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The role of amino acids and nucleic bases in turnover of nitrogen and carbon in soil humic fractions

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Summary

Incorporation of labelled ^{15}N and ^{14}C amino acids and nucleic bases into soil humus fractions as well as humus turnover was investigated under field conditions. The dynamics of ^{15}N and ^{14}C incorporation into organic matter was characterized by the following main steps: rapid incorporation of the labelled substance prevailing for the first 1–3 weeks, and decomposition of included labelled fragments prevailing beyond one month after substance addition. The annual turnover rates of N and C in humus fractions due to incorporation of amino acids and nucleic bases were calculated. The turnover rate of N in humus is two to three times that of C. The contribution of amino acids to organic matter generation is about twice as great as that of nucleic bases and other N-containing organic substances. This indicates the important role of amino acids in the humification process and humus turnover. Turnovers of humic acids (0.002 $year^{-1}$ for C and 0.02 for N) are the most rapid of humic fractions investigated, and humin is characterized by the slowest turnover (0.0002 $year^{-1}$ for C and 0.007 for N). There are no significant differences in the turnover rates of fulvic acid fractions (0.0002 $year^{-1}$ for C) with different molecular weight.

Introduction

It has been estimated that 90–95% of the total nitrogen (N) in surface soil is organic (Stevenson, 1982). Hydrolysis studies show that 20–50% of the total N is bound in amino acids, 3–10% in amino sugars, whilst less than 1% is present as purine and pyrimidine derivates (Stevenson, 1982). Most of the N input into soil is from microbial and plant remains in the form of proteins and nucleic acids with the ratio ranging from 1 : 1 to 10 : 1 (Alberts *et al.*, 1987; Rodin & Basilevich, 1965; Ladd *et al.*, 1981). Compared with proteins and nucleic acids, inputs of other organic N-forms (chlorophyll, chitin, amino sugars) are rather small. In the soil the high molecular weight substances are rapidly degraded (from 1 to 4 weeks) by microorganisms to low molecular substances (amino acids and nucleic bases) (Verma *et al.*, 1975; Haider, 1992). These low molecular substances then disappear by one or more of the following mechanisms.

1 Mineralization (Bondietti *et al.*, 1972; Verma *et al.*, 1975; Martin *et al.*, 1974; Martin & Haider, 1976).

2 Migration down the profile.

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3 Plant and microbial uptake (Ladd *et al.*, 1981; Van Veen *et al.*, 1987; Sørensen, 1987).

4 Participation in humification (Fokin, 1974, 1975; Verma *et al.*, 1975; Sørensen, 1987).

Fokin *et al.* (1992), have described the first three processes. In most investigations of humification the participation of high molecular compounds of plant and microbial residues has been postulated (Verma *et al.*, 1975; Jenkinson, 1977; Marumoto *et al.*, 1982; Sørensen, 1987; Müller, 1988).

The problem of the transformation of organic matter in soil remains acute, particularly because of the obscure role of individual compounds of plant and microbial residues in the biogeochemical cycles of the elements and their participation in the renewal and formation of the humus resource. Individual components of plant and microbial residues containing N play an important role as an exchangeable resource of nutrient elements especially of N in both natural and agricultural ecosystems.

The parallel investigation of the dynamics of N and C in soil in one experiment has many advantages. The employment of ^{14}C and ^{15}N labelled substances is especially useful in this context.

The aim of this investigation was to study the dynamics of two forms of organic N and C (amino acids and nucleic bases) in some humic fractions of a Podzoluvisol.

Test plot	Labelled substance	^{14}C-substances ^{14}C-activity /kBq g^{-1}	Amount /mg g^{-1}	^{15}N-substances atom ^{15}N-excess /%	Amount /mg g^{-1}
1	2-^{14}C-uracil	3.08	0.0002	—	—
	1,3-^{15}N-uracil	—	—	48.93	0.116
2	2-^{14}C-glycine	1.54	0.0001	—	—
3	1-^{14}C-alanine	1.54	0.0001	—	—
	^{15}N-alanine	—	—	70.42	0.072

Table 1 Some characteristics of the labelled substances used in the experiment.

