	C	S	C	S	C	S	C	C	S
Clover										
					1991					
48	7	* 14	38	38	11	10	5	6	61	68
67	1	1	28	24	16	12	9	13	54	50
135	0	0	19	* 48	6	5	14	15	39	68
					1992					
393	0	0	24	30	5	5	1	1	30	36
420	nd	nd	19	25	6	6	1	1	26	32
477	0	0	18	20	6	6	1	2	25	28
Wheat										
					1991					
48	5	4	54	57	16	13	3	5	78	79
67	1	1	37	45	22	* 14	8	9	68	69
135	0	1	52	* 70	5	2	10	12	67	85
					1992					
393	0	0	28	* 42	7	7	1	1	36	50
420	nd	nd	25	* 35	7	8	1	2	33	45
477	1	0	24	* 34	9	9	1	2	35	45

[1] shoots and roots
L.S.D.[2] inorganic N = 2
L.S.D. non biomass N = 9
L.S.D. microbial biomass N = 7
[2] least significant differences ($P < 0.05$)
* = significant differences between soils
nd = not determined

These figures for wheat-N ranged from 3% to 15% for both inorganic and organic-N. Particularly in bare and mixing soils, more leached organic-^{15}N was generally found in the clay than in the sand soil and more from wheat than clover. Organic-^{15}N decreased with the samplings, but some values remained higher, even as decomposition progressed.

Residual organic-^{15}N

Residue type. The proportion of labelled organic-^{15}N (0-15 cm) was found to decline in two phases: a rapid decomposition during the first 48 days of incorporation and a slow phase during days 48 to 477 days of decomposition, (Fig. 1). The recovery of ^{15}N on day 0 was assumed 100%.

Table 4. Total ^{15}N unaccounted for (% of input ^{15}N) in non N-fertilized cropping, bare and mixing treatments in sand (S) and clay (C) soil during 1991 and 1992 in the field

	Cropping (0-47 cm)			Bare (0-27 cm)			Mixing (0-27 cm)		
	S		C	S		C	S		C
Clover									
					1991				
48	39		32	18		16	19	*	2
67	46		50	51		49	49		45
135	51	*	32	47		42	37		46
					1992				
393	70		64	73		68	73	*	8
420	74		68	74		69	76		72
477	75		72	77		75	76		65
Wheat									
					1991				
48	22		21	7		0	5		0
67	32		31	27		24	25		25
135	33	*	15	31	*	15	23		19
					1992				
393	64	*	50	61		58	61		56
420	67		55	64	*	50	67		60
477	66		55	69		61	68		61

L.S.D.[1] = 13
[1] least significant differences ($P < 0.05$)
* = significant differences between soils

Rapid decline (0-48 days): significantly ($P < 0.05$) more clover decomposed than wheat. About 40%-55% derived from the clover and 65%-80% from wheat residue were present in organic form after 48 days of incorporation (Fig. 1).

Slow decline (67-477 days): the initial differences were maintained during the following days of decomposition; the organic-^{15}N curves for both clover and wheat declined nearly at the same rate. The mineralization rate constants between 48 and 477 days of decomposition, assuming first order kinetics, were slightly lower for clover, $k = 0.0018$ day^{-1} ($T_{1/2} = 385$ days) than for wheat, $k = 0.0020$ day^{-1} ($T_{1/2} = 346$ days), but no significant differences were observed (Table 6). After 477 days of decomposition, the amount of labelled N retained in organic forms ranged, from 15% to 35% for clover, and from 20% to 45% for wheat (Fig. 1).

Table 5. Transport of ^{15}N derived (% of input ^{15}N) from clover and wheat residue to the 15-47 cm layer in the cropping treatment and 15-27 cm in the bare and mixing treatments in sand (S) and clay (C) soil during 1991 and 1992 in the field

Sampling time (days)	Cropping				Bare				Mixing			
	Inorg.^{15}N		Org.^{15}N		Inorg.^{15}N		Org.^{15}N		Inorg.^{15}N		Org.^{15}N	
	S	C	S	C	S	C	S	C	S	C	S	C
Clover												
					1991							
48	5 *	11	2	1	18 *	13	3	5	15 *	25	4 *	8
67	0	1	3	3	0	1	2 *	6	0	4	5	7
135	nd	0	2	4	1 *	3	4 *	8	2 *	4	15 *	7
					1992							
393	0	0	2	3	0	0	0	3	0	0	2 *	6
420	nd	nd	1	1	0	0	2	3	0	0	2	3
477	0	nd	1	1	1	1	1	1	1	1	3 *	10
Wheat												
					1991							
48	3	3	3	5	9 *	13	3	3	9 *	15	9 *	15
67	0	1	2	5	0	1	5 *	11	0	1	6 *	10
135	nd	0	2 *	10	2	2	3 *	12	2	2	7 *	12
					1992							
393	0	0	2	2	0	0	1	4	0	0	3	5
420	nd	nd	1	2	0	0	2 *	12	0	0	2	4
477	0	0	2	3	1	1	1	4	1	1	1	3

L.S.D.[1] inorganic-^{15}N = 2
L.S.D. organic-^{15}N = 4
[1] least significant differences ($P < 0.05$)
* = significant differences between soils
nd = not determined

Cropping and mixing. The residual organic-^{15}N in sand soil followed a smooth curve throughout its decomposition (Fig. 1a and c), while in clay soils residual organic ^{15}N in the cropping treatment increased significantly ($P < 0.05$) after 135 days of incorporation (Fig. 1b and d). In spite of these differences the curves are regarded as essentially similar in both treatments, because the effect was not sustained. No effect of mixing was found. After 477 days of decomposition, after mixing the soil six times, no significant differences were established between mixed and bare soils (Fig. 1).

Nitrogen fertilization. Nitrogen fertilization had no significant effect on the decomposition of clover or wheat residue (Fig. 1c and d).

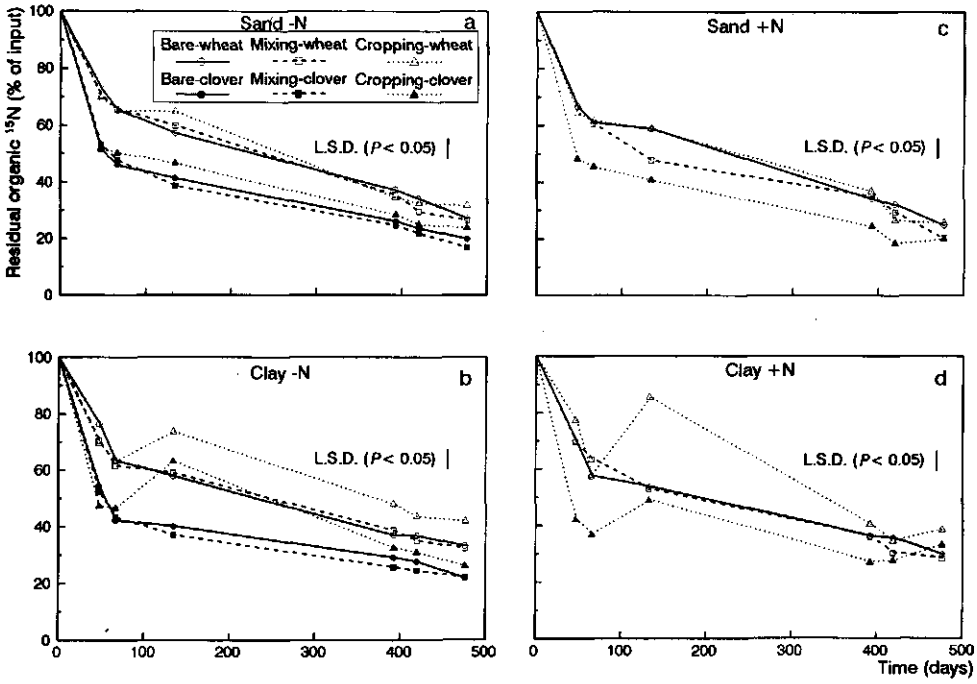

Fig. 1. Residual organic ^{15}N (0-15 cm) from clover and wheat residues incorporated without N fertilization in sand (a) and clay (b) and with N fertilization in sand (c) and clay (d) and allowed to decompose for 477 days (mixing on day 25, 67, 103, 351, 392 and 455).

Microbial biomass-^{15}N and inorganic-^{15}N

Residue type. No significant differences in microbial biomass-^{15}N were found (Fig. 2). Microbial biomass-^{15}N derived from clover tended to be lower than from wheat, however. After 48 days of decomposition (Fig. 2d) or 67 days (Fig. 2a-c) microbial biomass was found to be highest. The maximum values for residue N present in microbial biomass were about 18% for both clover and wheat. One immobilization-mineralization cycle may have occurred earlier, since a high concentration of inorganic-^{15}N was observed, in bare and mixing treatments, on day 48 (Fig. 4). Microbial biomass declined significantly ($P < 0.05$) from day 67 to 135 of decomposition. During the following samplings, this value remained more or less constant. Inorganic-^{15}N concentrations in cropped soil are presented in Fig. 3 and in the bare and mixing treatments in Fig. 4. Since inorganic-^{15}N from the cropping treatments was estimated as the total amount of ^{15}N contained in roots (0-47 cm) and shoots plus the inorganic-N concentration in the topsoil (0-15 cm), and the inorganic-N in the bare and mixing treatments was calculated in the topsoil only, comparisons

Table 6. Mineralization rate constants (day^{-1}) of residual organic ^{15}N (0-15 cm) from wheat and clover residue in sand and clay soil

Treatments	Sand	Clay
Wheat without N		
Bare	0.0020	0.0018
Mixing	0.0022	0.0017
Cropping	0.0020	0.0012
Clover without N		
Bare	0.0020	0.0017
Mixing	0.0024	0.0018
Cropping	0.0019	0.0015
Wheat with N		
Bare	0.0021	0.0017
Mixing	0.0023	0.0020
Cropping	0.0022	0.0018
Clover with N		
Cropping	0.0022	0.0009
Mean	0.0021	0.0016

between cropping and bare or mixing have not been made. Fig. 3 shows that inorganic-^{15}N from cropped soils increased as a result of mineralization; the increase was measured as the sum of inorganic N in soil and crop uptake. After 135 days of decomposition, N derived from clover was significantly higher than from wheat, except in sand soil (Fig. 3c); thereafter, no significant differences were observed. Fig (4a and b) shows that inorganic-^{15}N derived from clover in bare and mixing treatments, was significantly ($P < 0.05$) higher than from wheat after 48 days of decomposition. Between 48 and 67 days the inorganic-N concentration decreased sharply, coinciding with the observed strong increase in microbial biomass (Fig. 2) which also took place in the control (without N-labelled residue addition, data not shown). Thereafter, a significant increase in inorganic-^{15}N took place (Fig. 4). After 135 days of decomposition, the inorganic-N concentration decreased to values lower than 5% of the applied label.

Although differences between microbial biomass-^{15}N derived from clover and wheat residue were not found at any single sampling, the overall average of the two soils and sampling times was significantly ($P < 0,05$) lower for the clover than for the wheat residue. Conversely, the overall average inorganic-N levels were higher in the clover than in the wheat residue treatment (Table 7).

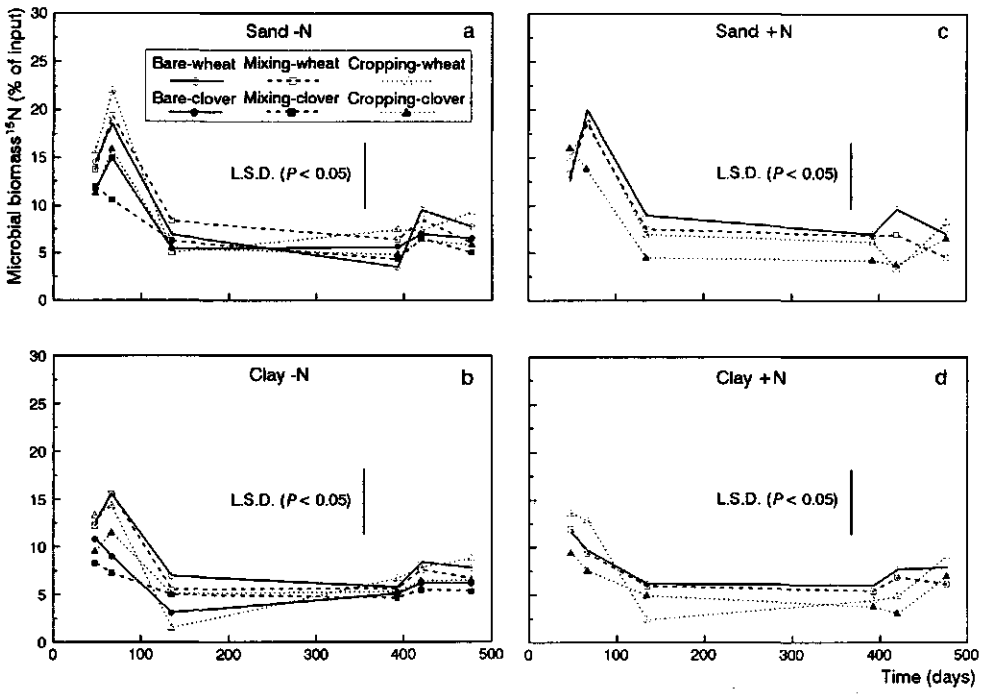

Fig. 2. ^{15}N incorporated in microbial biomass (0-15 cm) during decomposition without N fertilization in sand (a) and clay (b) and with N fertilization in sand (c) and clay (d).

Cropping and mixing. Cropping and mixing did not affect the labelled microbial biomass-N concentration at any time during decomposition (Fig. 2). Significant differences in inorganic-^{15}N concentration from mixed and bare clay soils without N and with fertilization were only after 48 days of incorporation, observed (Fig. 4b and d). This effect, however, disappeared as decomposition advanced.

Nitrogen fertilization. Nitrogen fertilization had no effect on microbial biomass-^{15}N (cf. Figs. 2a and b with Figs. 2c and d). No significant effects of N fertilization were observed on the inorganic-^{15}N concentration in the cropping treatment (cf. Figs. 3a and b with Figs. 3c and d), or in the bare or mixing treatments (cf. Figs. 4a and b with 4c and d).

Effect of soil texture on residual organic-^{15}N and microbial biomass-^{15}N

The mineralization rate constants of residual organic ^{15}N estimated between 48 and 477 days of decomposition from both, clay and sand soil (Table 6) and the mineralization rates obtained from

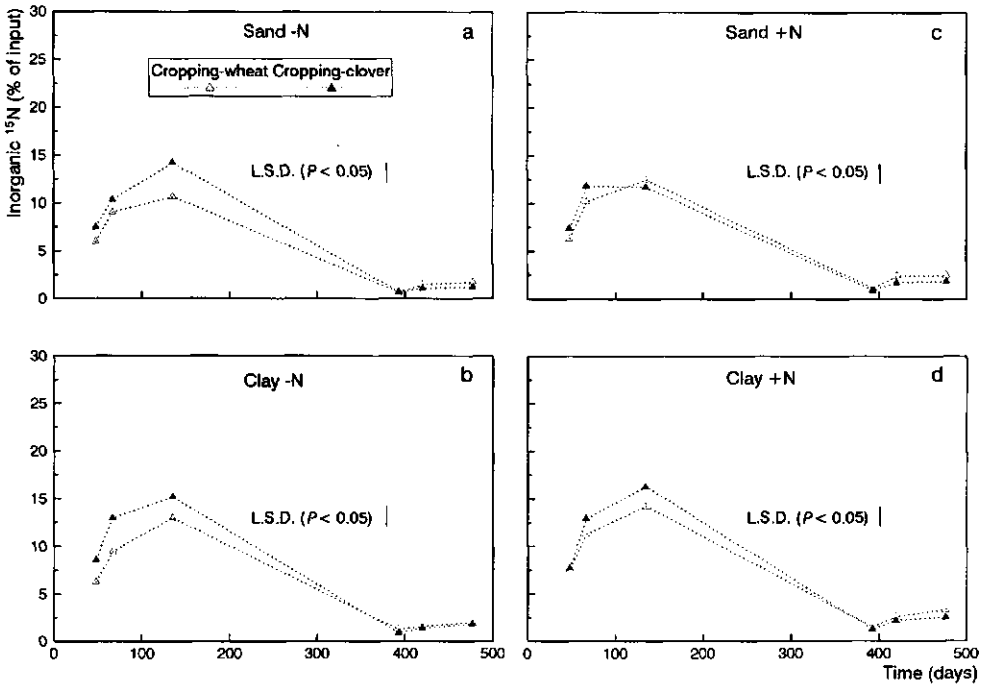

Fig. 3. Inorganic ^{15}N (NO_3^--N plus NH_4^+-N) from cropped soils without N fertilization in sand (a) and clay (b) and with N fertilization in sand (c) and clay (d).

the literature from several ^{15}N-labelled residues in different soils and the rates reported from the total N changes during cropping of some tropical soils are presented in Fig. 5. The rates were obtained from periods of decomposition or cropping longer than one year. All rates were adjusted to a temperature of 16°C (Matus and Rodríguez, 1994). After one year the rates decreased sharply. Apparently, the rates are independent of soil texture and residue type incorporated in the soil.

Microbial biomass-^{15}N values were very similar in clay and sand soil and no significant effect of soil texture was found.

DISCUSSION

Recovery of ^{15}N

The total recovery of ^{15}N showed that up to 20% and 40% of the wheat and clover residue, respectively, were unaccounted for after 48 days of decomposition in the cropping, bare and

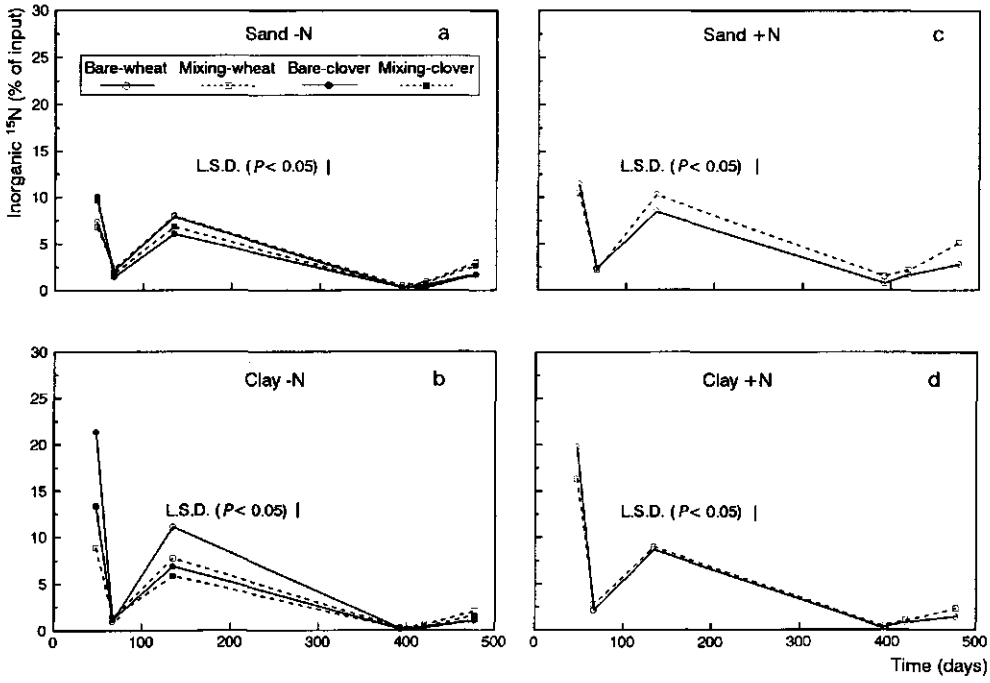

Fig. 4. Inorganic ^{15}N (NO_3^--N and NH_4^+-N) from bare and mixing treatments without N fertilization in sand (a) and clay (b) and with N fertilization in sand (c) and clay (d).

Table 7. Average of microbial biomass ^{15}N and inorganic ^{15}N (0-15 cm) on soils and samplings time (% of input ^{15}N)

Treatments	Wheat residue		Clover residue	
	M.biomass-^{15}N	Inorg.^{15}N	M.biomass-^{15}N	Inorg.^{15}N
With N				
Bare	9.9	3.9	7.6	4.2
Mixing	9.6	3.5	6.7	3.7
Cropping	9.9	5.3	8.2	6.3
Without N				
Bare	9.5	5.0	na	na
Mixing	8.6	5.3	na	na
Cropping	8.8	6.3	7.0	6.5

L.S.D.[1] microbial biomass N = 1.9
L.S.D. inorganic N = 0.5
[1] least significant differences ($P < 0.05$)
* = significant differences between soils
na = not applicable

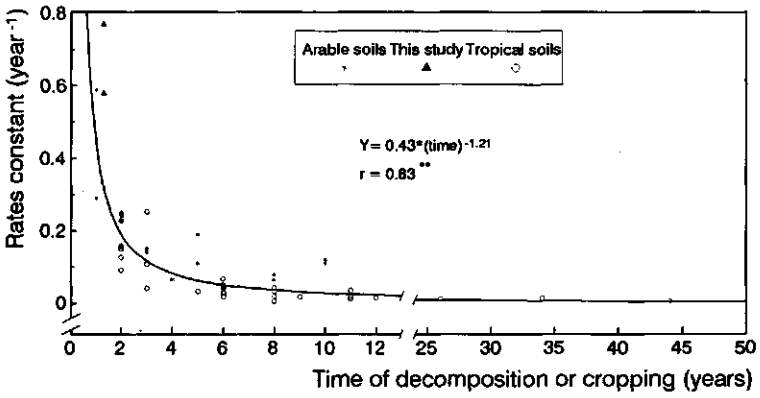

Fig. 5. Nitrogen mineralization rate constants from different ^{15}N-labelled crop residues in arable soils (Matus and Rodríguez, 1994; Jensen, 1994) and some tropical soils (Bartholomew, 1977) and a Indian soil (Gokhale, 1959).

mixing treatments (Table 4). This N may have been lost by leaching, denitrification or volatilization (recovery of ^{15}N at time 0 was assumed to be 100%) The low concentration of inorganic-^{15}N from the bare and mixing treatments after 67 days of decomposition (Fig. 4) was partly explained by leaching of mineral N and partly by immobilization when this effect was examined in a computer simulation model (Whitmore and Matus, 1995). Between 48 and 67 days of decomposition the precipitation recorded at the experimental site was 121 mm as compared with the long-term average of 42 mm (mean, 1951-1980). Soluble organic N loss by leaching below 47 cm from cropped soil and 27 cm from the mixing or bare treatments can neither be excluded. Both organic and inorganic-^{15}N were present in deeper layers throughout the six samplings (Table 5). A complete separation of the topsoil layer (0-15 cm) from the 15-27 or 15-47 cm subsoil layer at each sampling was not possible, and the subsoil layer may therefore have been contaminated with topsoil (Jensen, 1994). High microbial activity in the top soil and transport by faunal activity may also have moved organic N to deeper layers. Water-soluble organic N from both clover and wheat residue could have contributed as well.