Materials and methods

A Field experiment

Field experiments were done on an arable, loamy Dystric Podzoluvisol (FAO-UNESCO, 1990) at the experiment station of the Russian Research Institute of Fertilizers and Agricultural Soil Science at Barybino, 60 km south-west from Moscow. Topsoil (0–0.2 m) had a pH_{KCl} of 5.5, and contained about 0.75% organic C, 0.07% total N, and 29% clay fraction (0.01 mm).

Uracil, glycine and alanine, labelled at different positions with ^{15}N and ^{14}C (Table 1), were applied with red clover (*Trifolium pratense* L.) root residues (120 g, 20% dry weight) to 12 kg of topsoil on plots each of which was 0.2 m × 0.2 m square and with topsoil 0.2 m thick. The application of labelled substances together with the red clover residues simulated the incorporation of organic residues into the natural soil. Alanine and glycine were chosen for the experiment because they are the simplest amino acids and because their concentration in soil (Mamchenko, 1970; Umarov & Aseeva, 1971) and humus substances is larger than that of the other amino acids. Soil treated with the ^{14}C- and ^{15}N-labelled substances was sampled (750 g) from each test plot at 1 week, 1 month, 2.5 month, 4.5 month, and 1 year intervals. Some plots were additionally sampled immediately after application of the labelled substances (15 May, 1987) and at 1.5 year (15 October, 1988). One core (0.05 m × 0.05 m × 0.2 m = 0.0005 m^3 weighing 750 g) was taken from each replicated plot. The free space in the plots caused by sampling was demarcated from the labelled soil and filled up with similar soil without labelled substances. Soil samples were air-dried (60°C), ground and sieved (to pass 1 mm) before analysis.

Barley (*Hordeum vulgare* L.) was grown in the first year and wheat (*Triticum aestivum* L.) in the second year after adding the substances to the soil. The plant uptake of ^{14}C and ^{15}N has been reported earlier by Fokin *et al.* (1992).

B Fractionation of soil organic substances

Organic substances were extracted from soil cores and fractionated according to the scheme shown in Fig. 1. The soil samples were first treated three times with aqueous ethanol (20%) (for plots with labelled amino acid) or three times with distilled water at 70°C (for plots with labelled uracil) to extract free amino acids (Schmidt *et al.*, 1960; Cheng, 1975) or free nucleic bases (Fokin *et al.*, 1992) from soil. The soil residue was subsequently extracted with 0.1 M NaOH (1 : 5) several (5–7) times. The dissolved humic substances were first centrifuged at 4000 g for half an hour to remove soil particles, then at 6000 g for one hour to achieve separation from organo-mineral colloids. The ash contents of humic substances after centrifugation did not exceed 5%. Extracted humic substances were separated into humic and fulvic acid fractions by means of a cation exchange resin column (Dowex 50 × 8 in H^+-form). When the extracted humic substances passed through the cation exchange resin the pH of the solution changed from 14 to 3–4. The fraction passing through the column was regarded as a mixture of fulvic acids and nonhumic substances of soil organic matter of low molecular weight (organic acids, sugars). The fraction remaining on the column was eluted with 2 M NH_4OH. This fraction is referred to as the humic acid (HA) fraction. The results of ^{15}N-incorporation into the HA fraction may be distorted as a result of the contact with NH_4OH.

The fulvic acid (FA) fraction and nonhumic substances of soil organic matter eluted from the cation exchange resin column were separated into nonhumic substances (molecular weight, MW (700 Da) and FA (MW $\geq$ 700 Da) by gel filtration on Sephadex G-10 (Uppsala, Sweden, separation limits: 0–700). Details of the G-10 and G-50 Sephadex columns are presented in Table 2. The relation between MW, denoted by M_w in the equation, and the K_d-distribution coefficient typical for each column are calculated for MW identification (Karpukhin & Fokin, 1970):

$$\log_{10}(M_W) = A \times K_d + \log_{10}(M_{Wup}),$$

and

$$K_d = \frac{V_e - V_0}{V_i},$$

where A is a constant, M_{Wup} is the upper separation limit of molecular weight for the column, V_e is the elution volume of FA fraction by separation, V_0 is the volume of free solution unbounded on the gel, and V_i is the volume of solution bounded on the gel.