Effect of residue type

After 67 days of decomposition organic-^{15}N derived from clover ranged from 35% to 50% of added ^{15}N, these values being significantly ($P < 0.05$) lower than those for wheat, 55% and 70% (Fig. 1). The mixture of the ^{15}N-labelled plant material incorporated in soil, shoots and roots, had a C:N ratio of 8 (clover) and 11 (wheat). The classical N-mineralization-immobilization concept

predicts that decomposition of organic materials with a higher C:N ratio can be retarded because soil microbes do not find their requirement for N (Jansson, 1958). The critical N concentration in crop residues at which N-immobilization may occur has been found to be less than 1% (Jensen, 1929). This threshold value is far lower than the N contents in the materials used in the present experiment. Therefore, immobilization of N or any retardation of the decomposition was not expected (Nordmayer and Richter, 1985). Fig. 1 shows that there were no significant differences between soils with and without N fertilization. This suggests that during the first 48 days of decomposition clover residues mineralized faster than wheat residues due to the differences in the chemical composition rather than differences in N concentration. The proportions of structural carbohydrates, cellulose, hemicellulose and lignin of the two crop residues are shown in Table 2. On the basis of NDF (neutral detergent fibre) a distinction can be made between readily decomposable organic compounds (cellular content) and structural carbohydrates (mostly cell wall constituents). The cellular content in clover was considerably higher (60%) than in wheat (40%). This differences may have determined the observed faster N-mineralization from clover than from wheat. These results can also explain the higher ^{15}N unaccounted for in clover than in wheat residue after 67 days of decomposition (Table 4).

Effect of cropping

In the present study living roots did not decrease or increase the residual ^{15}N compared with uncropped bare and mixed soils, except in the cropped treatment on day 135 in the clay soil. This increase was partially attributable to sampling error because the coefficient of variation of this treatment ranged from 9% to 25% compared with bare and mixing treatments with less variation, between 1% and 9%. A low recovery of ^{15}N from roots (roots were subtracted from the total ^{15}N) was unlikely, because the total amount of root materials in clay and sand soils was similar and had a comparable ^{15}N concentration (data not shown). Consequently, the increase on day 135 can be disregarded because no similar effect was found in the sand soil and the increase was not sustained in the following samplings or in other treatments (Fig. 1). These results support the observation that living roots do not affect mineralization from fresh plant residues rich in N (Jingguo and Bakken, 1989). It seems that the presence of plants, rather than stimulating N mineralization, reduces microbial N immobilization through effective competition for mineral N (Jingguo and Bakken, 1989). It has been suggested that with high N concentration in soils, microbes prefer root derived material over native soil organic matter and that at low N concentration this preferences does not occur (Liljeroth *et al.*, 1990; Kuikman *et al.*, 1990). In the present investigation labelled microbial biomass in cropped soils was not significantly different from that in bare and mixing treatments, indicating that the residual ^{15}N from both, clover and wheat, was decomposed to the same extent in all three treatments.

Effect of mixing

It is possible that tillage of virgin soil causes disintegration of macro-aggregates (> 250 μm) into micro-aggregates (< 250 μm) and that micro-aggregates are relatively unaffected (Elliott, 1986). It is also possible that after long-term land cultivation the macro-aggregates remaining in the soil are resistent to disintegration by further cultivation. So, soil organic matter recently formed within macro- and micro-aggregates cannot be easily exposed to biodegradation by soil disaggregation (Matus, 1995[b]). Microbial metabolites and cellular materials may be largely protected against biodegradation within soil aggregates which cannot be easily disrupted by simple mixing. No effect of mixing on residual ^{15}N, microbial biomass-^{15}N and inorganic-^{15}N was observed in this experiment. Similar results, using ^{14}C labelled residues on the same soil, were reported by Matus (1995[a]).

Effect of clay and sand soil on residual ^{15}N and microbial biomass

The mineralization rate constants of N calculated for the net decline of organic ^{15}N between 48 and 477 days of decomposition were considered to represent the transformation of clover and wheat residue N into stable soil organic matter forms. The mineralization rate constants, 0.0021 day^{-1} in the sand and 0.0016 day^{-1} in the clay soil, were nearly similar, regardless of the considerable differences in silt and clay content between the two soils (Table 1). These values were also similar to the decay rates, 0.0015 day^{-1}, obtained from clay and sand soil amended with ^{14}C-labelled residues (Matus, 1995[a]). Matus and Rodríguez (1994) noted that the mineralization rate constants derived from several ^{15}N-labelled residues decomposing in different soils, when adjusted to the same temperature, were very similar for periods of decomposition longer than one year. An inverse relationship was found between the time of decomposition and the rate constants estimated here and those summarized by Matus and Rodríguez (1994), as well as the rates obtained by Jensen (1994) and others, estimated from the changes in total N during cultivation, including some tropical soils (Bartholomew, 1977; Gokhale, 1959) (Fig. 5). This illustrates that the rate constants fit a general curve, whatever crop residues or soil textures are included. Crop residues consist of both a readily decomposable and a resistant organic fraction. After one year, most of the readily decomposable materials are decomposed and the resistant fraction and microbial products are stabilized in soil (Matus and Rodríguez, 1994). The decomposition rate constants of organic matter in arable soils in a certain period of decomposition were equivalent to the rates in tropical soils for the same period of cultivation. Hassink *et al.*, (1995) suggested that the accumulation of soil organic matter in soil depends on the degree of saturation of its protective sites. If these protective sites are occupied, less soil

organic matter will be accumulated than in soils with protective sites available. Therefore, recently formed soil organic matter will decompose at similar rates whenever it is not protected from biodegradation. This was also consistent with similar values of microbial biomass ^{15}N between clay and sand soil, observed. Matus (1995[a]) showed similar residual ^{14}C decomposition rates and microbial biomass ^{14}C between the soils studied here. Similar decomposition rates rise the question if the protective sites in the investigated soils are saturated with soil organic matter so that recently formed organic matter cannot be physically protected.

Acknowledgements - I thank Mr. F.H. de Vries and Mr. W. Willems for their full collaboration and help with the experimental work. I also thank Ir. J. Hassink, Dr. K. B. Zwart and Dr. P. de Willigen for their comments on the manuscript. I am especially grateful to Dr. L. Brussaard and Dr. A.P. Whitmore for their support and helpful discussions throughout this investigation and final comments on the manuscript. Part of this work was supported by grant N° P806 under the sandwich PhD fellowship programme of the Wageningen Agricultural University and by the grant under the Presidential fellowship programme of Agencia de Cooperación Internacional (AGCI) del Ministerio de Planificación y Cooperación de Chile (MIDEPLAN).

REFERENCES

Amato M. and Ladd J.N. (1992) Decomposition of ^{14}C-labelled glucose and legume material in soils: properties influencing the accumulation of organic residue C and microbial biomass C. *Soil Biology & Biochemistry* **24**, 455-464.

Amato, M., Ladd J.N., Ellington, A., Ford, G., Mahoney J.E., Taylor A.C and Walsgott D. (1987) Decomposition of plant material in Australian soils. IV. Decomposition in situ of ^{14}C- and ^{15}N labelled legume and wheat materials in a range of southern Australian soils. *Australian Journal of Soil Research* **25**, 95-105.

Bartholomew, W.V. (1977) Soil nitrogen changes in farming systems in the humid tropics In *Biological Nitrogen Fixation in Farming Systems of the Tropics* (A. Ayanaba and P.J. Dart, Eds), pp. 27-44 *Chichester and Wiley.*

Bek, S. (1994) Mineralization of ^{14}C-labelled unripe straw in soil with and without rape (*Brassica napus* L). *Biology and Fertility of Soils* **17**, 21-26.

Brookes, P.C., Landman A., Pruden G. and Jenkinson D.S. (1985) Chloroform fumigation and the release of soil nitrogen: A rapid direct extraction method to measure microbial biomass nitrogen in soil. *Soil Biology & Biochemistry* **17**, 837-842.

Craswell, E.T and Waring, S.A. (1972) Effect of grinding on the decomposition of soil organic matter-I. The mineralization of organic nitrogen in relation to soil type. *Soil Biology & Biochemistry* **4**, 427-433.

Deijs, W.B. (1961) The determination of total nitrogen in herbage samples (in Dutch). *Instituut voor Biologisch en Scheikundig Onderzoek van Landbouwgewassen Wageningen, Jaarboek,* **89, 158,** 90.

Elliott, E.T. (1986) Aggregate structure and carbon, nitrogen and phosphorus in native and cultivated soils. *Soil Science Society of America Journal* **50,** 627-633.

Führ F. and Sauerbeck D. (1968) Decomposition of wheat straw in the field as influenced by cropping and rotation. In *Isotopes and Radiation in Soil Organic Matter Studies, pp. 241*-250. Proceedings IAEA/FAO Symposium, Vienna.

Gokhale, N.G. (1959) *Soil nitrogen status under continuous cropping and with manuring in the* case of unshaded tea. *Soil Science* **87,** 331-333.

Hart, P.B.S., Goh, K.M, and Ludecke, T.E. (1979) Nitrogen mineralization in fallow and wheat soils under field and laboratory conditions. *New Zealand Journal Agricultural Research* **22,** 115-125.

Hassink, J. (1992) Effect of soil texture and structure on carbon and nitrogen mineralization in grassland soils. *Biology and Fertility of Soils* **14,** 126-134.

Hassink, J., F.J. Matus, C. Chenu and J.W. Dalenberg (1995) Interaction between soil biota, soil organic matter and soil structure. *Advances in Agroecology* (Submitted)

Huntjens, J.L.M. (1971) The influence of living plants on mineralization and immobilization of nitrogen. *Plant and Soil* **35,** 77-94.

Jansson, S.L. (1958) Tracer studies on nitrogen transformation in soil with special attention to mineralization-immobilization relationships. *Annals of the Royal Agricultural College Sweden* **24,** 101-361.

Jensen, E.S. (1994) Dynamics of mature pea residue nitrogen turnover in unplanted soil under field conditions. *Soil Biology & Biochemistry* **26,** 455-462.

Jensen, H.L. (1929) On the influence of carbon:nitrogen ratios of organic materials on the mineralization of nitrogen. *Journal of Agricultural Science* **19,** 71-82.

Jingguo, W. and Bakken, L.R. (1989) Nitrogen mineralization in rhizosphera and non-rhizosphera soil: Effect of spatial distribution of N-rich and N-poor plant residues. In *Nitrogen in Organic wastes applied to soils* (J.A. Hansen and H. Henriksen, Eds), pp. 81-97 *Academic Press, London.*

Kuikman P.J., Liljeroth, E. and Van Veen, J.A. (1990) Interactions between plants and soil micro-organisms in the rhizosphere at different soil nutrient conditions and under predation by protozoa. *Congress of Soil Science, 14th*, Volume III, Japan, pp. 43-48

Ladd, J.N., Oades, J. M. and Amato, M. (1981) Microbial biomass formed from ^{14}C, ^{15}N- labelled plant material decomposing in soils in the field. *Soil Biology & Biochemistry* **13,** 119-126.

Liljeroth E., Van Veen, J.A. and Miller, H.J. (1990) Assimilate translocation to the rhizosphere of two wheat lines and subsequent utilization by rhizosphere microorganisms at two soil nitrogen concentrations. *Soil Biology & Biochemistry*, **22**, 1015-1021.

Matus, F.J. (1995a) Effect of soil texture, soil structure and cropping on the decomposition of crop residues. I. Residual organic C and microbial biomass C derived from decomposing ^{14}C-labelled wheat straw in a clay and sand soil. *Soil Biology & Biochemistry* (submitted).

Matus, F.J. (1995b) The distribution of soil organic matter of various aggregate size classes in arable soil. I. Relationships between clay content of aggregates of a sand and a clay soil and carbon mineralization, nitrogen mineralization and microbial biomass carbon. *Soil Biology & Biochemistry* (submitted).

Matus, F.J. and Rodríguez, J. (1994) A simple model for estimating the contribution of nitrogen mineralization to the nitrogen supply of crops from a stabilized pool of soil organic matter and recent organic input. *Plant and Soil* **162**, 259-271.

Mebius, L.J. (1960) A rapid method for the determination of organic carbon in soil. *Analytica Chemica Acta* **22**, 120-124.

Nordmeyer, H. and Richter, J. (1985) Incubation experiments on N mineralization in loess and sandy soil. *Plant and Soil* **83**, 433-445.

Rovira, A.D. and Greacen, E.L. (1957) The effect of aggregate disruption on the activity of microorganisms in the soil. *Australian Journal Agricultural Research* **8**, 659-673.

Sallih, Z. and Bottner, P. (1988) Effect of wheat (*Triticum aestivum*) roots on mineralization rates of soil organic matter. *Biology and Fertility of Soils* **7**, 67-70.

Sørensen, L.H. (1983) Size and persistence of the microbial biomass formed during the humification of glucose, hemicellulose, cellulose, and straw in soils containing different amounts of clay. *Plant and Soil* **75**, 121-130.

Van Soest, P.J. (1967) Development of a comprehensive system of field analysis and its application to forages. *Journal of Animal Science* **26**, 119-132.

Whitmore, A.P. and Matus, F.J. (1995). The decomposition of wheat and clover residues in soil: measurements and modelling. *Plant and Soil* (submitted).

CHAPTER 5

THE DISTRIBUTION OF SOIL ORGANIC MATTER OF VARIOUS AGGREGATE SIZE CLASSES IN ARABLE SOILS. I. RELATIONSHIPS BETWEEN CLAY CONTENT OF AGGREGATES OF A SAND AND A CLAY SOIL AND CARBON MINERALIZATION, NITROGEN MINERALIZATION AND MICROBIAL BIOMASS CARBON

F.J. Matus

DLO Research Institute for Agrobiology and Soil Fertility, P.O. Box 129, 9750 AC Haren, The Netherlands

Summary - The extent to which the aggregates of different size classes from a sand and a clay soil differ in their physical and biological characteristics and how this affects nitrogen mineralization rates was studied. Five size fractions were obtained: coarse sand (250-2000 μm), fine sand (50-250 μm), coarse silt (20-50 μm), fine silt (2-20 μm), and clay (< 2 μm). Total carbon and nitrogen concentration were relatively higher in the clay size fraction (< 2 μm) in both the sand and the clay soils; the sand soil, however, had a higher concentration. An inverse and significant relationship was established between carbon mineralization rate, nitrogen mineralization rate or microbial biomass carbon and the clay content of aggregates in the sand soil. There were, no similar relationships for carbon mineralization rate or microbial biomass carbon in the clay soil, whereas there was a positive correlation between nitrogen mineralization rate and clay content of the aggregates in this soil. This suggests that soil organic matter was strongly protected against decomposition in the finest fraction (< 20 μm) in the sand soil, apparently by adsorption, while in clay soil other mechanisms of soil organic matter protection were involved. It is not clear yet to what extent physical protection of soil organic matter in small pores is the main mechanism of protection in fine-textured soils.

INTRODUCTION

The amount and distribution of soil organic matter (SOM) in various aggregate size classes is affected by both soil texture and soil structure (Tisdall and Oades, 1982). An important mechanism affecting the carbon (C) and nitrogen (N) mineralization in soil is the physical protection of SOM from biodegradation. Indirect evidence of such protection may be obtained when soils are dried or rewetted (Sørensen, 1983; Van Gestel *et al.*, 1991) or in other cases when soil aggregates are disrupted, causing a temporary increase in SOM decomposition (Rovira and Greacen, 1957; Crasswell and Waring, 1972; Hassink, 1992). In sand soil the adsorption of SOM to the clay and

silt particles is the main mechanism of physical protection where these particles are more concentrated with SOM than in clay soil (Hassink *et al.*, 1993). Clay soils may protect more SOM than sand soils in small pores of aggregates rather than by adsorption (Hassink *et al.*, 1993). So, the mechanism of physical protection by adsorption to the clay and silt particles is more important in sand soils, while the mechanism of physical protection in small pores may be more important in clay soil (Hassink *et al.*, 1993). Aggregates of soil are considered to be the basic natural units: micro-aggregates are organo-mineral complexes < 250 μm (clay-polyvalent metal-organic matter complexes) which link together to form macro-aggregates defined as being larger than 250 μm (Tisdall and Oades, 1982). Macro- and micro-aggregates of one soil are different in clay content (Matus, 1995) and pore size distribution (Jocteur-Monrozier *et al.*, 1991). Soil texture and structure also determine the habitat for soil organisms. The accessibility of pores for soil organisms has been suggested as an important control mechanism of bacterial biomass turnover and consequently of C and N mineralization rates in soil (Kuikman and Van Veen, 1989; Rutherford and Juma, 1992). Bacteria are unable to enter the smallest pores with neck diameters < 0.2 μm. The number of micro-organisms in "inner" pores of soil aggregates are higher than on the external surfaces and in large pores (Vargas and Hattori, 1986) probably because they are protected against grazing by protozoa and nematodes which are unable to enter pores with neck diameters < 6 and < 15 μm, respectively (Jones, 1969; Postma *et al.*, 1989; Kuikman *et al.*, 1990; Heijnen *et al.*, 1991). There are no studies so far in which both mechanisms of protection of organic matter have been studied at the same time in aggregates of different clay content from soils of different textures.

The objectives of this study are 1) to compare the distribution of total C, total N and microbial biomass C of various aggregate size classes in a sand and a clay soil as influenced by different mechanisms of SOM protection and 2) to measure soil organic matter protection as indicated by the rates of C and N mineralization after soil disaggregation.

MATERIALS AND METHODS

Soils

The top 15 cm of a sandy and a clay soil were used in this study. Some properties of these soils are given in Table 1. The soils have been used in an experiment in which plant material was incorporated in soil and allowed to decompose for almost 500 days under field conditions. During this period spring wheat was sown twice. To assess the changes in the chemical and physical properties of soil aggregates after two growing seasons, soil samples taken before the plant material was added (1990) and soil samples at the end of the experiment (1992) were used for soil fractionation.

Table 1. Some properties of the soils

Soil properties	Sand	Clay
Total C (%)	1.62	1.76
Total N (%)	0.11	0.19
C:N	14.70	9.30
$N\text{-}NH_4^+$ (mg kg^{-1})	0.40	2.10
$N\text{-}NO_3^-$ (mg kg^{-1})	13.50	14.50
P (mg l^{-1})	21.80	12.20
K (mg kg^{-1})	107.90	273.90
Mg (mg kg^{-1})	116.00	nd
$CaCO_3$ (%)	0.60	0.30
pH-KCl	7.22	6.98
Bulk density (g cm^{-3})	1.30	1.15

nd = not determined

Physical measurements

Aggregate size distribution. The USDA scheme for primary particle size distribution (Gee and Bauder, 1986) was used: clay < 2 μm, silt between 2 and 50 μm and sand between 50 and 2000 μm. Two types of aggregates were distinguished: macro-aggregates (> 250 μm) and micro-aggregates (< 250 μm) (Tisdall and Oades, 1982). Soil fractionation was conducted on four separate occasions in subsamples of 100 g of soil. The soil was placed in the first of a series of three stacked round stainless steel sieves (Eijkelkamp, Holland) with mesh sizes of 250, 50 and 20 μm from top to the bottom; these were set on a vibrator (Haver & Boecker, 220 V and 50 Hz) provided with a sliding lid and a water sprinkler system connected to a source of demineralized water (flow rate 1.1 liter per minute). The soil was collected into a 5-liter bucket. Four minutes sieving was required to obtain clear water passing through the last sieve. In this way four aggregate size classes were obtained: coarse sand (250-2000), fine sand (50-250), coarse silt (20-50) and the fine silt plus clay (< 20 μm) and one unfractionated soil (< 2000 μm), referred to as "whole soil". The first three classes were recovered by filtration and the fine silt plus clay size class (< 20 μm) was separated from the soil suspension containing this fraction. The clay size fraction (< 2 μm) was separated by means of the following procedure: the suspension containing the < 20 μm fraction was transferred to a 5-liter bottle and then shaken end-over-end in order to homogenize it. One liter of suspension was poured into a sedimentation glass cylinder and again homogenized. A table showing the settling-times for clay particles at temperatures between 15 and 25°C was constructed by applying Stoke's law and a particle density of 2.675 g cm^{-3}. The temperature of the suspension was recorded. After the correct settling-time, clay fractions were isolated by siphoning the upper 5.5 cm of the suspension into a suction flask. The soil suspension remaining in the 5-liter bottles was treated by adding 0.1 M $CaCl_2$ solution to reach a final

concentration of 0.01 M. This allowed a complete settling of the < 20 μm fraction, as observed by a clear supernatant after flocculation overnight at 5°C. All fractions were oven-dried at 40°C for 24 or 48 hours and stored at room temperature for analysis. All sievings were performed in duplicate.

Distribution of primary particles. Standard analysis of particle sizes by the pipette method (Day, 1965) was performed in one replicate only for each aggregate size class and both whole soils. The content of clay, silt and sand in whole soils, and the content of clay in each aggregate class, were determined.

Pore size distribution. The pore size distribution in the classes 0.2-1.2, 1.2-6, 6-15 and 15-30 μm was estimated in various aggregate size classes and whole soils. Assuming that the pore volume is described as a set of vertical capillary tubes the effective pore necks diameters (d, μm) were calculated as (Hassink *et al.*, 1993):

$$d = 2 * r = -\frac{294}{h} \qquad (1)$$

where r = radius of curvature of the capillary pore (μm) and h = pressure head (kPa). The gravimetric water contents (on an oven-dry weight basis) in a range of pressure heads of -1,555; -246; -49; -20 and -10 kPa, equivalent to a pore diameter of 0.2, 1.2; 6; 15 and 30 μm, respectively, were determined from the water retention curve (Klute, 1986). The pore size distribution in each aggregate size class and whole soils was calculated as a percentage of soil porosity. The porosity (p, %) was estimated according to Hillel (1980):

$$p = \frac{(\rho_s - \rho_b)}{\rho_s} * 100 \qquad (2)$$

where ρ_b = dry bulk density (g cm^{-3}) and ρ_s = soil particle density (g cm^{-3}).

The estimation of pore neck diameter was considered to be a rough approximation because the hysteresis effect, irregular pore shapes and the shrink-swell characteristics of clay soils introduce errors in the determination (Darbyshire *et al.*, 1993). The following was assumed: (i) the amount of water from the whole soil was assumed to represent the summed amounts of water from each aggregate class. If this total was higher than that from the whole soil it was because of the water retained in the pores between aggregates. A proportional amount of water was then subtracted from each aggregate; (ii) each aggregate was considered to be stable in water, so water saturation prior to the determination did not produce further disaggregation; (iii) bulk densities, 1.2 and 1.3 g cm^{-3} for whole clay and sand soil, respectively, were taken for each size class. The particle density was taken to be 2.675 g cm^{-3}.

Chemical and biological measurements

Total C and total N. For the determination of total C and total N concentration the whole soil and aggregate size fractions from both soils were oven-dried at 70°C and finely ground to powder in a mortar. Total N (including mineral NO_3^--N plus NH_4^+-N) was determined by oxidation of each soil with sulphuric acid and salicylic acid and the N distilled according to Deijs (1961). Total C was determined in whole soils and aggregates by means of oxidation with a concentrated acid dichromate solution according to Mebius (1960).

Microbial biomass C and C mineralization rates. Prior to the determination of microbial biomass by the chloroform fumigation incubation technique (Jenkinson and Powlson, 1976), about 2 g of each aggregate and whole soils were moistened slowly to -10 kPa with a nutrient solution containing $Ca(H_2PO_4)_2.H_2O$, K_2SO_4, $MgSO_4.7H_2O$ and $Ca(SO_4)_2.H_2O$. The amount of Ca, P, Mg, S and K added was equivalent to 372, 226, 48.6, 208.1 and 195.2 mg l^{-1} in solution, respectively. The samples were pre-incubated during 8 days at 20°C. Then the samples were fumigated and incubated, and microbial biomass was calculated between day 8 and 18 as the difference between CO_2 evolved from fumigated and non-fumigated samples. No conversion factor K_c was used (Ritz *et al.*, 1992). The accumulated CO_2 from non-fumigated samples between 0-18 days (referred to 0-20 days) was used to calculate the C mineralization rates.

N mineralization. Eight gram of whole dry soil and of the various aggregate size fractions were moistened to -10 kPa as described above . All soils were incubated in the dark in a glass vial (15 cm^{-3}) at 20°C and 80% humidity. Drying of samples was prevented by covering the vials with a seal permeable to air but impermeable to water. The increase in mineral N was determined after 20 or 40 days of incubation. At each time, mineral N from the whole soils and the aggregate size fractions was extracted with 1 M KCl solution for 1 hour at a soil:water ratio of 1:2.5. Inorganic N (NH_4^+-N and NO_3^--N) was measured colorimetrically after extraction with 1M KCl solution for 1 h at a soil:water ratio of 1:2.5 using a Technicon autoanalyser (Traacs 800). The ^{15}N content in inorganic N was determined as mentioned above.

Statistical analysis

Analysis of variance (ANOVA; Genstat 5.0), and Student's t-test were used to analyze the differences of means between treatments. The level of probability at which significant differences were found was set at the 5%.

RESULTS

Physical properties of whole soil and various aggregate size classes

Aggregate size distribution. Micro-aggregates, 50-250 μm in diameter, predominated in the sand soil (Fig. 1a). In clay soil the largest amount of soil recovered was in the macro-aggregates, 250-2000 μm in diameter (Fig. 1b). On both sampling dates, the aggregate size distribution was not significantly different in the clay soil, while in the sand soil significant differences were found in the aggregates 250-2000 μm.

Distribution of primary particles. The standard textural analysis of the whole soil and the clay content of each aggregate size class are presented in Figs. 2a and 2b. Comparing the dry weight distribution of primary particle sizes and aggregate sizes showed a similar pattern between the two distributions in sand soil, while considerable differences were observed in clay soil (cf. Figs. 1 and 2a). In the sand soil the clay content varied considerably from one aggregate size class to another. Aggregates < 20 μm contained most clay (Fig. 2b). As in the sand soil, the largest amount of clay was found in the < 20 μm fraction in clay soil, but the clay content of the aggregates was more homogeneously distributed (Fig. 2b).

Pore size distribution. Pore size distribution in various aggregates and whole soils were found to be similar on both sampling dates and the results are presented only for 1992 (Table 2). In sand soil pores > 30 μm in diameter were the most abundant in aggregates > 50 μm, while in clay soil pores < 1.2 μm in diameter were equally distributed in all aggregates and few large pores were observed.

Chemical and biological properties

Total C. The largest concentrations of C were found in the clay (< 2 μm) and fine silt (2-20 μm) size fraction on both sampling dates in sand soil. The lowest C concentration, about 6 g kg^{-1}, was observed in the 50-250 μm size fraction, also in sand soil (Fig. 3a). In the clay soil, total C was equally distributed over various aggregate size classes and whole soil, and no significant differences were observed (Fig. 3b).

Total N. The largest concentrations of N, as for total C, were found in the clay (< 2 μm) and in the fine silt (2-20 μm) size fraction of the sand soil (Fig. 3c). In the clay soil, although smaller differences were observed, the clay size fraction contained significantly more N than the other size classes and the whole soil (Fig. 3d). On both sampling dates, even when significant

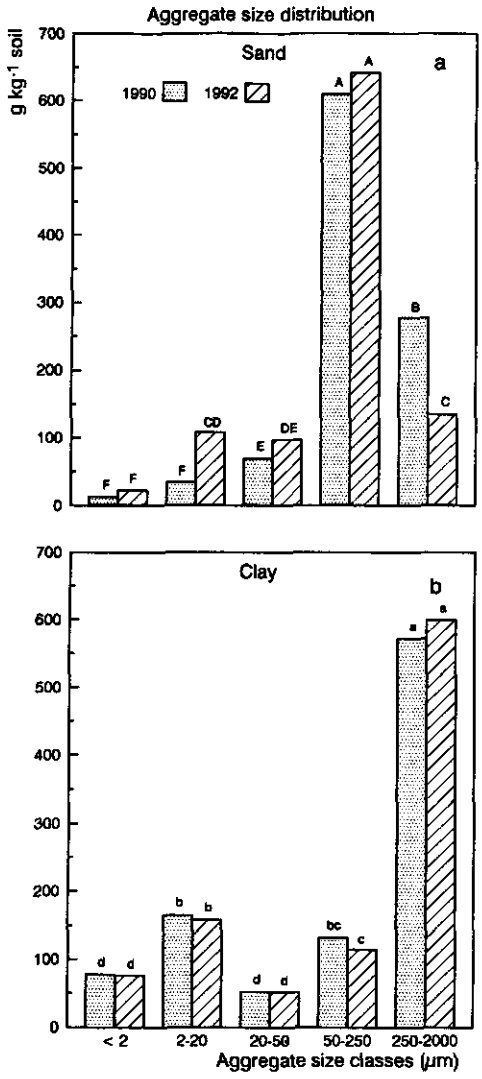

Fig. 1 Distribution of mass of various aggregate size classes in (a) sand soil and (b) clay soil. Bars carrying the same letter are not significantly different ($P < 0.05$).

differences ($P < 0.05$) were found, the distribution of N amongst aggregates from both soils was similar.

Carbon to nitrogen ratio. In both soils and on both sampling dates (except sand soil in 1992) the C:N ratios were significantly ($P < 0.05$) lower in the clay (< 2 µm) size fraction than in the other

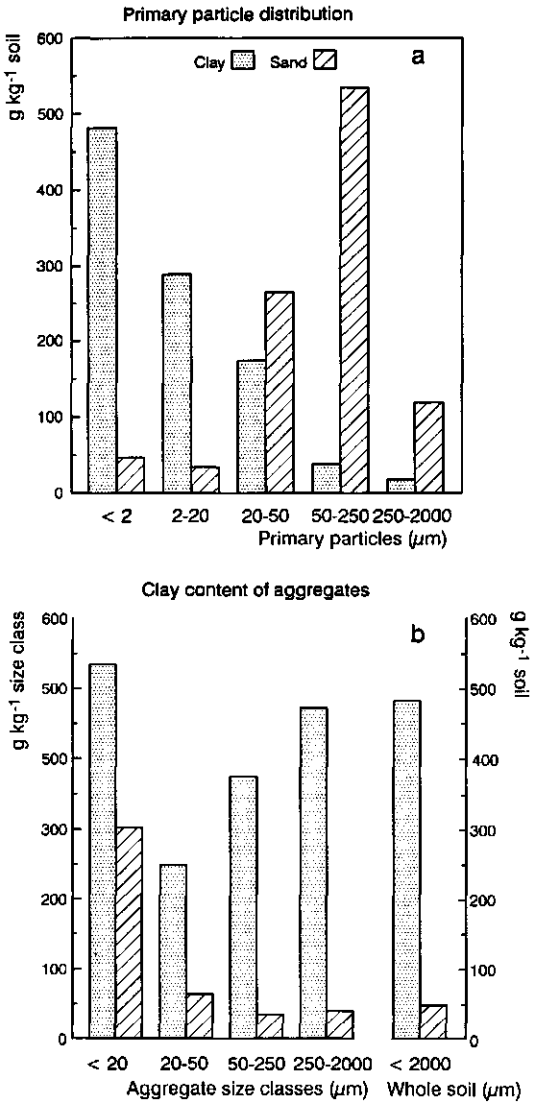

Fig. 2. Standard textural analysis of sand and clay soils (a) and content of clay (b) in various aggregate size classes in a sand and clay soil.

aggregate size classes and whole soils (Fig. 3 e and f). The C:N ratio in sand soil gradually decreased with the aggregate size (Fig. 3e), while in clay soil no significant differences were observed, except in the clay size fraction (< 2 μm) (Fig. 3f).

Table 2. Pore size distribution (% of soil porosity) in various aggregate size classes and in whole sand and clay soil in 1992

Aggregate size classes (μm)	Pore neck diameter (μm)				
	< 1.2	1.2-6	6-15	15-30	>30
Sand					
< 2000 (whole soil)	14.7	6.9	7.8	7.8	62.9
250-2000	10.8	6.5	1.7	5.1	76.0
50-250	5.8	9.2	7.8	4.2	73.0
20-50	11.9	24.5	14.1	27.2	22.1
< 20	45.7	11.8	nd	nd	nd
Clay					
< 2000 (whole soil)	46.2	12.0	15.6	2.0	24.1
250-2000	45.1	21.5	12.3	6.3	14.9
50-250	42.1	24.6	18.0	6.9	8.5
20-50	31.7	6.4	17.0	4.4	nd
< 20	55.1	20.3	12.7	0.9	11.0

nd = not determined

C mineralization rates and microbial biomass C. To compare the C mineralization rates and microbial biomass C between various aggregate size classes and between those and whole soils, the results were expressed per unit of total C present in each fraction or whole soil (Table 3). The highest rates of C mineralization were observed in the sand size fraction (50 - 250 and 250 - 2000 μm) and the lowest values were found in the fine silt plus clay size fraction (< 20 μm) in sand soil (Table 3). In clay soil the mineralization rates were not significantly different. In the sand soil the amount of microbial biomass C was found to be larger in the sand size fractions on both sampling dates, whereas in clay soil few significant differences were observed (Table 3).

N mineralization rates. The N mineralization rates were also expressed per unit of total N present in each fraction or whole soil (Table 4). As with the C mineralization rates, the highest N mineralization rates were found in the sand size fractions, while the lowest values, as for C mineralization, were observed in the < 20 μm size fraction in sand soil (Table 4). No significant differences were found in clay soil.

C and N concentration ratios of the clay fraction. The C and N concentration ratio (C_r, N_r) was defined as the total concentration of C or N in the clay size fraction (< 2 μm) divided by the total concentration of C or N in the whole soil. Both, C_r and N_r reached values between 7 and 10 in sand and between 1.2 and 2.4 in clay soil. Our C_r and N_r values, and those obtained from published data were plotted against the clay content of soils (Fig. 4a and b). Regardless of the

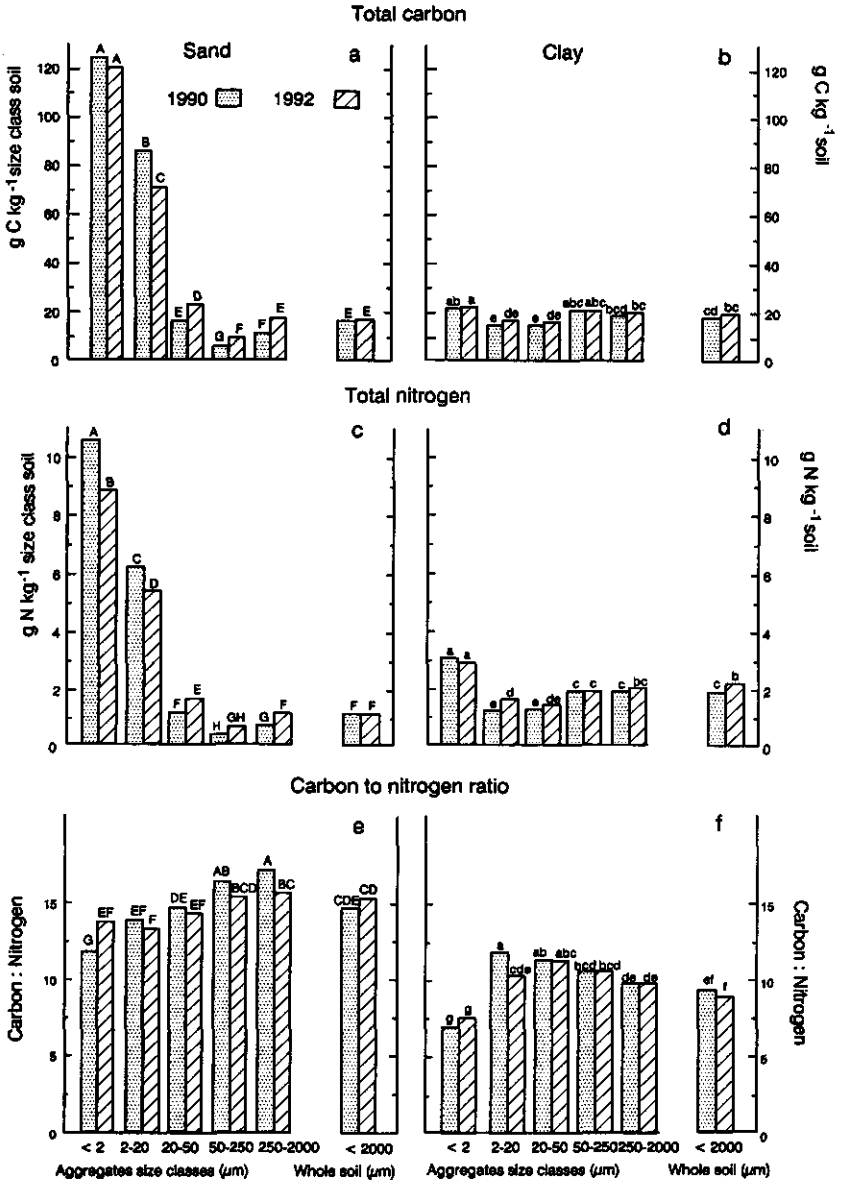

Fig. 3. Total C concentration in (a) sand soil and (b) clay soil, total N concentration in (c) sand soil and (d) clay soil and C:N ratio in (e) sand and (f) clay soil.

Table 3. C mineralization rates (g $C0_2$ kg^{-1} whole soil C or size class C day^{-1}) and microbial biomass C (g CO_2 kg^{-1} whole soil C or size class C) from various aggregate size classes and from whole sand and clay soil on both sampling dates

Aggregate size classes (µm)	C-mineralization 1990	C-mineralization 1992	Microbial biomass C 1990	Microbial biomass C 1992
Sand				
< 2000 (whole soil)	0.5	1.1	14.4	38.6
250-2000	1.8	0.7	151.2	107.6
50-250	1.5	1.3	309.1	189.7
20-50	0.7	0.7	116.1	75.2
< 20	0.5	0.5	13.1	1.9
Clay				
< 2000 (whole soil)	0.5	0.7	24.7	42.8
250-2000	0.6	0.7	47.4	88.6
50-250	0.6	0.7	85.6	75.7
20-50	1.0	1.4	113.1	109.9
< 20	0.6	1.8	83.9	72.8

L.S.D[1]. C-mineralization = 1.1
L.S.D. microbial biomass C = 52
[1] Least significant differences ($P < 0.05$)

Table 4. N mineralization rate (g N kg^{-1} whole soil N or size class N day^{-1}) from various aggregate size classes and from whole sand and clay soil on both sampling dates

Aggregate size classes (µm)	1990		1992	
	0-20[1]	0-40[1]	0-20	0-40
Sand				
< 2000 (whole soil)	nd	nd	0.40	0.30
250-2000	1.10	0.50	0.90	0.70
50-250	0.60	0.40	0.70	0.50
20-50	0.50	0.30	0.40	0.30
<20	nd	nd	0.20	0.10
Clay				
< 2000 (whole soil)	nd	nd	0.30	0.20
250-2000	0.50	0.30	0.50	0.30
50-250	nd	nd	0.10	0.10
20-50	nd	nd	0.01	0.01
<20	nd	nd	0.50	0.30

L.S.D.[2] N-mineralization = 0.4
[1] rates obtained after 20 or 40 days of incubation.
[2] Least significant differences ($P < 0.05$).
nd = not determined

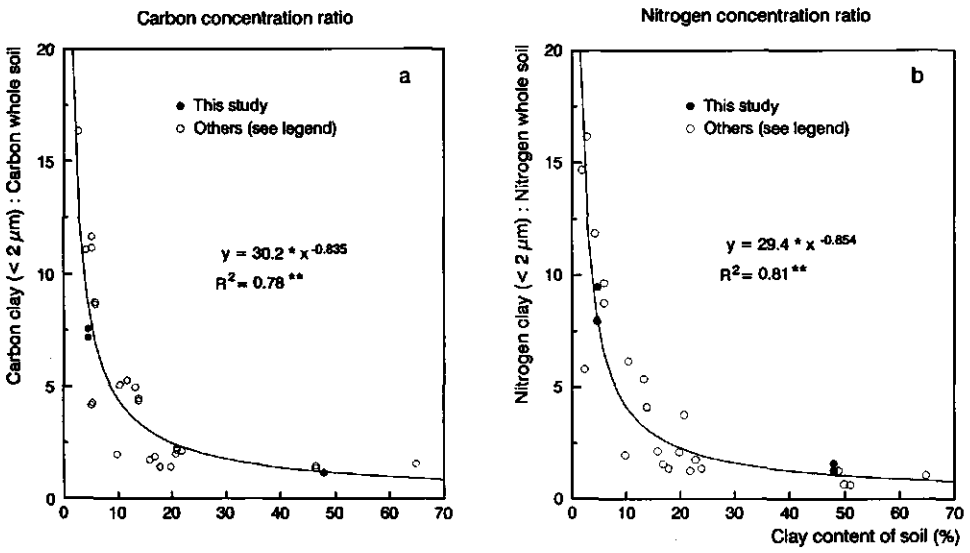

Fig. 4. Carbon (a) and nitrogen (b) concentration ratio of the clay size fraction (< 2 μm) and its relationship with the clay content of soil (Cameron and Posner, 1979; Tiessen and Stewart, 1983; Christensen, 1985; Christensen and Sørensen, 1985; Christensen, 1987; Christensen and Christensen, 1991; *Catroux and Schnitzer, 1987; Jocteur-Monrozier et al., 1991; Matus, 1995*).(**= *significant* at 0.01% level).

methods of soil dispersion used (wet sieving in this study or very destructive methods such as ultrasound), C_r and N_r were found to decrease with increasing clay content of soils (Fig. 4a and b).

Relationship of C and N mineralization rates and microbial biomass C with the clay content of aggregates. The percentage of aggregate C that mineralized and the percentage of aggregate C present in the microbial biomass decreased as the clay content of aggregates increased in sand soil (Figs. 5a and c). No such relationship were found in the clay soil (Figs. 5b and d). As with C mineralization, an inverse relationship was found between the clay content of aggregates and the percentage of aggregate N mineralized in sand soil (Fig. 6a). In contrast, there was a positive correlation between clay content of aggregates and N mineralization in clay soil (Fig. 6b).

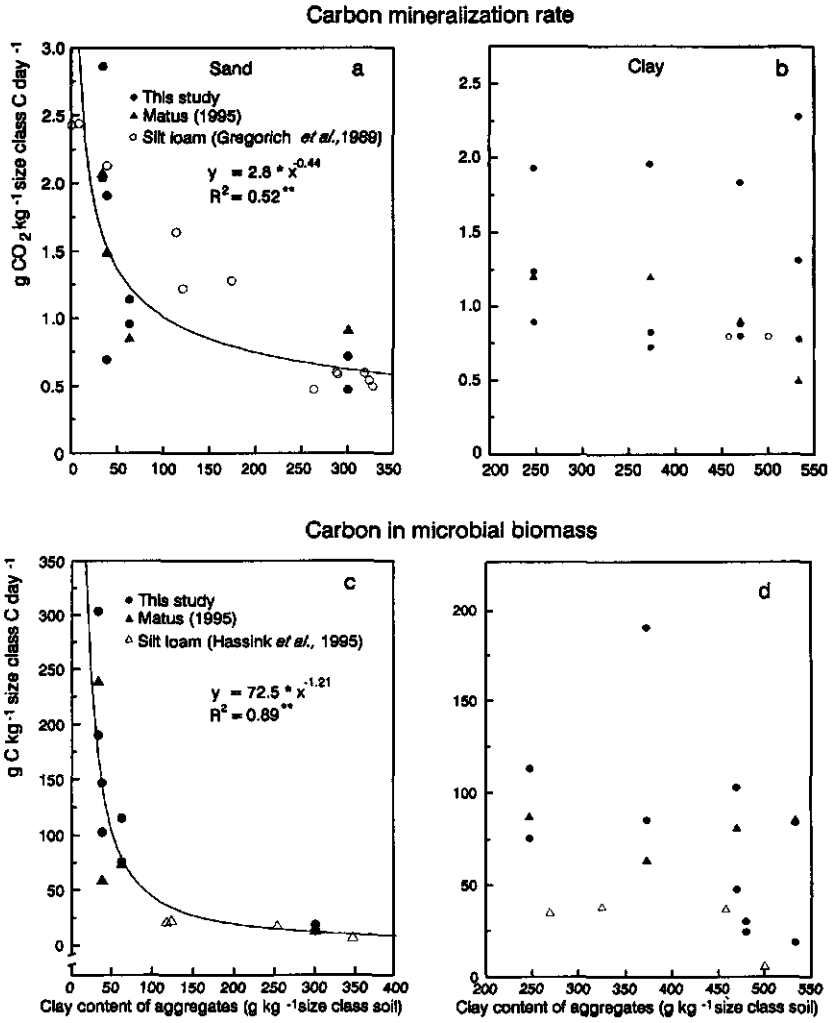

Fig. 5. Relationship between clay content of various aggregate size classes in sand soils and C mineralization rates (a) and microbial biomass C (c), and between clay content of various aggregate size classes in clay soil and C mineralization rates (b) and microbial biomass C (d).