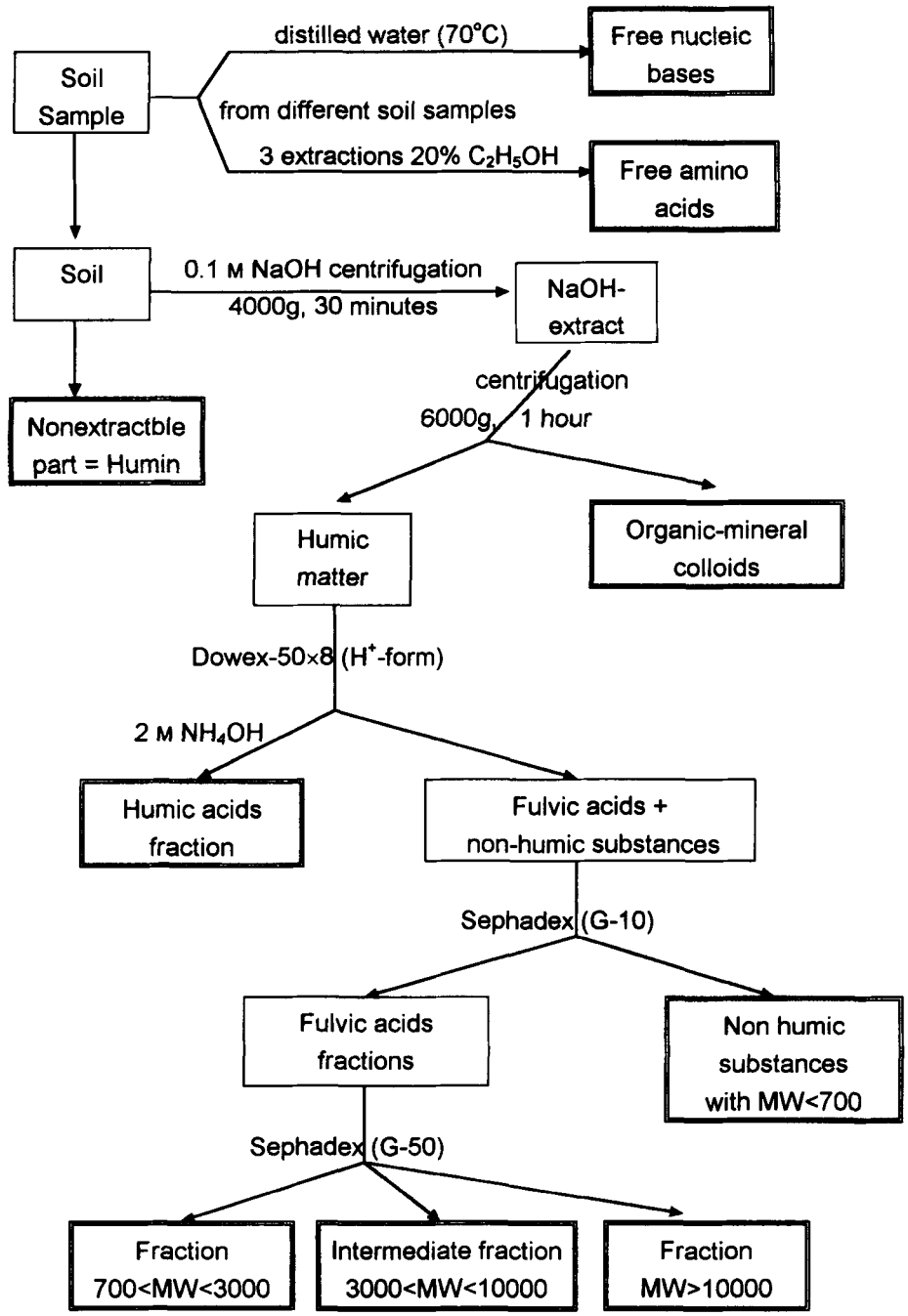

Fig. 1 Fractionation of soil organic matter. (Total C, total N and labelled ^{14}C, ^{15}N were measured in all fractions with two frames) [MW—molecular weight].