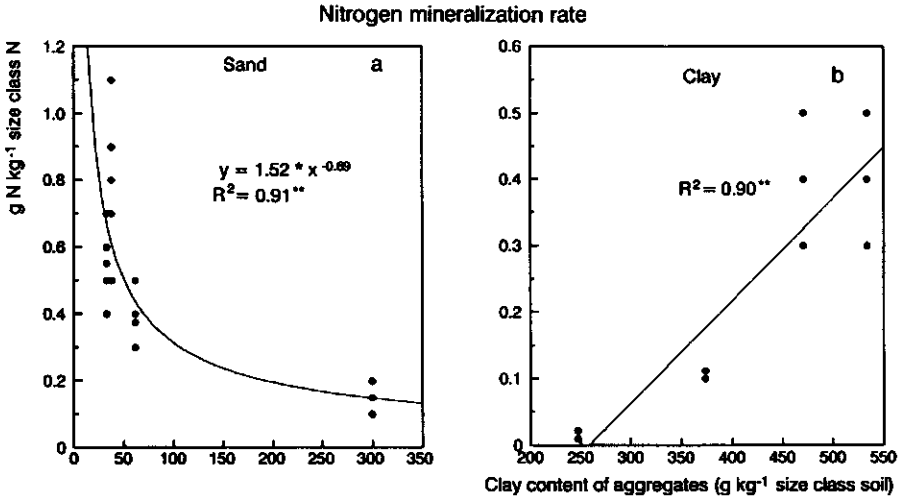

Fig. 6. Relationship between N mineralization rates and clay content of various aggregate size classes in sand (a) and clay (b).

DISCUSSION

Aggregate size distributions

After two years under field conditions, little changes were observed in the aggregate size distribution in both soils (Fig. 1a and b). Although, a significant less mass of soil was recovered from the macro-aggregate (> 250 μm) in 1992 than 1990 in the sand soil (Fig. 1a), this difference was not observed in the clay soil (Fig 1b). The results indicate that the macro-aggregates, especially in clay soil were stable to climatic events such as drying, wetting or freezing under field conditions.

Wet-sieving caused almost complete disaggregation in the sand soil but not in the clay soil. In sand soil most soil was found in the 50-250 μm aggregate fraction (Fig. 1a), coinciding with the largest recovery of primary particles (Fig. 2a). The term aggregate is justified in the sand soil, however, because a certain amount of clay (Fig. 2b) was measured in each fraction. How an incomplete disaggregation in clay soil can influence the SOM distribution in each fraction is discussed in the next section. The wet-sieving technique adopted in the present investigation was preferred to a more destructive mechanical treatment, such ultrasonic dispersion, in order to avoid losses of SOM and microbial biomass (Ladd *et al.*, 1993).

Effect of soil texture on the distribution of total C and total N of various aggregate size classes

The greatest concentrations of C and N were observed in the clay (< 2 μm) and in the fine silt (2-20 μm) size fraction in the sand soil (Fig. 3a and c). Carbon was rather uniformly distributed among aggregates in the clay soil, while the N concentration was found to be highest in the clay size fraction (Fig. 3d). C_r and N_r, the C and N concentration ratios of the clay size fraction, were 7.4 and 9 in sand soil, and 1.2 and 1.5 in clay soil. It may be argued that a direct comparison of SOM between clay and sand soil is not possible because a complete soil dispersion was not reached in clay soil. The partitioning of SOM from whole soil into several aggregate size classes depends on the extent of soil dispersion reached (Christensen, 1992; Cambardella and Elliott, 1993). However, there are no studies aimed directly at examining the redistribution of SOM detached from organo-mineral complexes during soil dispersion (Christensen, 1992). In the present study the C_r and N_r from the soils studied here and other ratios obtained from the literature were inversely correlated with the soil clay content (Fig. 4a and b), i.e., C or N was much more concentrated in the clay (< 2 μm) size fraction of coarse- than in that of fine-textured soils. From the data of Gregorich *et al.* (1988) C_r of the silt plus clay (< 50 μm) size fraction of a silt loam soil could be calculated to increase only from 1.0 to 1.2 when the energy input increased from 0 to 1500 J ml^{-1}. This means that most of the organic C is associated with silt and clay particles and that its redistribution after different levels of ultrasonic energy had been applied did not alter the C concentration in these fractions. From these results it can be concluded that no matter what intensity is applied to disperse the soil, the C content in the fine silt plus clay size fraction (< 20 μm) was greater in coarse- than in fine-textured soils (Fig. 4a and b). Microscopical studies have revealed that clay aggregates are a massive assembly of clay particles forming dense lattices, whereas in sand aggregates clay particles are loosely arranged (Oades and Waters, 1991; Hassink *et al.*, 1995). As a consequence more SOM can be stabilized per unit of surface of clay in the sand than in the clay soil (Hassink *et al.*, 1995). For example, the fine silt plus clay size fraction (< 20 μm) in the clay soil had a concentration of clay almost two times higher than in the sand soil (Fig. 2b). It can be calculated that the total C and N concentration in the fine silt plus clay size fraction in clay soil was 32 mg C and 3.6 mg N g^{-1} clay and in the sand soil 294 mg C and 22 mg N g^{-1} clay. The higher concentration of SOM found in the < 20 μm size fraction in sand soil may have been physically protected by adsorption onto the surface of the clay lattice forming very stable aggregates (Tisdall and Oades, 1982). Soil organic matter also may be isolated in pores out of reach of microorganisms (Van Veen *et al.*, 1985; Elliott and Coleman, 1988; Hassink *et al.*, 1993).

Effect of clay content on C mineralization, N mineralization and microbial biomass C of various aggregate size classes

An inverse relationship was found between the clay content of aggregates and C mineralization rate or microbial biomass C in the sand soil (Fig. 5a and c). The highest C mineralization rate and amount of microbial biomass C were found in the sand size fraction (50-250 μm) with the lowest clay content (Fig. 2b) and the lowest total C (Fig. 3a), whereas the lowest C mineralization rate and microbial biomass C were observed in the < 20 μm fraction with the highest clay content (Fig. 2b) and the highest total C (Fig. 3a). The results suggest that SOM associated with the < 20 μm fraction may have been strongly protected against decomposition, presumably coated by silt and clay particles, while SOM associated with sand size fractions (> 50 μm) was not. It is unlikely that the C mineralization in the < 20 μm fraction was reduced by the presence of charcoal found in similar sand soils used as pastures (Hassink, 1994) for two reasons:

1) visual inspection of both whole sand and whole clay soil did not show the presence of burnt organic matter, similar to that described by Hassink (1994);

2) similar reduction of the N mineralization rate in the same fraction (< 20 μm) was also observed (Fig. 6a).

In the present study sand size aggregates may include particulate macro-organic matter in addition to the aggregates proper because these organic materials were not washed away. Others have found that SOM associated with sand size fractions decomposes faster than SOM associated with the silt and clay fractions (Tiessen and Stewart, 1983). Sand fractions contain partially humified macro-organic matter (Elustondo *et al.*, 1990). Angers and Mehuys (1990) reported an increase in the amount of plant-derived carbohydrates in the sand fractions as well. Contrary to the observation in the sand soil, a similar relationship between C mineralization or microbial biomass C with the clay content of aggregates was not found in the clay soil (Fig. 5b and d). As for C mineralization, an inverse relationship between N mineralization rates and clay content of aggregates was found in sand soil (Fig. 6a). There was, however, a positive and significant ($P < 0.01$) correlation between N mineralization rate and the clay content of aggregates in the clay soil (Fig. 6b). This coincided with a positive and significant ($P < 0.05$) correlation ($R^2 = 0,94$) in the same soil between pores < 1.2 μm in diameter and the clay content of the aggregates (cf. Fig 2b and Table 2). The distribution of pore spaces in several aggregates and whole soils indicated that most pores found in the clay soil were in the class < 1.2 μm in diameter, while in sand soils most of the pores were > 30 μm in diameter (Table 2). The good correlation found between the clay content of aggregates with the N mineralization rate and the proportion of pores < 1.2 μm in diameter in clay soil may indicate that SOM is physically protected in small pores, as suggested before by Hassink *et al.* (1993). However, similar correlations of clay content of aggregates with C mineralization rates or microbial biomass C were not found, while the

positive correlation with N mineralization rate may simply be the consequence of the low C:N ratio in the clay fraction in clay soil (Fig.3f). The explanation of the differences in C:N ratio between size fractions is beyond the scope of this study. So, from the present results it is not clear yet to what extent the organic materials are entrapped in small pores, protected against decomposition in clay soil.

CONCLUSIONS

Clay (< 2 µm) size fraction had the highest total C and N concentration of all aggregate size classes in both sand and clay soils. Total C and N were much more concentrated in the clay size fraction of the sand than of the clay soil. This higher concentration in sand soil coincided with the lowest C and N mineralization rates and microbial biomass C. This indicates that SOM was protected mainly by adsorption to the silt and clay particles. In contrast, there was no relationship between the clay content of the aggregates with the C mineralization rate and microbial biomass C in clay soil, except for a positive correlation established for N mineralization. It is not clear yet to what extent SOM in clay soil is physically protected against decomposition in small pores.

Acknowledgements - I thank K. Kampen, A. Vos, A. Lahr and M. Wolters and J. Zwiers for their technical assistance. I also thank Ir. J. Hassink, Dr. K. B. Zwart, Dr. L.A. Bouwman and Dr. J. Bloem for their helpful discussions and comments on the manuscript. I especially thank Dr. L. Brussaard and Dr. A.P. Whitmore for their support in the course of this investigation, critical comments, discussions and final revision of the manuscript. Part of this work was supported by a grant N° P806 under the sandwich PhD Fellowship Programme of the Wageningen Agricultural University and by a grant under the Presidential Fellowship Programme of the Agencia de Cooperación Internacional (AGCI) del Ministerio de Planificación y Cooperación de Chile (MIDEPLAN).

REFERENCES

Angers, D.A. and Mehuys, G.R. (1990) Barley and alfalfa cropping effects on carbohydrate contents of a clay soil. *Soil Biology & Biochemistry* **22**, 285-288.

Cambardella, C. A. and Elliott, E.T. (1993) Carbon and nitrogen distribution in aggregates from cultivated and native grassland soils. *Soil Science Society of America Journal* **57**, 1071-1076.

Cameron, R.S. and Posner, A.M. (1979) Mineralisable organic nitrogen in soil fractionated according to particle size. *Journal of Soil Science* **30**, 565-577.

Catroux, G and Schnitzer, M. (1987) Chemical, spectroscopic, biological characteristics of soil organic matter in particle size fractions separated from an Aquoll. *Soil Science Society of America Journal* **51**, 1200-1207.

Craswell, E.T. and Waring, S.A. (1972) Effect of grinding on the decomposition of soil organic matter -II. Oxygen uptake and nitrogen mineralization in virgin and cultivated soils. *Soil Biology & Biochemistry* **4**, 435-442.

Christensen, B.T. (1985) Carbon and nitrogen in particle-size fractions isolated from Danish arable soils by ultrasonic dispersion and gravity-sedimentation. *Acta Agriculturae Scandinavica* **35**, 175-187.

Christensen, B.T. (1987) Decomposability of organic matter in particle size fraction from field soils with straw incorporation. *Soil Biology & Biochemistry* **19**, 429-453.

Christensen, B.T. (1992) Physical fractionation of soil organic matter in primary particles size and density separates. *Advances in Soil Science* **20**, 1-90.

Christensen, S. and Christensen, B.T. (1991) Organic matter available for denitrification in different soil fractions: effect of freeze/thaw cycles and straw disposal. *Journal of Soil Science* **42**, 637-647.

Christensen, B.T. and Sørensen, L.H. (1985) The distribution of native and labelled carbon between soil particle size fraction isolated from long-term incubation experiments. *Journal of Soil Science* **36**, 219-229.

Darbyshire, J.F., Chapman, S.J., Cheshire, M.V., Gauld, J.H., McHardy, W.J. Paterson, E. and Vaughan, D. (1993) Methods for the study of interrelationships between micro-organism and soil structure. *Geoderma* **56**, 3-23.

Day, P.R. (1965) Particle fractionation and particle-size analysis. In *Methods of Soil Analysis, Part 1, Physical and Mineralogical Properties, Including Statistics of Measurements and Sampling* (C.A. Black, Ed), pp. 545-566. ASA Publ., Madinson, WI, USA.

Deijs, W.B. (1961). The determination of total nitrogen in herbage samples (in Dutch). Instituut voor *Biologisch en Scheikundig Onderzoek van Landbouwgewassen Wageningen*, Jaarboek Mededelingen **89**, **158**, 90.

Elustondo, J., Angers, D. A., Laverdiere, M. R. and N'Dayegamiye, A. (1990) Étude comparative de l'agrégation et de la matière organique associée aux fractions granulométriques de sept sols sous culture de maïs ou en prairie. *Canadian Journal of Soil Science* **70**, 395-402.

Elliot, E.T. and Coleman, D.C. (1988) Let the soil work for us. *Ecological Bulletins* **39**, 23-32, Copenhagen.

Gee, G.W. and Bauder, J.W. (1986) Particle-size analysis. In *Methods of soil Analysis, Part I, Physical and Mineralogical Properties*, (A. Klute Ed.), pp. 383-441. 2nd Edition ASA and SSSA Publ., Madison, WI, USA.

Gregorich, E.G., Kachanoski, R.G. and Voroney, R.P. (1988) Ultrasonic dispersion of aggregates: distribution of organic matter in size fractions. *Canadian Journal of Soil Science* **68**, 395-403.

Gregorich, E.G., Kachanoski, R.G. and Voroney, R.P. (1989) Carbon mineralization in soil size fractions after various amounts of aggregate disruption. *Journal of Soil Science* **40**, 649-659.

Hassink, J. (1992) Effects of soil texture and structure on carbon and nitrogen mineralization in grassland soils. *Biology and Fertility of Soils* **14**, 126-134.

Hassink, J. (1994) Effect of soil texture and grassland management on soil organic C and N and rates of C and N mineralization. *Soil Biology & Biochemistry* **26**, 1221-1231.

Hassink, J. Bouwman, L.A., Zwart, K.B., Bloem, J. and Brussaard, L. (1993) Relationships between soil texture, physical protection of soil organic matter, soil biota, and C and N mineralization in grassland soils. *Geoderma* **57**, 105-128.

Hassink, J., Matus, F.J., Chenu, C. and Dalenberg, J.W. (1995) Interaction between soil biota, soil organic matter and soil structure. *Advances in Agroecology* (Submitted).

Heijnen, C.E., Hok-A-Hin, C.H. and Van Veen, J.A. (1991) Protection of *Rhizobium* by bentonite clay against predation by flagellates in liquid cultures. *FEMS Microbiology Ecology* **85**, 73-80.

Hillel, D. (1980) Fundamentals of soil physics. *Academic Press, New York.*

Jenkinson, D.S. and Powlson, D.S. (1976) The effects of biocidal treatments on metabolism in soil -V. A method for measuring soil biomass. *Soil Biology & Biochemistry* **8**, 209-213.

Jocteur-Monrozier, L., Ladd, J.N., Fitzpatrick, R.W., Foster, R.C. and Raupach, M. (1991) Components and microbial biomass content of size fractions in soils of contrasting aggregation. *Geoderma* **49**, 37-62.

Jones, F.G.W., Larbey, D.W. and Parrott, D.M. (1969) The influence of soil structure and moisture on nematodes, especially Xiphinema, Longidorus, Trichdorus and Heterodera spp. *Soil Biology & Biochemistry* **1**, 153-165.

Klute, A. (1986) Water retention: Laboratory methods. In *Methods of soil Analysis, Part I, Physical and Mineralogical Properties*, (A. Klute Ed.), pp. 635-662. 2nd Edition ASA and SSSA Publ., Madison, WI, USA.

Kuikman, P.J. and Van Veen, J.A. (1989) The impact of protozoa on the availability of bacterial nitrogen to plants. *Biology and Fertility of Soils* **8**, 13-18.

Kuikman, P.J., Van Elsas, J.D., Jansen, A.G., Burgers, S.L.G.E. and Van Veen, J.A. (1990) Population dynamics and activity of bacteria and protozoa in relation to their spatial distribution in soil. *Soil Biology & Biochemistry* **22**, 1063-1073.

Ladd, J.N., Foster, R.C. and Skjemstad, J.O. (1993) Soil structure: carbon and nitrogen metabolism. *Geoderma* **56**, 401-434.

Matus, F.J. (1995) The distribution of soil organic matter of various aggregate size classes in arable soil. II. Residual organic ^{14}C, residual ^{15}N, microbial biomass ^{14}C and ^{14}C and ^{15}N mineralization rates in a sand and a clay soil. *Soil Biology & Biochemistry* (submitted).

Mebius, L.J. (1960) A rapid method for the determination of organic carbon in soil. *Analytica Chimica Acta* **22**, 120-124.

Oades, J.M. and Waters, A.G. (1991) Aggregate hierarchy in soils. *Australian Journal of Soil Research* **29**, 815-828.

Postma, J, Walter, S. and Van Veen, J.A. (1989) Influence of different initial soil moisture contents on the distribution and population dynamics of introduced *Rhizobium leguminosarum* biovar *trifolii*. *Soil Biology & Biochemistry* **21**, 437-442.

Ritz, K., Griffiths, B.S. and Wheatly, R.E. (1992) Soil microbial biomass and activity under a potato crop fertilized with N with and without C. *Biology and Fertility of Soils* **12**, 265-271.

Rovira, A.D. and Greacen, E.L. (1957) The effect of aggregate disruption on the activity of microorganisms in the soil. *Australian Journal of Agricultural Research* **8**, 659-673.

Rutherford, P.M. and Juma, N.G. (1992) Influence of texture on habitable pore space and bacterial-protozoan populations in soil. *Biology and Fertility of Soils* **12**, 221-227.

Sørensen, L.H. (1983) The influence of stress treatments on the microbial biomass and the rate of composition of humified matter in soils containing different amounts of clay. *Plant and Soil* **75**, 107-119.

Tiessen, H. and Stewart, J.W.B. (1983) Particle-size fractions and their use in studies of soil organic matter:II. Cultivation effects on organic matter composition in size fractions. *Soil Science Society of America Journal* **47**, 509-514.

Tisdall, J.M. and Oades, J.M. (1982) Organic matter and water-stable aggregates in soils. *Journal of Soil Science* **33**, 141-163.

Van Gestel, M., Ladd, J.N. and Amato, M. (1991) Carbon and nitrogen mineralization from two soils of contrasting texture and microaggregates stability: influence of sequential fumigation, drying and storage. *Soil Biology & Biochemistry* **23**, 313-322.

Van Veen J.A., Ladd J.N. and Amato M. (1985) Turnover of carbon and nitrogen through the microbial biomass in a sandy loam and a clay soil incubated with [^{14}C(U)]glucose and [^{15}N](NH_4)SO_4 under different moisture regimes. *Soil Biology & Biochemistry* **17**, 747-756.

Vargas, R. and Hattori, T. (1986) Protozoan predation of bacterial cells in soil aggregates. *FEMS Microbial Ecology* **38**, 233-242.

CHAPTER 6

THE DISTRIBUTION OF SOIL ORGANIC MATTER OF VARIOUS AGGREGATE SIZE CLASSES IN ARABLE SOILS. II. RESIDUAL ORGANIC ^{14}C, RESIDUAL ^{15}N, MICROBIAL BIOMASS ^{14}C AND ^{14}C AND ^{15}N MINERALIZATION RATES IN A SAND AND A CLAY SOIL

F.J. Matus

DLO Research Institute for Agrobiology and Soil Fertility, P.O. Box 129, 9750 AC Haren, The Netherlands

Summary - The distribution of residual ^{14}C, residual ^{15}N, microbial biomass ^{14}C and the mineralization rates of ^{14}C and ^{15}N derived from labelled wheat residues in various aggregate size classes in a sand and a clay soil were studied. Five fractions were obtained: coarse sand (250 - 2000 μm), fine sand (50-250 μm), coarse silt (20-50 μm), fine silt (2-20 μm) and clay (< 2 μm). Total ^{14}C enrichment in the fine silt plus clay (< 20 μm) size fraction in the sand soil was four times more higher than in the clay soil, and total ^{15}N enrichment of the clay (< 2 μm) size fraction in sand soil was four times higher than in the clay soil. No correlations were found between the clay content of aggregates and the ^{14}C or ^{15}N mineralization rates and microbial biomass ^{14}C in sand soil. This suggests that recently produced soil organic matter was less protected from biodegradation than unlabelled SOM in sand soil. Contrary to what was found in the sand soil, there was a significant and positive correlation between the clay content of aggregate and ^{15}N-mineralization rate, calculated for 20 days of incubation, for clay soil. The correlation for ^{15}N mineralization, calculated for 40 days of incubation, however, was not significant. It is concluded that soil texture and structure played major roles in determining soil organic matter protection, but had a less important function in the protection of newly formed soil organic matter in sand soil. It is not clear yet to what extent soil organic matter localized in small pores is the main mechanism of physical protection in clay soil.

INTRODUCTION

The interaction between biological and physical properties of different aggregate size classes may provide valuable information on the mechanisms involved in soil organic matter (SOM) protection. Indirect evidence of such protection may be obtained when soils are dried or rewetted (Sørensen, 1983; Van Gestel *et al.*, 1991) or when soil aggregates are disrupted otherwise, causing a temporary increase in the decomposition of SOM (Rovira and Greacen, 1957; Crasswell and Waring, 1972; Hassink, 1992). The binding of SOM to clay and silt particles has been found to be

the most important protection mechanism in sand soils (Tidall and Oades, 1982; Hassink *et al.*, 1993). In clay soils a high proportion of SOM may be physically separated from micro-organisms in small pores within micro-aggregates (Elliott and Coleman, 1988). In a previous paper (Matus, 1995) it was concluded that adsorption of SOM to clay minerals was the main mechanisms in sand soil while in clay soil it was not clear to what extent SOM was physically protected in small pores. The present paper describes how recently formed SOM, derived from ^{14}C- and ^{15}N-labelled wheat straw was distributed in several aggregate size classes in the same soils studied by Matus (1995). The CO_2 evolution and ^{15}N mineralization from various aggregate size classes with different clay content and pore size distribution were used as a measure of SOM protection. The objectives of the present investigation were 1) to compare the distribution of total ^{14}C and total ^{15}N and microbial biomass ^{14}C of different aggregate size classes in a sand and a clay soil as influenced by different mechanisms of SOM protection, and 2) to measure soil organic matter protection as indicated by the rates of ^{14}C and ^{15}N mineralization after soil disaggregation.

MATERIALS AND METHODS

Soils

Soil labelled with ^{14}C. Two soils, a sand and a clay, were used; the properties of these soils are given in Table 1. The soils were used in a pot experiment under controlled ambient conditions where ^{14}C-labelled wheat straw was incorporated and spring wheat was sown twice. The fate of the label was traced in the soil during almost 300 days. At the end of the experiment, moist soil samples were used for soil fractionation.

Soils labelled with ^{15}N. The top 15 cm of a sand and a clay soil were used. Soil properties are given in Table 1. ^{15}N-labelled plant material was incorporated in 1992 and was allowed to decompose for almost 500 days under field conditions. Spring wheat was sown twice. To assess the changes in the chemical and physical properties of soil aggregates after cropping, the soils sampled by Matus (1995) were taken before the labelled plant material was added in 1990 and at the end of the experiment in 1992. The results of the soil fractionation are given by Matus (1995).

Physical measurements

Aggregate size distribution. The USDA scheme for primary particles (Gee and Bauder, 1986) was used: clay < 2 μm, silt between 2 and 50 μm and sand between 50 and 2000 μm. Two types of aggregates were distinguished: macro-aggregates (> 250 μm) and micro-aggregates (< 250 μm) (Tisdall and Oades, 1982). Soil labelled with ^{14}C was fractionated as described by Matus (1995).

Table 1. Some properties of the soils

Soil properties		Clay	Sand
Chemical			
Total C (%)		1.76	1.62
Total N (%)		0.19	0.11
C:N		9.30	14.70
$N\text{-}NH_4^+$ (mg kg^{-1})		2.10	0.40
$N\text{-}NO_3^-$ (mg kg^{-1})		14.50	13.50
P (mg l^{-1})		12.20	21.80
K (mg kg^{-1})		273.90	107.90
Mg (mg kg^{-1})		nd	116.00
$CaCO_3$ (%)		0.30	0.60
pH-KCl		6.98	7.22
Physical			
Particle size distribution (g kg^{-1})			
Clay	(< 2 µm) (%)	481	47
Fine silt	(2-20 µm)	289	34
Coarse silt	(20-50 µm)	174	265
Fine sand	(50-250 µm)	38	535
Coarse sand	(250-2000 µm)	18	119
Water holding capacity (%)			
-0.2 kPa		48	24
-25.0 kPa		40	21
-1555.0 kPa		22	5
Bulk density (g cm^{-3})		1.15	1.30

nd = not determined

Four aggregate size classes were obtained: coarse sand (250-2000 µm), fine sand (50-250 µm), fine silt (20-50 µm), fine silt plus clay (< 20 µm) and one unfractionated soil (< 2000 µm), referred to as "whole soil". The first three classes were recovered by filtration and the fine silt plus clay size fraction (< 20 µm) was separated from the soil suspension flocculated by adding 0.1 M $CaCl_2$ (Matus, 1995).

Distribution of clay content of aggregates. The clay content of each aggregate class was determined by standard analysis of particle sizes by the pipette method (Day, 1965). One replicate from each aggregate size class was used.

Chemical and biological measurements

Residual ^{14}C. Radioactivity of SOM was analyzed by a wet oxidation method. A duplicate subsample (1 g) was placed on the bottom of a screw glass tube (3.6 cm diameter, approximately 250 cm^3) with 5-ml of a concentrated acid-dichromate solution, consisting of 117.7 g H_2SO_4, 78.4 g H_3PO_4 and 160 g $K_2Cr_2O_7$. Each tube containing a glass vial with 10 ml 0.1 M NaOH was immediately sealed (teflon screw lid) and left standing for 30 min at room temperature. Thereafter, the tubes were heated for 2 hours at 160°C and left overnight. After precipitating the carbonate with excess $BaCl_2$ (0.75 M) the total amount of carbon was determined in a 5-ml aliquot of the NaOH trapping solution and titrated with a standard solution of 0.1 M Hcl to end point Ph 8.3 (Dalal, 1979). The labelled carbon was measured as trapped CO_2 in a 1-ml aliquot of the NaOH, diluted with 1-ml of demineralized water plus 10 ml of a scintillation cocktail (Insta-Gel, Packard Instruments Company) in a low potassium glass scintillation vial. Samples were counted in a liquid scintillation counter, Rackbeta II 1215, Wallac, programmed according to a previously prepared quenching curve. One or two hours counting were needed to reach a standard deviation less than 2%. Soluble ^{14}C lost during fractionation was measured from 20 g of whole soils extracted with 0.5 M of K_2SO_4 (1:4 = soil:solution). An aliquot of 0.4 ml of this extract was diluted with 1-ml of water and 12 ml of a scintillation cocktail and counted in a liquid scintillation counter as described above.

Microbial biomass ^{14}C and ^{14}C-mineralization rates. Prior to the determination of the microbial biomass by the chloroform fumigation-incubation method (Jenkinson and Powlson, 1976) the soils were moistened to -10 kPa with a nutrient solution containing $Ca(H_2PO_4)_2.H_2O$, K_2SO_4, $MgSO_4.7H_2O$ and $Ca(SO_4)_2.H_2O$. The amount of Ca, P, Mg, S and K added was equivalent to 372, 226, 48.6, 208.1 and 195.2 mg l^{-1} in solution, respectively. The soils were preincubated for 20 days at 20°C. Microbial biomass was calculated between day 20 and 30 as the difference between CO_2 evolved from fumigated and non-fumigated samples. No conversion factor was used (Ritz *et al.*, 1992). The accumulated CO_2 from non-fumigated samples between 0 and 20 days was used to calculate the C mineralization rates. The radioactivity of trapped $^{14}CO_2$ was measured as previously described.

Total ^{15}N. The ^{15}N concentration in the soil was determined using a Carlo Erba NA 1500 nitrogen analyzer linked to a SIRA 10 V.G. Isogas micromass spectrometer. The ^{15}N content of inorganic N extracts was determined after steam-distillation with $FeSO_4$ and Ag_2SO_4. The distillate was collected in dilute boric acid, neutralized with sulphuric acid and taken to dryness in a hot block. The NH_4^+-N was converted to N_2 by reaction with alkaline lithium hypobromite at the inlet to the mass spectrometer and analyzed for the isotopic ratio of ^{15}N:^{14}N.

^{15}N mineralization. The ^{15}N mineralization rates was determined between 20 and 40 days after the start of incubation as described by Matus (1995). The ^{15}N in inorganic forms (NO_3^--N and NH_4^+-N) was determined as above.

Statistical analysis

Analysis of variance (ANOVA; Genstat 5.0), and Student's t-test were used to assess the differences of means between treatments. The level of probability at which significant differences were found was set at 5%.

RESULTS

Physical properties of whole soil and various aggregate size classes

Aggregate size distribution. Aggregate size distributions from non-labelled soils from 1990 and soil labelled with ^{15}N from 1992 are presented by Matus (1995). Aggregate size distributions for ^{14}C-labelled soil are presented in Fig. 1a. Micro-aggregates < 20 μm in diameter and macro-aggregates between 250 and 2000 μm were more abundant in the clay than in the sand soil (Fig. 1a). These two fractions held more than 80% of the total soil mass in the clay soil. In the sand soil, however, micro-aggregates, 50-250 μm and macro-aggregates 250-2000 μm dominated (Fig 1a). In general, the distribution of aggregate sizes was similar to that presented by Matus (1995).

Distribution of clay content in each aggregate size class. The distribution of clay particles (< 2 μm) in the aggregates and whole soil (Fig. 1b) was clearly similar to the distribution of aggregate sizes in the clay soil (Fig. 1a), whereas in sand soil most of the clay was found in the < 20 μm fraction. This fraction also had the highest clay content in the clay soil. In sand soil, the lowest clay contents were found in the 50-250 μm size fraction, and in the clay soil in the 20-50 μm fraction.

Chemical and biological properties

Total ^{14}C. The concentration of total ^{14}C was higher in the < 20 μm fraction in the sand soil (Fig. 2). The labelled C in clay soil, however was almost uniformly distributed over the aggregate sizes, except for the significantly ($P < 0.05$) higher concentration of ^{14}C found in the 50-250 μm size fraction (Fig. 2).

Total ^{15}N. The largest concentration of ^{15}N were found in the clay (< 2 μm) and in the fine silt size

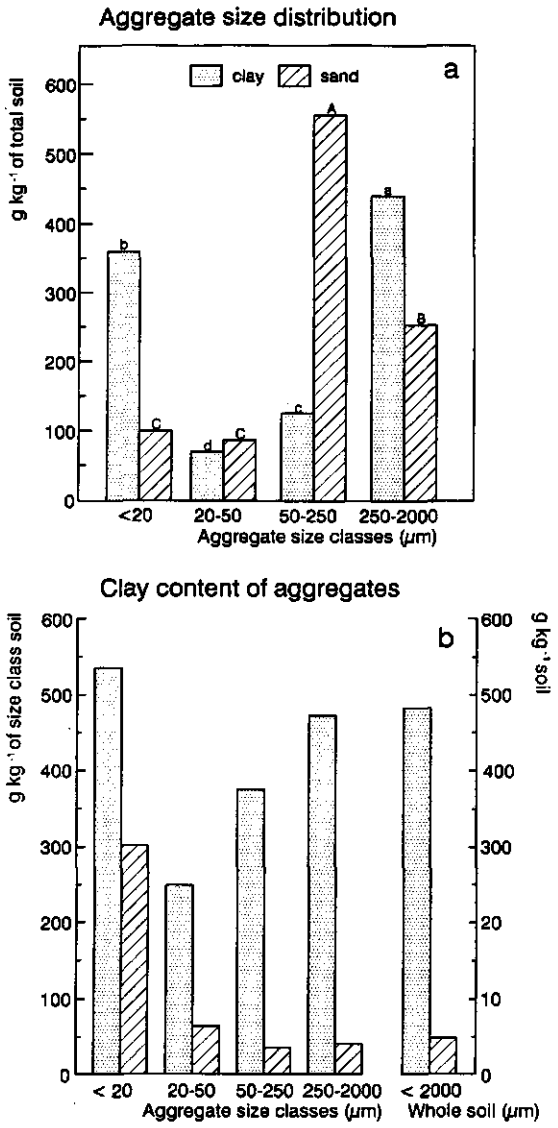

Fig. 1. Mass distribution of various aggregate size classes in clay and sand soils (a), and clay content of various aggregate size classes and whole clay and sand soils (only one replication available) (b). Bars carrying the same letter are not significantly different ($P < 0.05$).

fractions (2-20 μm) of the sand soil (Fig. 3). In the clay soil, although smaller differences were

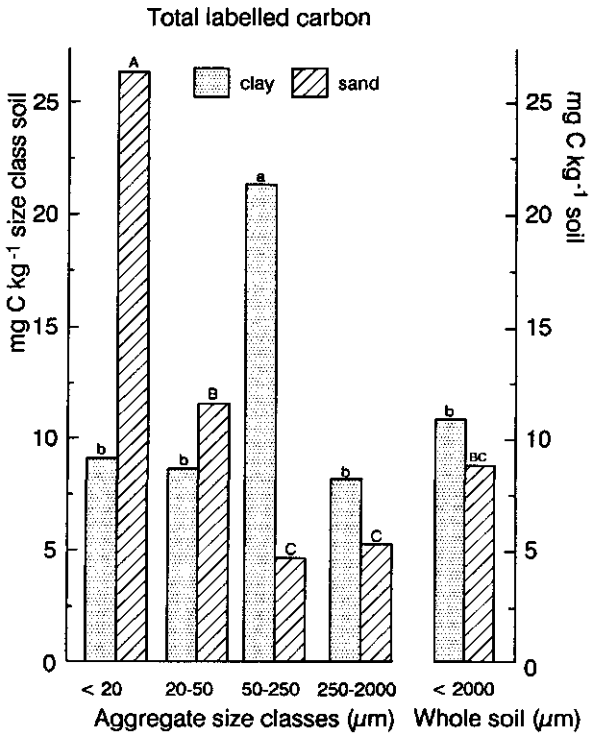

Fig. 2. Total ^{14}C concentration in clay and sand soil. Bars carrying the same letter are not significantly different ($P < 0.05$).

observed, the N concentration was significantly higher in the clay size fraction than in the other size classes and the whole soil (Fig. 3).

Non-labelled C mineralization rates and microbial biomass C. To compare the C mineralization rates and the C present in the microbial biomass of the various aggregates size classes and between those and whole soils, the results were expressed per unit of total C present in each aggregate size class or whole soil (Table 2). No significant differences in the percentage of aggregate C and whole C mineralized from each fraction were found (Table 2). The highest percentage of aggregate C present in the microbial biomass was observed in the 50-250 μm size fraction in the sand soil (Table 2), while the lowest value was observed in the < 20 μm fraction. In clay soil such differences amongst size classes were not observed, but the C in the microbial biomass was significantly higher in the whole soil than in any other of the size classes.

Labelled ^{14}C mineralization rates and microbial biomass ^{14}C. The ^{14}C mineralization rates and

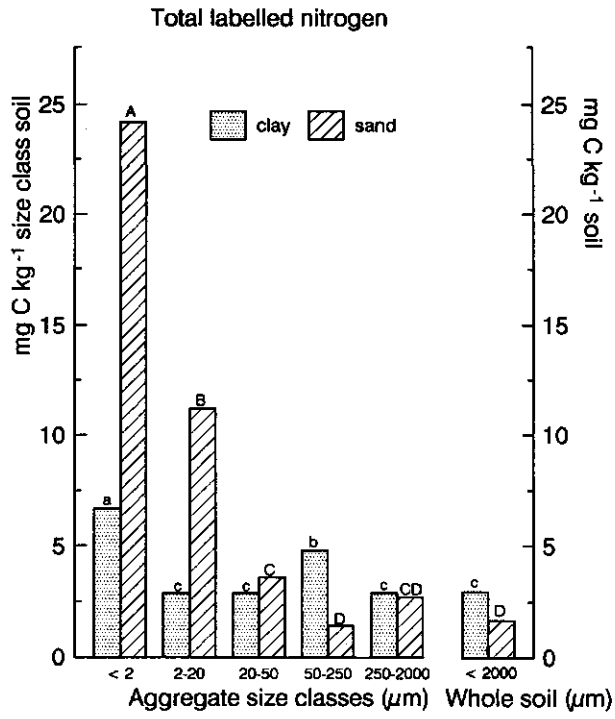

Fig. 3. Total ^{15}N concentration in clay and sand soil. Bars carrying the same letter are not significantly different ($P < 0.05$).

microbial biomass ^{14}C were expressed (Table 3) in the same way as non-labelled C. No significant differences were established in the percentage of aggregate labelled C or whole labelled C. The mineralization rates of ^{14}C from each aggregate size and whole soil were always higher than those from non-labelled C, especially in the < 20 μm fraction in sand soil, where the highest concentration of residual ^{14}C was observed (cf. Table 3 and Fig. 2). Significant differences ($P < 0.05$) in microbial biomass ^{14}C were observed (Table 3). Micro-aggregates 20-50 μm and 50-250 μm in diameter in sand soil were found to be lower in microbial biomass than any other fraction and whole soil. In clay soil these differences were even greater.

^{15}N-mineralization. No significant differences in ^{15}N mineralization rates between whole soils and amongst aggregate size classes were found (Table 4). The rates of labelled N mineralization obtained after 20 or 40 days of incubation were rather similar. Table 4 also shows the large standard errors of the data. The labelled ^{15}N mineralization in the 50-250 μm fraction for the

Table 2. Rates of C mineralization (g CO_2 kg^{-1} whole soil C or size class C day^{-1}) and microbial biomass C (g CO_2 kg^{-1} whole soil C or size class C) from various aggregate size classes and from whole sand and clay soil

Aggregate size classes (µm)	C mineralization	Microbial biomass C
Sand		
< 2000 (whole soil)	2.1	75.0
250-2000	1.6	60.0
50-250	2.2	235.6
20-50	0.9	75.8
< 20	1.0	12.7
Clay		
< 2000 (whole soil)	2.0	127.7
250-2000	0.9	80.8
50-250	1.2	63.8
20-50	1.2	87.4
< 20	0.5	85.4

L.S.D.[1] C-mineralization = 1.6
L.S.D. Microbial biomass = 32.6
[1] Least significant differences ($P < 0.05$)

Table 3. Rate of ^{14}C mineralization (g $^{14}CO_2$ kg^{-1} whole soil or size class ^{14}C) and microbial biomass ^{14}C (g $^{14}CO_2$ kg^{-1} whole soil or size class ^{14}C) from various aggregate size classes and from whole sand and clay soil

Aggregate size class (µm)	^{14}C-mineralization	Microbial biomass ^{14}C
Sand		
< 2000 (whole soil)	2.7	25.0
250-2000	3.6	15.2
50-250	2.7	1.4
20-50	3.1	7.8
< 20	3.3	23.8
Clay		
< 2000 (whole soil)	5.3	53.0
250-2000	2.9	36.1
50-250	2.0	10.4
20-50	2.8	0.0
< 20	1.8	22.2

L.S.D.[1] ^{14}C-mineralization = 3.5
L.S.D. Microbial biomass ^{14}C = 13.6
[1] Least significant differences ($P < 0.05$)

period 0-20 days did not agree with the absence of N mineralization found between 0-40 days of incubation in the same fraction.

Table 4. Rates of ^{15}N mineralization (g kg^{-1} whole soil ^{15}N or size class ^{15}N day^{-1}) from various aggregate size classes and from whole sand and clay soil in 1992 after different periods of incubation

Aggregate size classes (µm)	Days[1]	
	0-20	0-40
Sand		
<2000 (whole soil)	1.2	1.2
250-2000	2.7	1.9
50-250	1.7	1.4
20-50	1.8	1.5
<20	2.7	1.8
Clay		
<2000 (whole soil)	0.6	0.7
250-2000	1.2	1.0
50-250	0.4	0.0
20-50	0.2	0.5
<20	1.6	1.1

[1] rates obtained after 20 or 40 days of incubation
L.S.D. [2] N-mineralization = 1.5
[2] Least significant differences ($P < 0.05$)

Relationship between ^{14}C mineralization, ^{15}N mineralization rates and microbial biomass ^{14}C and clay content of aggregates. No significant correlations between clay content of aggregates and labelled ^{14}C mineralization rates were found in sand soil ($R^2 = 0.07$). Labelled microbial biomass showed a better correlation, but no significant differences were established ($R^2 = 0.66$). In clay soil, as in the sand soil, no significant correlations were observed. Labelled ^{14}C-mineralization yielded a lower coefficient of determination ($R^2 = 0.21$) than labelled microbial biomass ($R^2 = 0.68$). No significant correlations were established between clay content of aggregates and ^{15}N-mineralization rates obtained either in 20 days ($R^2 = 0.30$) or 40 days ($R^2 = 0.20$) of incubation in sand soil. In clay soil, however, a significant ($P < 0.05$) and positive correlation was found for the ^{15}N mineralization rates obtained in 20 days ($R^2 = 0.91$). No correlation, however, was obtained for the labelled N mineralization in 40 days ($R^2 = 0.43$).

DISCUSSION

Aggregate size distribution

The highest proportion of the total mass of the sand soil was found in the sand size fractions 50-250 µm and 250-2000 µm (Fig. 1a). In the present study the aggregate size distribution in sand soil was similar to the pattern obtained by Jocteur-Monrozier *et al.* (1991) and by Oades and

Waters (1991) in similar soils. The extent to which a total or limited soil disaggregation can influence the SOM distribution in each aggregate size class was discussed by Matus (1995).

Effect of soil texture on the distribution of total ^{14}C and total ^{15}N over various aggregate size classes

The summed amount of ^{14}C from each aggregate size class were found to be lower than the concentration of ^{14}C in whole clay or sand soil and therefore losses during the fractionation procedure occurred. The recoveries were 94% and 86% of total ^{14}C. Total ^{14}C in whole sand was 8.7 and in whole clay 10.8 mg ^{14}C kg^{-1}. These differences were in agreement with the 0.8 mg ^{14}C kg^{-1} soil measured as water-soluble ^{14}C in the soil solution from whole clay soil. Christensen and Sørensen (1985) also reported smaller losses of water-soluble C (between 2% and 9% of residual ^{14}C left in soil), even when they used ultrasonic energy to disperse the soils.

Total labelled C concentrations were about 3 times higher in the fine silt plus clay size fraction (< 20 μm) in the sand soil than in the clay soil (Fig. 2). The ^{14}C concentration ratio ($^{14}C_r$), i.e. the concentration of ^{14}C in the fine silt plus clay size fraction (< 20 μm) divided by the concentration of ^{14}C of the whole soil, was 3 in sand soil and 1 in clay soil. When this concentration is expressed per unit of clay, the < 20 μm size fraction was about 5 times more concentrated in sand than in clay soil. These results are in agreement with the data presented for non-labelled C and a similar conclusion can be drawn: the clay and silt size fraction were much more concentrated with recently formed SOM in coarse- than in fine-textured soils. A similar conclusion was drawn by Christensen and Sørensen (1985). This means that more SOM per unit of clay surface is found in sand than in clay soil (Hassink *et al.*, 1995). The distribution of total ^{15}N over the aggregate sizes was in agreement with the data presented for labelled C. Although the ^{15}N was measured in the clay (< 2 μm) size fraction instead of the < 20 μm size fraction (as with residual labelled C), the largest concentration of ^{15}N were observed in the clay and in the fine silt (2-20 μm) size fractions in the sand soil (Fig. 3), whereas for the clay soil ^{15}N was found to be highest in the clay (< 2 μm) size fraction. The ^{15}N concentration ratio of clay size fraction ($^{15}N_r$), i.e. the contentration of ^{15}N in the clay size fraction divided by the concentration of ^{15}N of the whole soil, was 14 in sand soil and 2.2 in clay soil. The results confirm that recently produced soil organic matter is similarly distributed as non-labelled SOM over various aggregate size classes (Matus, 1995).

Effect of clay content on non-labelled C mineralization rate and non-labelled microbial biomass C of various aggregates size classes

No significant differences in C mineralization rates were found (Table 2). The amount of

aggregate C present in microbial biomass (Table 2) was found to be too high with respect to the usual amount measured in soils (17-37 g C kg^{-1} soil C) (Jenkinson and Powlson, 1976) and significantly ($P < 0.05$) higher in whole clay soil than in any of the size classes. These high figures may have been the result of:

1) Fumigation with chloroform. It seems that the chloroform penetration is different in disaggregated soils than in the whole soils. More C was present in the microbial biomass from the sum of each fraction than in the microbial biomass of the whole soil.

2) Sampling errors. Standard errors were large, because the amount of soil used (2 g) was smaller than the usual quantity (250 g) (Jenkinson and Powlson, 1976).

Matus (1995) reported that the C mineralization rates in the same sand soil studied here and microbial biomass C showed an inverse relationship with the clay content of aggregates, while in a clay soil no similar relationships were established. The figures for C mineralization and microbial biomass C presented here were also included in his data. Matus (1995) suggests that in sand soil both C and N mineralization rate were strongly protected from biodegradation in the < 20 μm size fraction, whereas SOM associated with sand size fractions (> 50 μm) was not.

Effect of clay content on ^{14}C and ^{15}N mineralization rates and microbial biomass ^{14}C of various aggregate size classes

Contrary to the findings of Matus (1995) no similar relationship between the clay content of aggregates and labelled ^{14}C-mineralization and ^{15}N-mineralization were observed in sand soil. The results suggest that in sand soil recently formed soil organic matter was less protected from biodegradation than older SOM. In clay soil, although a positive correlation was found between clay content of aggregates and ^{15}N-mineralization calculated for 20 days of incubation, no significant correlation was established for the rates estimated for 40 days of incubation. The present results did not support the idea that recently formed soil organic matter in clay soil was protected against decomposition in small pores, because no correlation was established for ^{14}C mineralization in clay soil. Matus (1995) also found a positive and significant correlation between clay content of aggregates with non-labelled N mineralization and with the proportion of pores, < 1.2 μm in diameter in clay soil. This could indicate that SOM was physically protected in small pores as suggested before by Hassink *et al.* (1993). However, similar correlations of clay content of aggregates with non-labelled C mineralization or non-labelled microbial biomass C were neither established by Matus (1995). He concluded that it is not clear yet to what extent the organic materials are entrapped in small pores, protected against decomposition in clay soil, because the positive correlation with N mineralization rate may simply be the consequence of the low C:N ratio in the clay fraction in the clay soil (Matus, 1995). On the other hand, Tables 2 and 3 show that non-labelled C and ^{14}C mineralization rates from each fraction and whole soils were

comparable when expressed per unit of total C or total ^{14}C of each fraction or whole soil. Table 3 shows that ^{14}C mineralization rates were higher than non-labelled C mineralization rates. These differences were especially greater in the < 20 μm size fraction, where SOM was found to be more protected in sand soil (Table 2 and 3). The higher values in both clay and sand soil indicate that labelled recently formed organic C decomposes faster than the non-labelled total C (Christensen and Sørensen, 1985) reflecting that a greater fraction of the recently produced SOM was not protected against biodegradation. This conclusion was also supported when ^{15}N-mineralization rates were compared with non-labelled N-mineralization rates obtained by Matus (1995) (data not shown).

CONCLUSIONS

Soil texture and structure play major roles in determining soil organic matter protection, but are less important in the protection of newly formed soil organic matter because no relationship of the C mineralization rates and microbial biomass C of newly formed SOM with the clay content of aggregates was found in sand soil. There was no relationship between clay content of aggregates with labelled C mineralization rate and labelled microbial biomass C in clay soil, except for a positive correlation established for ^{15}N mineralization at 20 days of incubation. It is not clear yet to what extent labelled SOM in clay soil is physically protected against decomposition in small pores. In both soils, higher rates of labelled C mineralization as compared with non-labelled C mineralization indicated that labelled recently formed organic C decomposed faster than the non-labelled total C.

Acknowledgements - I thank K. Kampen, A. Lahr and M. Wolters for their technical assistance. I also thank Ir. J. Hassink and Dr. K.B. Zwart for their helpful discussion and comments on the manuscript. I especially thank Dr. L. Brussaard and Dr. A.P. Whitmore for their support and useful discussions during this investigation. Part of this work was supported by grant N° P806 under the sandwich PhD Fellowship Programme of Wageningen Agricultural University and by a grant under the Presidential Fellowship Programme of the Agencia de Cooperación Internacional (AGCI) del Ministerio de Planificación y Cooperación de Chile (MIDEPLAN).

REFERENCES

Craswell, E.T. and Waring, S.A. (1972) Effect of grinding on the decomposition of soil organic matter -II. Oxygen uptake and nitrogen mineralization in virgin and cultivated soils. *Soil Biology & Biochemistry* 4, 435-442.

Christensen, B.T. and Sørensen, L.H. (1985) The distribution of native and labelled carbon between soil particles size fraction isolated from long-term incubation experiments. *Journal of Soil Science* **36**, 219-229.

Dalal, R.C. (1979) Simple procedure for the determination of total carbon and its radioactivity in soils and plant materials. *Analyst* **101**, 151-154.

Day, P.R. (1965). Particle fractionation and particle-size analysis. In *Methods of Soil Analysis, Part 1, Physical and Mineralogical Properties, Including Statistics of Measurements and Sampling* (C.A. Black, Ed.), pp. 545-566. ASA Publ., Madinson, WI, USA.

Elliot, E.T. and Coleman, D.C. (1988). Let the soil work for us. IN *Ecological Bulletins* **39**, 23-32, Copenhagen.

Gee, G.W. and Bauder, J.W. (1986) Particle-size analysis. In *Methods of soil Analysis, Part I, Physical and Mineralogical Properties*, (A. Klute Ed.), pp. 383-441. 2nd Edition ASA and SSSA Publ., Madison, WI, USA.

Hassink, J. Bouwman, L.A., Zwart, K.B., Bloem, J and Brussaard, L (1993) Relationships between soil texture, physical protection of soil organic matter, soil biota, and C and N mineralization in grassland soils. *Geoderma* **57**, 105-128.

Hassink, J., Matus, F.J., Chenu, C. and Dalenberg, J.W. (1995) Interaction between soil biota, soil organic matter and soil structure. *Advances in Agroecology* (Submitted).

Jenkinson D.S. and Powlson, D.S. (1976) The effects of biocidal treatments on metabolism in soil -V. A method for measuring soil biomass. *Soil Biology & Biochemistry* **8**, 209-213.

Jocteur-Monrozier, L., Ladd, J.N., Fitzpatrick, R.W., Foster, R.C. and Raupach, M. (1991) Components and microbial biomass content of size fractions in soils of contrasting aggregation. *Geoderma* **49**, 37-62.

Matus, F.J. (1995). The distribution of soil organic matter of various aggregate size classes in arable soil. I. Relationships between clay content of aggregates of a sand and a clay soil and carbon mineralization, nitrogen mineralization and microbial biomass carbon. *Soil Biology & Biochemistry* (submitted).

Oades, J.M. and Waters, A.G. (1991) Aggregate hierarchy in soils. *Australian Journal of Soil Research* **29**, 815-828.

Ritz, K., Griffiths, B.S. and Wheatly, R.E. (1992) Soil microbial biomass and activity under a potato crop fertilized with N with and without C. *Biology and Fertility of Soils* **12**, 265-271.

Rovira, A.D. and Greacen, E.L. (1957) The effect of aggregate disruption on the activity of microorganisms in the soil. *Australian Journal of Agricultural Research* **8**, 659-673.

Sørensen, L.H. (1983) The influence of stress treatments on the microbial biomass and the rate of decomposition of humified matter in soils containing different amounts of clay. *Plant and Soil* **75**, 107-119.

Tisdall, J.M. and Oades, J.M. (1982) Organic matter and water-stable aggregates in soils. *Journal of Soil Science* **33**, 141-163.

Van Gestel, M., Ladd, J.N. and Amato, M. (1991) Carbon and nitrogen mineralization from two soils of contrasting texture and microaggregates stability: influence of sequential fumigation, drying and storage. *Soil Biology & Biochemistry* **23**, 313-322.

Van Veen J.A., Ladd J.N. and Amato M. (1985) Turnover of carbon and nitrogen through the microbial biomass in a sandy loam and a clay soil incubated with [^{14}C(U)]glucose and [^{15}N](NH_4)SO_4 under different moisture regimes. *Soil Biology & Biochemistry* **17**, 747-756.

CHAPTER 7

THE DECOMPOSITION OF WHEAT AND CLOVER RESIDUES IN SOIL: MEASUREMENTS AND MODELLING

A.P. Whitmore and F.J. Matus

DLO Research Institute for Agrobiology and Soil Fertility, P.O. Box 129, 9750 AC Haren, The Netherlands

Key words: Crop residues, decomposition, modelling

Summary - A computer model is described that is able to trace the fate of nitrogen added to field soils in crop residues. Clover and wheat residues labelled with ^{15}N were added to a clay and a sand soil and the fate of the label traced over a period of almost 16 months under field conditions. Using a simple function to retard the decomposition of crop residues according to how much fibrous tissue they contain, the model was able to estimate the organic N remaining in soil, and the mineral N and microbial biomass N derived from the crop residues. It proved necessary, however, to postulate the existence of a pool of organic matter derived from crop residues that was more labile than native humus in soil. The results from both clover and wheat residue decomposition in the clay and in the sand soils could be simulated by the same organic matter turnover model using the initial C, N and fibre content of the two residues without any changes in the parameter values, except the moisture content of the soils.

INTRODUCTION

Computer simulation models of carbon or nitrogen turnover in soil have enjoyed much success in recent years in their ability to estimate the mineralization of C or the mineralization or immobilization of N (*e.g.* Bradbury *et al.*, 1993, Verberne *et al.*, 1990, Parton *et al.*, 1987). The relative amounts of C and N, or their ratio in organic material or crop residues added to soil, have proved an excellent indicator of the speed of release of N or C and serve as a predictor of whether N is immobilized or mineralized. Nonetheless, not all the variation in residue decomposition is accounted for with this simple hypothesis and a number of authors have noticed that the residual variation is well correlated with the fibre content of the organic materials (*e.g.* Matus, 1995; Whitmore and Groot, 1995; Vanlauwe *et al.*, 1993). Here we show that a simple adaptation of a computer simulation model to take account of the lignin, cellulose and hemi-cellulose contents of crop residues is able to produce very good simulations of the amount of

nitrogen remaining behind in soil as the residues decompose. The same model satisfactorily estimated the amount of labelled N found in the microbial biomass and in the mineral form in soil.

MATERIALS AND METHODS

Incubation experiments

The shoots (12.7 g) or roots (3.2 g) from each of clover or wheat residues labelled with ^{15}N were added to either 6100 g of sand soil (containing 5% clay) or 5400 g of clay soil (containing 48% clay) in a series of field incubation experiments (Matus, 1995). All weights are expressed on a dry matter basis. The plant materials differed in their fibre content; these and other relevant chemical properties are given in Table 1. Dried residues were mixed with field moist soil and the mixture confined within PVC cylinders (20 cm in diameter by 30 cm in length) that were kept in the field under ambient conditions for about 500 days beginning in May 1991. The fate of the ^{15}N-label derived from these plant materials has been described in detail by Matus (1995) who has presented the distribution of label with depth in the soil. Since virtually no organic ^{15}N was found below 15 cm the results simulated here are for the top 15 cm of soil in each cylinder only.

Table 1. Chemical properties of the wheat and clover

Residue	C (%)	N (%)	Cellulose + Hemi-cellulose (%)	Lignin (%)
Wheat shoot	35.1	3.6	36.6	3.7
Wheat root	32.8	1.8	59.7	4.0
Clover shoot	33.3	4.4	20.5	3.3
Clover root	32.0	3.5	31.4	6.9

Computer simulations

Whitmore and Groot (1995) have described a simple computer simulation model of the decomposition of crop residues; in this model the decomposition of both crop residues and humified organic matter follows first-order kinetics, the only difference being that the turnover times differ. A proportion α of the carbon from the decomposing residues becomes biomass and a proportion β becomes humified (Bradbury *et al.*, 1993), the remainder $(1-\alpha-\beta)$ is lost as CO_2. Whether nitrogen is mineralized or immobilized depends on the C:N ratios of the residues (Z), biomass (X) and humified organic matter (Y). Equation (1) shows the necessary condition for mineralization directly from crop residues.

$$\frac{1}{Z} > \left(\frac{\alpha}{X} + \frac{\beta}{Y}\right) \quad (1)$$

Whitmore (1994) has described a simple adaptation of the kinetic law describing the turnover of the microbial biomass in soil. The amount of biomass B remaining in soil as an initial amount B_0 turns over in time t is described by equation (2)

$$B = \frac{B_0}{1 + kB_0 t} \quad (2)$$

A combination of these models was used to estimate the fate of ^{15}N-labelled crop residues in soil. Because the cylinders containing the residues were kept outside in the field, Burns' leaching equation (1976) was used to estimate the loss of mineralized N from the soil cylinders assuming that the mineralization of N was uniform throughout the 15 cm length and assuming that all ammonified N was rapidly nitrified. If P is the percolating excess rainfall (rain minus evaporation) falling in time t, h is the depth of soil (15 cm here) and θ is the volumetric moisture holding capacity of the soil.

$$N_{leached} = \left\{\frac{P}{P + \theta}\right\}^{\frac{h}{Z}} \quad (3)$$

The fibre content was assumed to retard the decomposition of the crop residues. As the easily decomposable parts of the residues break down the fibrous parts become relatively more concentrated and retard the decomposition process to a greater and greater extent. The retardation factor f_R is calculated as follows:

$$f_R = \frac{k_R F_0}{(1 + k_R F_t)} \quad (4)$$

where k_R is a constant and F_0 is the initial concentration of fibre in crop residues (cellulose plus lignin in these simulations) and F_t the concentration at time t. Moisture and temperature are assumed to modify the rates of turnover and organic matter in the same way as described by Bradbury *et al.* (1993). The factor f_R reduces the rate of residue decomposition along with f_T (effect of temperature) and f_W (effect of moisture) as follows:

$$k^1 = f_R * f_W * f_T * k \quad (5)$$

note that both f_T and f_R can be greater than unity (*e.g.* the temperature can exceed the base temperature of 9°C) and that there is no interaction between these rate-modifying factors.

Daily values of the rainfall and soil temperature were recorded on site. Evaporation was not recorded locally and values for the long-term mean were taken from Eelde airport, 5 km distant. Occasional missing values among the rain and temperatures were also supplied from the mean monthly value at Eelde.

RESULTS

The residual ^{15}N remaining in soil and the simulations of these data are presented in Fig. 1. Here, all the organic ^{15}N estimated with the model to be remaining in soil microbial biomass and

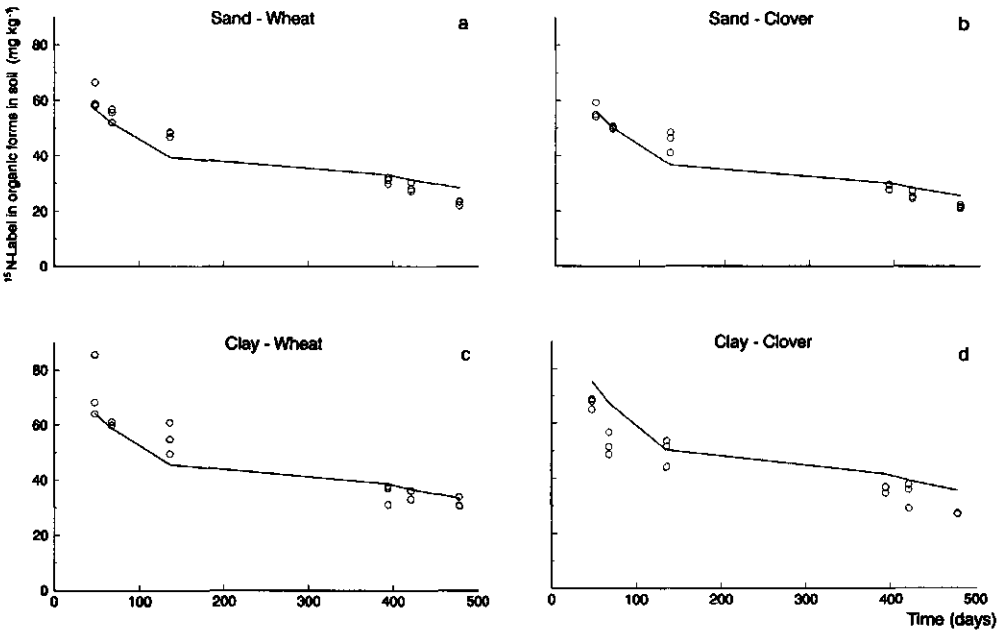

Fig. 1. Measured and modelled amounts of ^{15}N-label remaining in soil in organic forms: (a) sand soil with wheat, (b) sand soil with clover, (c) clay soil with wheat, (d) clay soil with clover.

humus has been totalled. Measurements of total ^{15}N-label in soil have had mineral ^{15}N subtracted. It is clear that the model simulates the retention of ^{15}N-label in organic forms in soil very well indeed. Of particular interest is the fact that results from both the clay and sand soils and with clover and wheat residues can be simulated by the same model without having to make any changes in the parameter values apart from θ which determines the moisture holding capacity of the soil. The behavior of the N derived from crop residues can be described with exactly the same organic matter turnover model.

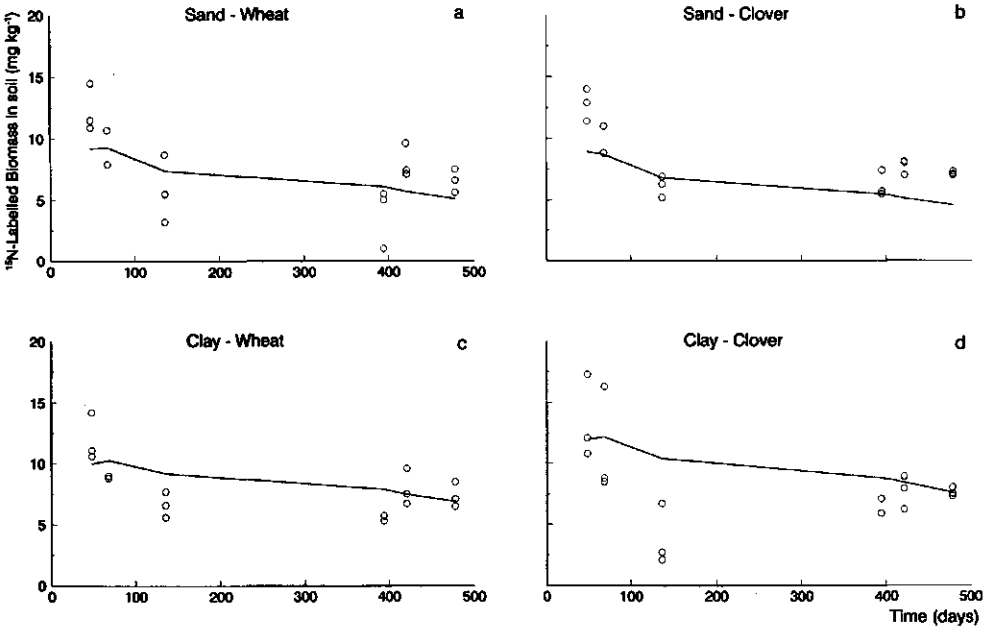

Fig. 2. Measured and modelled amounts of ^{15}N-label remaining in soil in the microbial biomass: (a) sand soil with wheat, (b) sand soil with clover, (c) clay soil with wheat, (d) clay soil with clover.

Fig. 2 shows the amounts of ^{15}N-label recovered in the microbial biomass. Relatively large amounts of microbial biomass were found in these soils and the amounts decline throughout the experiment (also in the control). The model does not predict a sharp increase in the amount of ^{15}N-label found in the biomass but rather a gradual enrichment over the first month or so followed, as shown here, by an equally gradual decline. One set of outlying measurements on day 67 has been excluded, however. The soils used here were taken shortly after harvest and they already contained large amounts of crop residues. The model predicts that the total biomass N in the control soil declines over the course of the experiment as indeed was found with the measured values (data not shown). As such it is not surprising therefore that the model predicts no rapid increase in the microbial biomass ^{15}N.

The ^{15}N-label found in mineral forms in soil is shown in Fig. 3, together with the simulations. Here the most striking feature is the rapid decline to almost nothing and increase in mineral N between the first and third sampling times (48 and 135 days). Although these measurements took place in the early summer and autumn a great deal of ^{15}N-label was lost from top 15 cm of soil. Matus (1995) did not recover a great deal more in the 35 cm of soil below these cylinders. Assuming that all the loss was leaching, Burns' (1976) leaching equation has simulated the pattern

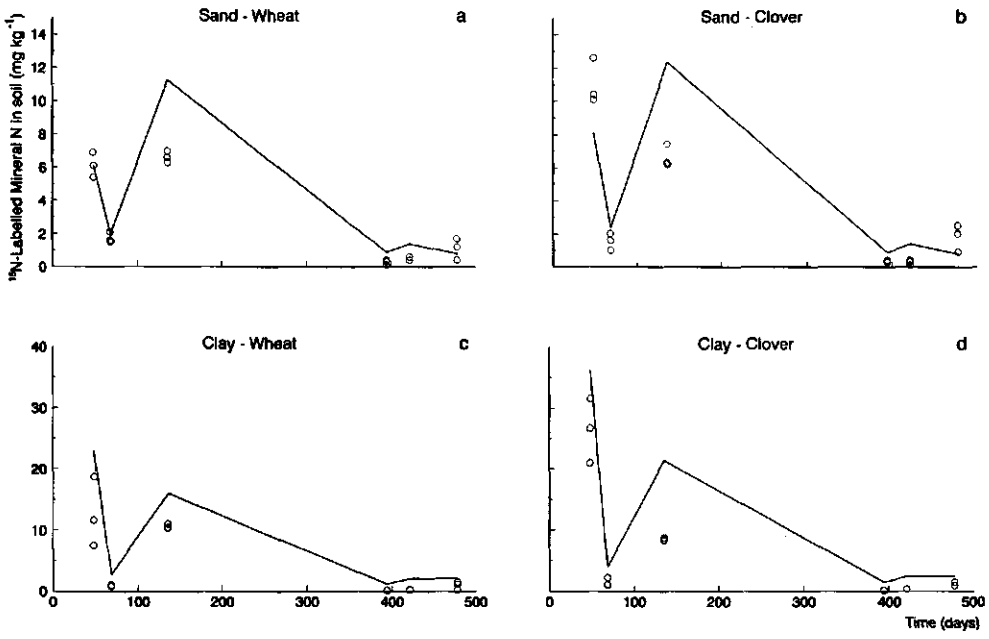

Fig. 3. Measured and modelled amounts of ^{15}N-label remaining in soil as mineral N: (a) sand soil with wheat, (b) sand soil with clover, (c) clay soil with wheat, (d) clay soil with clover.

of loss very well indeed. Even so some of the loss undoubtedly was denitrification and it is clear in the clay soil, for example (Figs. 3c and d), that the model overestimated the amount of mineral N in the soil on the first sampling date; more denitrification might reasonably be expected in this soil under these conditions. Careful examination of the data in all figures reveals that much ^{15}N-label is missing. Nitrogen equivalent to 50 mg N per 100 g soil was lost during the first 48 days. During this period some 90 mm of rain fell, much of it during thunderstorms. Indeed on this first sampling day 21.9 mm of rain fell, which probably accounts for much of the variation found in the measurements made on the clay soil.

Although almost all of the parameters in the organic matter model are as previously described (Whitmore and Groot, 1995; Bradbury *et al.*, 1993), we could not obtain the fits shown in these diagram for the decline in ^{15}N-label in organic forms in soil without increasing the rate of turnover of ^{15}N-labelled humified substances in soil. The value of this rate constant needed to be about three times faster than was calculated for the previous articles where either non-labelled materials were studied or the long-term fate of resistant crop residues was traced (mature wheat roots and stubble). One novel element in these simulations is the use of a 2nd-order rate law to

describe the decomposition of the microbial biomass. Equally good simulations of the retention of ^{15}N-label in organic forms in soil as those shown in Fig. 1 were obtained using a 1st-order rate law in which the microbial biomass turned over more slowly during the initial months of the experiment; the amounts of microbial biomass N simulated were unreasonably high, however. In effect the biomass was fulfilling the same function of storing ^{15}N-label and releasing it more quickly than the humified organic matter. Either way it seems likely that an active fraction of organic matter was produced from these easily decomposable crop residues and that this itself is relatively rapidly re-mobilized in soil.

Acknowledgements - F.J. Matus gratefully acknowledges the support of a Presidential Fellowship Programme from Agencia de Cooperación Internacional (AGCI) del Ministerio de Planificación y Cooperación de Chile (MIDEPLAN) and of the Sandwich-Programme Fellowship (P806) from the Agricultural University, Wageningen, The Netherlands.

REFERENCES

Bradbury N J, Whitmore A P, Hart P B S and Jenkinson D S 1993 Modelling the fate of nitrogen in crop and soil in the years following application of ^{15}N-labelled fertilizer to winter wheat. *J. Agric. Sci., Camb.* **121**, 363-379.

Burns I G 1976 Equations to predict the leaching of nitrate uniformly incorporated to a known depth or uniformly distributed throughout a soil profile. *J. Agric. Sci., Camb.* **86**, 305-313.

Matus F J 1995 Effects of soil texture, soil structure and cropping on the decomposition of crop residues. II: Residual organic N, microbial biomass N and inorganic N derived from decomposing ^{15}N-labelled wheat and clover in a clay soil and sand soil. Submitted to *Soil Biol. Biochem.*

Parton W J, Schimel D S, Cole C V and Ojima D S 1987 Analysis of factors controlling soil organic matter levels in great plains grasslands. *Soil Sci. Soc. Am. J.* **51**, 1173-1179.

Verberne E L J, Hassink J, de Willigen P, Groot J J R and van Veen J A 1990 Modelling organic matter dynamics in different soils. *Neth. J. Agric. Sci.* **38**, 221-238

Vanlauwe B, Dendooven, L and Merckx R 1994 Residue fractionation and decomposition: The significance of the active fraction. *Plant and Soil* **158**, 263-274.

Whitmore A P and Groot J J R (in press) The mineralization of N from finely or coarsely chopped crop residues: measurements and modelling. *European J. Agron.*

Whitmore A P 1994 Alternative kinetic laws to describe the turnover of the microbial biomass. *Plant and Soil* (submitted)

CHAPTER 8

GENERAL DISCUSSION AND CONCLUDING REMARKS

Practical models and mechanistic studies

Chapter 2 presented a practical model to estimate the mineralization of nitrogen (N) from the input of crop residues in different agricultural systems in Chile. The model was built on published data showing that recently formed soil organic matter (SOM) derived from crop residues in the long term (> 1 year) decomposed at similar rates, apparently independent of the nature of the residue and soil types considered. The model predicted the mineralization of N reasonably well. Mechanistic models often perform no better and sometimes worse than practical models like this one in the estimation of N mineralization (De Willigen, 1991). Mechanistic models may be applicable to a wide range of conditions, but need detailed information and the results are often sensitive to specific parameter values. Practical models require few parameters, but are applicable to a limited range of conditions (De Willigen, 1991). Our model (chapter 2) estimated the N mineralization on an annual basis from the organic N input in a crop rotation. Cropping, residue type, soil texture and soil structure were not considered. These factors, often reported to influence crop residue and SOM decomposition in soil in the short term (< 1 year) in soil and, *mutatis mutandis,* to influence SOM physical protection, were reported in this thesis and are discussed here.

Effects of cropping and plant residue type on decomposition of residues and residual soil organic matter

Cropping neither influenced the total amount of ^{14}C nor microbial biomass ^{14}C and the residual ^{14}C in humus forms as measured by acid hydrolyzation (chapter 3) at any time during incubation. No significant differences in residual organic ^{15}N, microbial biomass ^{15}N and inorganic ^{15}N derived from clover and wheat residues as affected by cropping were observed (chapter 4). It has been well documented that the decomposition of labelled plant residues in soil is retarded by the presence of living plants, both under field (e.g. Führ and Sauerbeck, 1968; Jenkinson 1977) and under laboratory conditions (e.g. Reid and Goss, 1982). It has also been reported that the presence of roots induces a higher microbial activity in the rhizosphere by exudation of organic compounds and stimulates the decomposition of SOM (Helal and Sauerbeck, 1986) and the mineralization of nitrogen (Hart *et al.*, 1979; Jingguo and Bakken, 1989). The retardation of the decomposition of labelled plant residues may occur soon after incorporation and continue during the course of decomposition (e.g. Jenkinson, 1977) or may be small initially, disappearing later

(e.g. Führ and Sauerbeck, 1968). The reduction in the decomposition of labelled plant residues in the presence of living roots has been related to the C:N ratio of the incorporated crop residues in soil, the effect occurring with C:N ratios > 20 (e.g. Shields and Paul, 1973; Jenkinson, 1977; Sallih and Bottner, 1988; Beck, 1994). The results presented in the figures 1 of chapters 3 and 4 support the hypothesis that soil microorganisms prefer crop residues with a low C:N ratio over root-derived substrate. An effect of roots of wheat plants on the ^{14}C or ^{15}N present in microbial biomass was not evident either (figures 2 of chapter 3 and chapter 4). From more than 60 papers examined by Fog (1988) it was concluded that not only the C:N ratio of incorporated organic matter affected N mineralization but also the microflora present, competition between decomposers, suppression of microbial activity by N addition and preferences for labile rather than more recalcitrant organic compounds also play a role. In Chapter 4 ^{15}N-labelled clover residues, containing 4.2% N were shown to decompose significantly faster than wheat materials, containing 3.2% N, during the first 48 days of decomposition. This difference was sustained during the total incubation period of 477 days. Since (1) both residues had a low C:N ratio (about 10), but clover contained 50% more fibre (cellulose and hemicellulose) than wheat and (2) the decline of residual organic ^{15}N either with or without N fertilization was similar, the difference in decomposition between clover and wheat can be explained from the difference in fibre content rather than from microbial N immobilization or effective competition for N between plants and microbes in the rhizosphere.

Effects of soil disruption on decomposition of residual soil organic matter

Disruption of the soil did not affect the decline of residual ^{14}C or residual ^{15}N derived from labelled residues in either the clay or sand soil (chapters 3 and 4). On the other hand, figures 3a-d of chapter 5 and figures 2 and 3 of chapter 6 show that non-labelled SOM was distributed similarly to recently labelled SOM within soil aggregates. The soils used in this study have been under long-term arable management. It is possible that the macro-aggregates (> 250 μm) and the micro-aggregates (< 250 μm) left in soil after continuous tillage are not disrupted by further tillage. In addition, mixing of the soil may not have been sufficiently disruptive to break up the remaining macro- and micro-aggregates, especially in the clay soil. In chapter 6 it was demonstrated that a large proportion of recently formed SOM of micro- and macro-aggregates was not physically protected against decomposition at all. Therefore, if no effect of soil disruption on soil aggregates left after long-term cultivation and if recently formed SOM is not physically protected, mixing could hardly affect the decomposition of recently formed SOM.

Effects of soil texture on the distribution and physical protection of soil organic matter

The distribution of total C, total N and microbial biomass C of macro- and micro-aggregates was described in chapter 5. The rates of C and N mineralization after soil dispersion were used as indicators of the degree of SOM protection in aggregates. Soil organic matter in both soils was associated to a greater extent with the fine silt (2-20 μm) and clay (< 2 μm) size fractions than with any of the other fractions. Soil organic matter was found to be much more concentrated in the clay size fraction of the sand soil than in that of the clay soil. The results were consistent with the hypothesis that more SOM is stabilized per unit of clay in sand than in clay soil (Hassink *et al.*, 1995). Microscopic studies have revealed that in clay soil aggregates are a massive assembly of clay particles forming dense lattices, whereas in the aggregates in sand soil clay particles are loosely arranged (Oades and Waters, 1991; Hassink *et al.*, 1995). Although the < 20 μm fraction in sand soil had the highest SOM concentration and clay content, there was an inverse relationship between the clay content of the aggregates and the microbial biomass C and with the rates of C and N mineralization in sand soil. Such relationships were not found in the clay soil, except for a positive, significant correlation between clay content of the aggregates and N mineralization. The results suggest that in sand soil, SOM was protected by silt and clay particles, presumably by adsorption. Soil organic matter can be physico-chemically adsorbed onto the surface of clay minerals (Edwards and Bremner, 1967; Tisdall and Oades, 1982), encrusted by clay particles (Tisdall and Oades, 1982), or possibly entrapped in small pores, especially in clay soils (Hassink *et al.*, 1993). In clay soil the positive correlation with N mineralization may simply be the consequence of the low C:N ratio observed in the silt plus clay fraction (< 20 μm) which coincided with the highest clay content. Hence, it is not yet clear to what extent SOM is protected in small pores in the clay soil (chapter 5).

Chapter 6 described the distribution of total ^{14}C, total ^{15}N and microbial biomass ^{14}C of macro- and micro-aggregates and the rates of ^{14}C and ^{15}N mineralization to assess whether recently formed SOM behaves similarly to non-labelled SOM. The distribution of labelled SOM was similar to that of non- labelled SOM in the various aggregate size classes. Similarly to what was found for unlabelled SOM, labelled SOM was more concentrated in the < 20 μm size fraction in both clay and sand soil, but in the sand soil the concentration of SOM per unit of clay was higher. Contrary to what was found in the study of unlabelled C and N, no correlation was established between the clay content of aggregates and the mineralization rates of ^{14}C, ^{15}N and microbial biomass ^{14}C in sand soil. In clay soil ^{15}N mineralization was correlated only after 20 days of incubation. In sand soil recently produced SOM was apparently less physically protected against biodegradation than older SOM. This conclusion is supported by the higher rate of ^{14}C mineralization as compared with the non-labelled C mineralization, indicating that newly formed SOM decomposes faster than non-labelled native SOM. The results for clay soil were again not

conclusive as regards the protection of recently formed SOM.

Effects of soil texture on C and N mineralization rates

Although there was a considerable difference between the texture of the sand and the clay soil studied, the decomposition of labelled plant residues was found to be very similar (chapters 3 and 4). Chapter 4 show that the measured decomposition rates of soil organic N from these soils and other soils obtained from the literature leveled off in soil after two years of decomposition and similar rates of decomposition were observed as the period of decomposition progressed. Comparable results were obtained by Kolenbrander (1974). Later, experiments with ^{14}C- and ^{15}N-labelled crop residues confirmed this finding (Ladd *et al.*, 1981; Sauerbeck and Gonzales, 1977; Jenkinson, 1977). However, Amato and Ladd (1992) found a significant positive correlation between the clay content of soil and the residual organic ^{14}C from legumes decomposing in 23 Australian soils with pH > 7. When the statistical analysis was confined to soils of neutral and acid pH (6-7), however, no significant correlation was established. Amato and Ladd (1992) explained the observed differences by the effect of pH. In the present thesis, both the clay and sand soils had a neutral pH. In chapters 3 and 4 it was suggested that the accumulation of organic matter in soil depends on the degree of saturation of protective sites on the mineral surfaces (Hassink *et al.*, 1995). If these protective sites are saturated, less SOM can be stabilized than in soils that have protective sites available (Hassink *et al.*, 1995). If in both soils studied here the protective sites were already occupied, stabilization of recently formed SOM will be impeded and fresh organic materials will decompose at similar rates because they are not protected from biodegradation. Recently, Hassink (1995) was able to estimate the amount of SOM required to saturate all the protective sites in the silt and clay particles (< 20 μm) of a wide range of grassland soils. With his equation and the data presented in chapters 5 and 6 it was possible to estimate the degree of saturation of our soils. The sand soil turned out to be saturated with SOM, but the clay soil was not. Identical decomposition rates of labelled plant material in the sand and clay soil (chapters 3 and 4) can still be explained if recently formed soil organic matter was not (yet) protected against decomposition in either soil (chapters 5 and 6). But similar rates of long-term decomposition (chapter 4) in saturated and non-saturated soil are difficult to explain. It may be possible that labelled SOM is not protected at the beginning of decomposition but that the mineralization rates are reduced as the organic matter gradually becomes physically protected.

Chapter 7 described the decomposition of wheat and clover residues as simulated in a computer model of soil organic matter turnover. The model did not contain any of the factors which were shown to determine the decomposition of SOM in chapters 5 and 6, except residue type. Using a simple function to retard the decomposition of crop residues according to the fibre content, the model was able to estimate the differences between clover and wheat in organic ^{15}N

remaining in soil during the first 48 days of decomposition. Thereafter, equally good simulations of the similar patterns of decomposition of organic ^{15}N in clay and sand soil could be obtained. The use of the model strongly suggested, however, that a portion of the added residues incorporated in SOM was recycled more rapidly than native SOM. This recycling may come about because not all the recently formed SOM can be physically protected immediately.

In conclusion, when the C and N mineralization has to be estimated from crop residues in arable soils, we do not need to include soil structure, broken up by soil disruption and soil texture as a modifying factors of the rate of C and N mineralization. In order to assess the (potential for) protection of SOM against biodegradation in arable soils, the mechanisms of such protection need further research, especially in fine-textured soils.

REFERENCES

Amato, M. and Ladd, J.N. (1992) Decomposition of ^{14}C-labelled glucose and legume material in soils: properties influencing the accumulation of organic residue C and microbial biomass C. *Soil Biology & Biochemistry* **24**, 455-464.

De Willigen, P. (1991) Nitrogen turnover in the soil-crop system; comparison of fourteen simulation models. *Fertilizer Research* **27**, 141-149

Edwards, A.P. and Bremner, J.M. (1967) Microaggregates in soils. *Journal of Soil Science* **18**, 65-73.

Fog, K. (1988) The effect of added nitrogen on the rate of decomposition of organic matter. *Biological Reviews* **63**, 433-462.

Führ, F. and Sauerbeck, D. (1968) Decomposition of wheat straw in the field as influenced by cropping and rotation. In *Isotopes and Radiation in Soil Organic Matter Studies, pp.* 241-250. Proceedings IAEA/FAO Symposium, Vienna.

Hart, P.B.S., Goh, K.M. and Ludecke, T.E. (1979) Nitrogen mineralization in fallow and wheat soils under field and laboratory conditions. *New Zealand Journal of Agricultural Research* **22**, 115-125.

Hassink, J (1995) Soil texture determines the capacity of soils to protect organic C and N against decomposition. *Soil Science Society of America Journal* (submitted)

Hassink, J., Bouwman, L.A., Zwart, K.B., Bloem, J. and Brussaard, L. (1993) Relationships between soil texture, physical protection of soil organic matter, soil biota, and C and N mineralization in grassland soils. *Geoderma* **57**, 105-128.

Hassink, J., Matus, F.J., Chenu, C. and Dalenberg, J.W. (1995) Interaction between soil biota, soil organic matter and soil structure. *Advances in Agroecology* (Submitted).

Helal, H.M. and Sauerbeck D.R. (1986) Influence of plant roots on the stability of soil organic matter and soil aggregates. In *Trans XIII ISSS Congress* **3**, pp. 776-777. ISSS, Hamburg.

Jenkinson, D.S. (1977) Studies on the decomposition of plant material in soil. V. The effects of plant cover and soil type on the loss of carbon from ^{14}C labelled ryegrass decomposing under field conditions. *Journal of Soil Science* **28**, 424-434.

Jingguo, W. and Bakken, L.R. (1989) Nitrogen mineralization in rhizosphere and non-rhizosphere soil: Effect of spatial distribution of N-rich and N-poor plant residues. In: *Nitrogen in organic wastes applied to soils* (J.A. Hansen and H. Henriksen Eds.), pp. 81-97. Academic Press, London.

Kolenbrander, G. J. (1974) Efficiency of organic manure in increasing soil organic matter content. *Trans. 10th Int. Congress of Soil Science*, Moscow, Vol. **2**, pp. 129-136.

Ladd, J. N., Oades J. M. and Amato, M. (1981) Microbial biomass formed from ^{14}C, ^{15}N- labelled plant material decomposing in soils in the field. *Soil Biology & Biochemistry* **13**, 119-126.

Oades, J.M. and Waters, A.G. (1991) Aggregate hierarchy in soils. *Australian Journal of Soil Research* **29**, 815-828.

Reid, J.B. and Goss, M.J. (1982) Suppression of decomposition of ^{14}C-labelled plant roots in the presence of living roots of maize and perennial ryegrass. *Journal of Soil Science* **33**, 387-395.

Sallih, Z. and Bottner, P. (1988) Effect of wheat (*Triticum aestivum*) roots on mineralization rates of soil organic matter. *Biology and Fertility of Soils* **7**, 67-70.

Sauerbeck, D.R. and Gonzalez, M.A. (1977) Field decomposition of carbon-14-labelled plant residues in various soil of the Federal Republic of Germany and Costa Rica. *Soil Organic Matter Studies*. International Atomic Energy Agency, Vienna, **1**, 159-170.

Shields, J.A. and Paul, E.A. (1973) Decomposition of ^{14}C-labelled plant material under field conditions. *Canadian Journal of Soil Science* **53**, 297-306.

Tisdall, J.M. and Oades, J.M. (1982) Organic matter and water-stable aggregates in soils. *Journal of Soil Science* **33**, 141-163.

SUMMARY

The main objective of this thesis is to evaluate the significance of cropping, soil texture and soil structure for the decomposition of crop residues and soil organic matter, the concomitant mineralization of carbon (C) and nitrogen (N) and, *mutatis mutandis*, for the physical protection of organic matter in soil.

Chapter 2 presents a practical model developed to estimate, with time steps of one year, the mineralization of N from the input of crop residues in four agricultural systems in Chile. The model distinguishes between an active and a passive pools of soil organic matter (SOM). Annual soil N mineralization is derived from the active pool which comprises stabilized and labile soil organic N. The accumulation of inputs of fresh organic N during a crop rotation build up the stabilized N, while the labile N is derived from a fraction of the annually added organic N which mineralizes faster than the stabilized N. Using daily averages for rainfall, the model estimated the N losses by leaching as well. The model described the large differences in N mineralization at the four sites studied reasonably well. Cropping, residue type, soil texture and soil structure were not considered.

Chapter 3 presents a greenhouse experiment in which ^{14}C-labelled plant residues were incorporated in a clay and sand soil with similar organic C contents and crop management histories. The hypothesis that the interaction between soil texture, soil structure and root-induced microbial activity can modify the C dynamics in soil was tested. The ^{14}C-labelled residues were mixed with the soils and incubated in pots for 266 days. The experiment included three treatments or their combination: cropping (spring wheat sown twice), soil disruption (mixing of soil six times) and bare soils. Cropping and mixing did not influence residual ^{14}C as compared with bare soils at any time during incubation, neither the residual ^{14}C present in the microbial biomass, nor the residual ^{14}C in humus as measured by acid hydrolyzation.

Chapter 4 describes a similar experiment. Wheat and clover plant residues labelled with ^{15}N were incorporated in the same soils. The residues were allowed to decompose for almost 16 months under field conditions. During the first 48 days of decomposition the residual organic-^{15}N derived from clover materials, containing 4.2% N, declined faster than that from wheat materials, containing 3.2% N. Cropping and mixing did not affect the organic-^{15}N remaining in the soil or microbial biomass-^{15}N. The amounts of inorganic-^{15}N derived from clover and wheat were similar in cropped or mixed soils and there were no differences with the bare treatments. From chapters 3 and 4 it is concluded that the living roots of wheat plants did not suppress or stimulate the decomposition of residues, labelled with ^{14}C or ^{15}N. Apparently soil microbes preferred the labelled residue rich in N used here (C:N ratio 10-17) over root-derived materials. Since the content of fibre (cellulose, hemicellulose and lignin) from clover (40%) was considerably lower than in wheat (60%), the differences in decomposition between clover and wheat during the first 48 days of

decomposition could be explained by the differences in fibre content rather than from microbial N immobilization or effective competition for N between plant and microbes in the rhizosphere.

Soil mixing was not found to be sufficiently disruptive to break up soil aggregates and to expose the labelled residual SOM to further decomposition. The decline of residual ^{14}C and residual ^{15}N during decomposition was not affected by soil texture, and the decay rates between clay and sand soil were identical. This was partly explained by recently formed labelled soil organic matter not (yet) being physically protected against decomposition.

Chapter 5 presents the distribution of unlabelled total C, total N and microbial biomass C in the macro- (> 250 μm) and micro- (< 250 μm) aggregates from the soils used in chapter 4. The C and N mineralization rates after soil dispersion were used as indicators of the degree of SOM protection in aggregates. Five size fractions were obtained: coarse sand (250-2000 μm), fine sand (50-250 μm), coarse silt (20-50 μm), fine silt (2-20 μm) and clay (< 2 μm). Soil organic matter in both soils was more associated with the fine silt (2-20 μm) and clay (< 2 μm) size fractions than with any of the other fractions. However, soil organic matter was found to be much more concentrated in the clay size fraction of the sand soil than in that of the clay soil. An inverse and significant relationship was established between C mineralization rate, N mineralization rate or microbial biomass C and the clay content of aggregates in the sand soil. Such relationships were not found in the clay soil, except for a positive and significant correlation between clay content of aggregates and N mineralization. This suggests that soil organic matter is strongly protected against decomposition in the finest size fraction (< 20 μm) of the sand soil, apparently by adsorption, while in the clay soil other mechanisms of soil organic matter protection are involved. The positive correlation between clay content of aggregates and N mineralization in clay soil may simply be the consequence of the low C:N ratio in the fine silt plus clay (< 20 μm) size fraction which coincided with the highest clay content. Hence it is not yet clear to what extent SOM is protected in small pores in the clay soil.

Chapter 6 reports the distribution of total ^{14}C, total ^{15}N-labelled organic matter and ^{14}C-labelled microbial biomass of the macro- (> 250 μm) and micro- (< 250 μm) aggregates found in the soils used in both chapters 3 and 4. The rates of ^{14}C and ^{15}N mineralization were studied to see if recently formed SOM behaves differently from non-labelled SOM. The distribution of labelled SOM was similar to that of non-labelled SOM in the various aggregate size classes. Both unlabelled and labelled soil organic matter were more concentrated in the < 20 μm size fraction in both clay and sand soils but the concentration of SOM per g of clay was greater in the sand soil. Contrary to what was found in the study of unlabelled C and N, no correlations was established between the clay content of aggregates and the mineralization rates of ^{14}C, ^{15}N and microbial biomass ^{14}C in sand soil. This suggests that recently produced SOM was apparently less physically protected from biodegradation than unlabelled SOM in sand soil. This conclusion is supported by the higher rate of ^{14}C mineralization as compared with the non-labelled C

mineralization, indicating that newly formed SOM decomposes faster than non-labelled native SOM. In clay soil there was a significant and positive correlation between the clay content of the aggregates and the ^{15}N mineralization rate, calculated for 20 days of incubation. It is concluded that soil texture and structure play major roles in determining soil organic matter protection in both soils, but are less important in the protection of newly formed soil organic matter in sand soil. It is not clear yet to what extent small pores constitute an important mechanism of physical protection in clay soil.

Chapter 7 describes the decomposition of wheat and clover residues as simulated in a computer model of the short-term turnover of soil organic matter. The model did not contain any of the SOM decomposition factors (soil texture and physical protection of SOM) dealt with in chapters 5 and 6, except residue type. Using a simple function to retard the decomposition of crop residues according to the fibre content, the model was able to estimate the differences between clover and wheat in organic ^{15}N remaining in soil during the first 48 days of decomposition. Thereafter, equally good simulations of the similar patters of decomposition of organic ^{15}N in clay and sand soil could be obtained.

In conclusion because soil texture and structure were found to have no effect on the rate of decomposition of recently formed organic matter in soil, we do not need to include these factors when the C and N mineralization from crop residues in arable soils has to be estimated. However, because both labelled and unlabelled organic matter were distributed differently over the aggregates size classes of a sand and clay soil and because the mineralization rates of organic matter from within these classes differed in a similar manner, soil texture may play an important role in determining the (potential for) physical protection of SOM against biodegradation in arable soils; the mechanisms of such protection need further research, especially in fine-textured soils.

SAMENVATTING

Het belangrijkste doel van dit proefschrift is het evalueren van de invloed van gewasgroei, bodemtextuur en bodemstructuur op de afbraak van gewasresten en bodem-organische stof (SOM) en de gelijktijdig optredende koolstof- (C) en stikstof- (N) mineralisatie en *mutatis mutandis* op de fysische bescherming van organische stof in de bodem.

Hoofdstuk 2 beschrijft een praktisch model dat is ontwikkeld om, in stappen van een jaar, de mineralisatie te berekenen vanuit gewasresten in vier landbouwkundige systemen in Chili. Het model maakt onderscheid in een actieve en een passieve 'pool' van bodem-organische stof (SOM). De jaarlijkse N-mineralisatie wordt afgeleid uit de actieve 'pool' die bestaat uit gestabiliseerde en instabiele bodem-organische stof. De ophoping van toegevoegde verse organische stikstof gedurende een gewasrotatie leidt tot de opbouw van gestabiliseerde N, terwijl de instabiele N ontstaat uit een deel van de jaarlijks toegevoerde organische N die sneller mineraliseert dan de gestabiliseerde N. Door gebruik te maken van de dagelijkse gegevens van de neerslag is het model in staat om de N-verliezen door uitspoeling eveneens te berekenen. Het model beschreef de grote verschillen in N-mineralisatie van de vier verschillende lokaties betrekkelijk goed. Er werd geen rekening gehouden met gewasgroei, type gewasrest, bodemtextuur en bodemstructuur.

Hoofdstuk 3 laat een *kas-experiment* zien waarin ^{14}C-gelabelde planteresten werden gemengd met een klei- en een zandgrond met hetzelfde organische-stofgehalte en dezelfde historie voor wat betreft gewasbeheer. De hypothese werd getest dat de interactie tussen bodemtextuur, bodemstructuur en de door plantewortels geïnduceerde microbiële activiteit de koolstofdynamiek in de bodem kan beïnvloeden. De met ^{14}C gelabelde planteresten werden gemengd met beide grondsoorten en in potten geïncubeerd gedurende 266 dagen. Het experiment bestond uit drie behandelingen en alle combinaties daarvan: een gewas (zomertar we die twee keer gezaaid werd), grondbewerking door het door elkaar mengen van de grond (6 keer) en grond zonder gewas (braak). Gewasteelt en grondbewerking hadden op geen enkel tijdstip gedurende de incubatie een effect op de residuele ^{14}C in vergelijking met de braak-behandeling; noch op de ^{14}C aanwezig in de residuele microbiële biomassa, noch in de residuele ^{14}C van de humusfractie bepaald met behulp van zure hydrolyse.

Hoofdstuk 4 beschrijft een vergelijkbaar experiment, maar nu vond de uitvoering plaats onder *veldomstandigheden* en werden er resten van tarwe en klaverplanten gebruikt die gelabeld waren met ^{15}N. Deze werden door dezelfde gronden gemengd als in hoofdstuk 3. De planteresten werden onderworpen aan afbraak gedurende bijna 16 maanden. Gedurende de eerste 48 dagen van de afbraak, verdween de residuele ^{15}N uit klaver (met 4.2% N) sneller dan die uit tarwe (met 3.2% N). Gewasgroei en het mengen van de grond had geen effect op de residuele ^{15}N in de grond, noch op de ^{15}N van de microbiële biomassa. De hoeveelheden anorganische ^{15}N uit klaver en tarwe waren gelijk in grond met gewasgroei en gemengde grond, en er waren geen

verschillen met de braakbehandeling. De conclusie uit het werk van hoofdstuk 3 en 4 was dat levende wortels van tarweplanten geen remmend of stimulerend effect hadden op de afbraak van planteresten die gelabeld waren met ^{14}C of ^{15}N. Blijkbaar hadden de micro-organismen die voor de afbraak zorgden een voorkeur voor de gelabelde resten die hier gebruikt werden (met een C:N ratio van 10-17) ten opzichte van materiaal uit levende plantewortels. Aangezien het gehalte aan vezels (cellulose, hemicellulose en lignine) van klaver (40%) behoorlijk lager was dan van tarwe (60%), kon hieruit het verschil in afbraaksnelheid gedurende de eerste 48 dagen beter worden verklaard dan uit een eventuele N-immobilisatie door micro-organismen of een effectieve competitie om N tussen planten en micro-organismen in de rhizosfeer. Verder werd geconcludeerd dat het mengen van de grond niet voldoende was om de aggregaten open te breken, waardoor residuele gelabelde bodem-organische stof aan verdere afbraak blootgesteld zou worden. De afname in residuele ^{14}C en ^{15}N werd niet beïnvloed door de bodemtextuur, en de afbraaksnelheid in klei en zand waren gelijk. Dit kon gedeeltelijk verklaard worden door aan te nemen dat recent gevormde gelabelde bodem-organische stof (nog) niet beschermd was tegen afbraak.

Hoofdstuk 5 laat de verdeling van ongelabelde koolstof, stikstof en microbiële koolstof zien over macro-aggregaten (> 250 µm) en micro-aggregaten (< 250 µm) van de grondsoorten die werden gebruikt in hoofdstuk 4. De snelheid van koolstof- en stikstofmineralisatie in de aggregaatfracties werd gebruikt als een aanwijzing voor de mate van bescherming van SOM in de aggregaten. Vijf fracties werden verkregen: grof zand (250-2000 µm), fijn zand (50-250 µm), grof silt (20-50 µm), fijn silt (2-20 µm) en klei (< 2 µm). Bodem-organische stof in beide grondsoorten was meer geassisocieerd met de fijne silt en klei-fracties dan met enige andere fractie. Echter, de concentratie bodem-organische stof in de kleifractie van de zandgrond was veel hoger dan die in de kleigrond. Er bestond een omgekeerde en betrouwbare relatie tussen de snelheid van koolstof- en stikstofmineralisatie of de koolstof in microbiële biomassa en het kleigehalte van de aggregaten van de zandgrond. Dergelijke relaties werden niet gevonden in de kleigrond, op één na: er was een positieve en betrouwbare correlatie tussen het kleigehalte van aggregaten en de stikstofmineralisatie. Dit wijst erop dat bodem-organische stof in hoge mate wordt beschermd tegen afbraak in de fijnste bodemfractie van de zandgrond, wellicht door adsorptie, terwijl er in klei andere beschermingsmechanismen optreden. De positieve correlatie met de N-mineralisatie in de kleigrond kan eenvoudigweg het gevolg zijn van de lage C:N-ratio in de silt plus klei-fractie die samenging met het hoogste kleigehalte. Vandaar dat het nog niet geheel duidelijk is in welke mate SOM in de kleine poriën van de kleigrond wordt beschermd.

Hoofdstuk 6 vermeldt het resultaat van onderzoek naar de verdeling van ^{14}C en ^{15}N uit de totale organische stof en de ^{14}C uit microbiële biomassa over macro- en micro-aggregaten uit de gronden van hoofdstuk 3 en 4. De snelheid van ^{14}C- en ^{15}N-mineralisatie werd bestudeerd om vast te stellen of recent gevormde en gelabelde SOM zich op dezelfde wijze gedraagt als oudere, niet

gelabelde SOM. De verdeling van gelabelde en niet gelabelde SOM was gelijk over de verschillende aggregaatgrootte-klassen. Zowel gelabelde als ongelabelde SOM was meer geconcentreerd in de fractie < 20 µm, van zowel klei als zandgrond, maar de concentratie per eenheid klei was groter in de zandgrond. In tegenstelling tot wat was gevonden in het onderzoek met ongelabelde C en N, bestond er geen correlatie tussen het kleigehalte van de aggregaten en de mineralisatiesnelheid van ^{14}C en ^{15}N en de ^{14}C van de microbiële biomassa in zandgrond. Dit wijst erop dat recent gevormde SOM minder goed tegen afbraak beschermd was dan oudere ongelabelde SOM. Deze conclusie werd ondersteund doordat de ^{14}C-mineralisatie hoger was dan de niet gelabelde C-mineralisatie, wat erop wees dat recent gevormde SOM sneller werd afgebroken dan niet gelabelde natieve SOM. In de kleigrond bestond een betrouwbare positieve correlatie tussen het kleigehalte van de aggregaten en de ^{15}N-mineralisatiesnelheid gedurende de eerste 20 dagen van de incubatie. De conclusie was dat de bodemtextuur en -structuur een hoofdrol speelden in de wijze waarop organische stof in de bodem werd beschermd in beide grondsoorten, maar dat hun rol in zandgrond minder groot is dan in kleigrond in het geval van recent gevormde SOM. Het is nog niet duidelijk in welke mate kleine poriën van kleigrond een belangrijk mechanisme zijn in de fysische bescherming van organische stof.

Hoofdstuk 7 beschrijft de afbraak van tarwe- en klaver-resten, gesimuleerd met behulp van een computermodel van de korte-termijn turnover van bodem-organische stof. In het model waren geen van de SOM afbraakfactoren als bodemtextuur en fysische bescherming van SOM opgenomen, met uitzondering van het type planterest. Met behulp van een eenvoudige functie om de afbraak van gewasresten te laten afnemen met het vezelgehalte, was het model in staat om het verschil te berekenen tussen klaver en tarwe, met betrekking tot de organische ^{15}N die in de grond achterblijft gedurende de eerste 48 dagen van de afbraak. Voor de daaropvolgende periode kon ook een goede simulatie voor het gelijke afbraakpatroon van de afbraak van organisch ^{15}N in zand en klei worden verkregen.

Omdat gevonden werd dat bodemtextuur en bodemstructuur (als gevolg van grondbewerking) geen invloed hadden op de afbraaksnelheid van recent gevormd organisch materiaal, kan geconcludeerd worden dat deze bodemfactoren niet hoeven te worden betrokken bij het berekenen van de C- en N-mineralisatie uit recent toegevoegde gewasresten in landbouwakkers.

Echter, zowel gelabelde verse als ongelabelde 'oude' organisch stof was verschillend verdeeld over de verschillende aggregaat-grootteklassen. Een vergelijkbare verdeling werd gevonden voor de mineralisatie van organische stof uit deze aggregaten. Daarom kan geconcludeerd worden dat de bodemtextuur een belangrijke rol moet spelen in de (potentiële) fysische bescherming van bodem-organische stof.

RESUMEN

El objetivo principal de esta tesis es evaluar la importancia del efecto del cultivo, textura y estructura del suelo sobre la descomposición de los residuos vegetales y materia orgánica del suelo, al mismo tiempo evaluar la mineralización de carbono (C) y nitrógeno (N) y, *mutatis mutandis*, para la protección física de la materia orgánica en el suelo.

El capítulo 2 presenta un modelo práctico desarrollado para estimar la mineralización anual de N a partir de la incorporación de los residuos vegetales en cuatro sistemas agrícolas chilenos. El modelo distingue entre un 'pool' activo y un 'pool' pasivo de materia orgánica del suelo (SOM). La mineralizacón anual de N es derivada del 'pool' activo, el cual consiste de un nitrógeno orgánico estabilizado y otro lábil. El N estabilizado es acumulado por la permanente incorporación de N orgánico proveniente de los residuos vegetales frescos de la rotación de cultivos, mientras que el N lábil es derivado de una fracción de los residuos vegetales incorporados anualmente y su tasa de mineralización es más rápida que la del N orgánico estabilizado. Usando el promedio diario de precipitación, el modelo estima también, las pérdidas de N por lixiviación. El modelo describe razonablemente bien las grandes diferencias de mineralización de N en los cuatro sistemas agrícolas estudiados. El efecto del cultivo, el tipo de residuo vegetal, la textura y estructura del suelo no fueron considerados.

El capítulo 3 presenta un experimento en invernadero, en el cual fueron incorporados residuos vegetales marcados con ^{14}C en un suelo arcilloso y uno arenoso, ambos con contenidos de C orgánico e historia de rotación de cultivos similares. La hipótesis de que la dinámica del C en el suelo puede ser modificada por la interacción entre la textura, la estructura y la actividad microbial inducida por las raíces fue probada. Los residuos vegetales marcados con ^{14}C fueron mezclados con los suelos e incubados en macetas experimentales por 266 días. El experimento incluyó tres tratamientos o sus combinaciones: cultivación (trigo de primavera sembrado dos veces), desintegración de la estructura del suelo (mezclado del suelo, seis veces) y suelo descubierto no disturbado. La cultivación y la destrucción de la estructura del suelo no influyeron en el ^{14}C residual durante la incubación comparado con el suelo no disturbado ni sobre el ^{14}C presente en la biomasa microbiana, ni en el ^{14}C presente en forma húmica, medido por hidrolización ácida.

El capítulo 4 describe un experimento similar. Dos tipos de residuos vegetales, trigo y trébol, marcados con ^{15}N fueron incorporados en los mismos suelos. La descomposición fue llevada a cabo por casi 16 meses en condiciones de campo. Durante los primeros 48 días de descomposición el ^{15}N orgánico residual derivado de los residuos vegetales de trébol, conteniendo 4.2% N, declinó más rápido que aquel derivado de trigo, conteniendo 3.2% N. Cultivación y desintegración de la estructura del suelo no afectaron el ^{15}N orgánico residual o el ^{15}N en la biomasa microbiana. La concentración de ^{15}N inorgánico derivado (NO_3^--N + NH_4^+-N) de los residuos vegetales de trébol

y trigo fueron similares en los suelos cultivados, mezclados o no disturbados. Del capítulo 3 y 4 es concluido que las raíces vivas del cultivo de trigo no estimuló ni suprimió la descomposición de los residuos vegetales marcados con ^{14}C o ^{15}N. Aparentemente los microbios del suelo prefirieron el substrato marcado rico en N usado en estos experimentos (C:N 10-17) más que el substrato derivado de las raíces. Debido a que el contenido de fibra (celulosa, hemicelulosa y lignina) del residuo vegetal de trébol (40%) fue considerablemente más bajo que en el residuo de trigo (60%), la diferencia en la descomposición entre el residuo de trébol y trigo durante los primeros 48 días de incubación pudo ser explicado por las diferencias en los contenidos de fibra más que por la inmobilización de N por la biomasa microbiana o por la competencia efectiva por N entre la planta y los microbios de la rizosfera. Por otra parte, la intensidad con que fue mezclado el suelo no fue suficiente para romper los agregados y exponer la SOM marcada a la descomposición. La declinación del ^{14}C y ^{15}N residual durante la incubación no fue afectada por la textura del suelo, y las tasas de descomposición del suelo arcilloso y arenoso fueron idénticas. Estos resultados fueron parcialmente explicados debido a que la materia orgánica marcada, recientemente formada no es (no todavía) físicamente protegida contra la biodegradación.

El capítulo 5 muestra la distribución del C y N total no marcados y del C no marcado en la biomasa microbiana de los macro-agregados (> 250 μm) y micro-agregados (< 250 μm) de los suelos usados en el capítulo 4. Las tasas de mineralización de C y N fueron usadas como indicadores del grado de protección física de la SOM. Cinco agregados fueron obtenidos: arena gruesa (250-2000 μm), arena fina (50-250 μm), limo grueso (20-50 μm), limo fino (2-20 μm) y arcilla (< 2 μm). La materia orgánica fue más concentrada en las fracciones de limo fino (2-20 μm) y arcilla (< 2 μm) que en las otras fracciones. Sin embargo, la materia orgánica fue mucho más concentrada en la fracción de arcilla en el suelo arenoso. Una relación inversa y significante ($P < 0.01$) fue establecida entre la tasa de mineralización de C, la tasa de mineralización de N o el C presente en la biomasa microbiana y el contenido de arcilla de los agregados en el suelo arenoso. La misma relación en el suelo arcilloso no fue encontrada, excepto por una correlación significante ($P < 0.01$) y positiva entre el contenido de arcilla de los agregados y la mineralización de N. Estos resultados sugieren que la materia orgánica es fuertemente protegida contra la descomposición en la fracción fina (< 20 μm) del suelo arenoso, aparentemente por adsorción, mientras que en el suelo arcilloso otros mecanismos de protección de la materia orgánica parecen estar involucrados. La correlación positiva entre la mineralización de N y el contenido de arcilla de los agregados en el suelo arcilloso pudo ser una consecuencia de la baja relación C:N en la fracción limo y arcilla (< 20 μm) la cual, a su vez, coincidió con el contenido más alto de arcilla. Por lo tanto, no es claro aún en qué grado la SOM es físicamente protegida en pequeños poros en el suelo arcilloso.

El capítulo 6 muestra la distribución del ^{14}C total, ^{15}N total y ^{14}C presente en la biomasa microbiana de los macro- y micro-agregados de los suelos usados en el capítulo 3 y 4. Las tasas

de mineralización de ^{14}C y ^{15}N fueron estudiadas para comprobar si la recientemente formada SOM se comporta en forma similar a la SOM no marcada. La distribución de SOM marcada fue similar como la SOM no marcada en los diferentes clases de agregados. La SOM marcada y no marcada fue más concentrada en la fracción < 20 µm en el suelo arcilloso y arenoso, pero la concentración de SOM por gramo de arcilla fue mucho mayor en el suelo arenoso. Al contrario de lo encontrado en el estudio del C y N no marcados, no correlación fue establecida entre el contenido de arcilla de los agregados y las tasas de mineralización de ^{14}C, ^{15}N y ^{14}C de la biomasa microbiana en el suelo arenoso. Esto resultados sugieren, aparentemente, que la SOM recientemente formada esta menos protegida que la SOM no marcada en el suelo arenoso. Esta conclusión fue respaldada por las mayores tasas de mineralización de ^{14}C comparadas con las mismas tasas de mineralización de C no marcado, indicando que la SOM recientemente formada descompone más rápido que la SOM nativa no marcada. Hubo una significante ($P < 0,05$) y positiva correlación entre el contenido de arcilla de los agregados y la tasa de mineralización de ^{15}N, calculada durante 20 días de incubación en el suelo arcilloso. De los resultados del capítulo 5 y 6 es concluido que la textura y estructura determinan la protección física de la SOM en ambos suelos, sin embargo son menos importantes en la protección física de la SOM recientemente formada en el suelo arenoso. En el suelo arcilloso, no es claro aún en qué grado los pequeños poros del suelo constituyen un mecanismo de proteccion física de la SOM.

El capítulo 7 describe la descomposición de los residuos vegetales de trigo y trébol, simulados en un modelo computacional (por día). El modelo no contiene ninguno de los factores de la descomposición de la SOM (textura del suelo y protección física de la SOM) tratados en los capítulos 5 y 6, excepto el tipo de residuo vegetal. Usando una función simple para retardar la descomposición de los residuos vegetales de acuerdo al contenido de fibra, el modelo fue capaz de estimar las diferencias entre la descomposición del residuo de trébol y trigo durante los primeros 48 días de incubación. Después de este período la declinación de ^{15}N orgánico fueron simulados igualmente bien, tanto en el suelo arcilloso como en el suelo arenoso.

En conclusión, debido a que la textura y la estructura del suelo no tuvo ningún efecto sobre las tasas de descomposición de la SOM recientemente formada, no se necesita, entonces, tomar en cuenta estos factores si el propósito es estimar la mineralización de C y N a patir de los residuos vegetales en los suelos arables. Sin embargo, la textura y estructura del suelo tienen una importante función que determina la (potencial) protección física de la SOM contra la biodegradación, ya que ambas la SOM marcada y no marcada fueron diferentemente distribuídas en las distintas clases de agregados en ambos suelos, y debido a que las tasas de mineralización de la SOM de los agregados difirieron en una manera similar.

CURRICULUM VITAE

Francisco Javier Matus was born on July 20, 1958 in Santiago, Chile. He began his undergraduate study at the Agronomy School of Pontificia Universidad Católica de Chile in March of 1978, receiving his Title Engineer in Agronomy (Crop Science) in 1984. He continued his study as a graduate student at the same University and obtained his MSc in Soil Science (Soil Fertility) in November 1987. He worked as Research Assistant Professor from 1985 to 1987. Thereafter he worked at the Agronomy School of Talca University as Assistant Professor in Soil Fertility. In 1990 he joined the Research Institute for Agrobiology and Soil Fertility (AB-DLO) to complete his PhD study, supported by a grant under the Presidential Fellowship Programme of the Agencia de Cooperación Internacional (AGCI) del Ministerio de Planificación y Cooperación de Chile (MIDEPLAN) and by grant N° P806 under the sandwich PhD Fellowship Programme of Wageningen Agricultural University. After his return to Chile he will join his former position at the Talca University.