The FA was separated into three fractions with MW = 700–3000, 3000–10000, >10000 by gel filtration on Sephadex G-50 (Uppsala, Sweden, separation limits: 500–10000). The intermediate fraction of FA with MW = 3000–10000 was used only for recovery calculations. The FA with MW > 10000 was not further separated into different fractions and the MW of this FA fraction was not exactly determined.

Reported results are means of three replicates. The coefficient of variation for replicates of ^{14}C and ^{15}N content does not exceed 20%. The coefficient of variation for replicate organic matter extraction and separation (C_{total} and N_{total} content) was 10%.

C Analyses

The amount of total and labelled C and N was determined in soil samples as well as in the humic fractions. The ^{14}C-activity was measured by scintillation counting (RackBeta, model

Gel	Separation limits /daltons	Column height /cm	Column diameter /cm	V_t /ml	V_i /ml	V_0 /ml	V_g ml
G-10	0–700	26	3.5	250	65	104	79
G-50	500—10000	38	2.2	144	88	46	11

V_t, general column volume.
V_i, volume of solution bounded on the gel.
V_0, volume of free solution unbounded on the gel.
V_g, the gel volume.

Table 2 Details of Sephadex columns for gel filtration of fulvic acids.

1219 of the LKB Wallac company of Finland) in Bray cocktails (for solutions) and Luma Gel (Lumac Company) (for dried solid soil samples and nonextractable humin). A special method using gel-scintillation cocktails (Kukushkin & Kuzyakov, 1991) was used to determine ^{14}C-activity directly in solid soil samples. In this method the ground soil particles ($\leqslant 0.25$ mm) are evenly distributed throughout the entire volume of the gel, giving counting efficiencies of 85–95% for solutions and 65–80% for solid samples of soil and humin. The ^{14}C-activity of humic substance dissolved in NaOH or NH_4OH was measured after the ending of the chemiluminescence. The absolute ^{14}C-activity was standardized using SQP(E) method separately for the solutions (with 0.1 M NaOH solution) and for the solid samples (with soil), allowing a comparison of the ^{14}C-activity of the soil samples and humic fraction solutions and the calculation of an overall recovery of ^{14}C. The ^{14}C-activity measurement error of the solutions and of the solid samples did not exceed 3 and 10%, respectively ($P \leqslant 0.01$).

The total C content in humic fractions was determined by dichromate oxidation at 100°C (2 ml humic fraction solution + 1.5 ml 15M H_2SO_4 + 1.5 ml saturated aqueous solution of $K_2Cr_2O_7$). The residual $K_2Cr_2O_7$ was determined photometrically at 565 nm. The calibration of the total C content measurements was done with glucose.

The total nitrogen content of the air-dried soil samples and the solutions of the humic fractions was determined by Kjeldahl digestion and titration. The $^{15}N/^{14}N$ ratio was determined on a mass spectrometer (Mi-1201B) after conversion of all N-forms into N_2. Soil and humic fraction N was converted to NH_4^+ by the Kjeldahl method and then to N_2 by the hypobromite method. The natural ^{15}N content was 0.365%. The error of mass spectrometric determinations did not exceed 1%.

Results and discussion

The fractionation method, described above, separates all organic substances into two fractions of nonhumic, low molecular weight organic substances (amino acids or nucleic bases, and nonhumic substances with MW $\leqslant$ 700) and six fractions of humic substances (humin, HA, three FA fractions and organo-mineral colloids). The recovery of C (Table 3) is similar to that from traditional fractionation methods for the Podzoluvisol (Stevenson, 1982; Orlov, 1985). The HA content using the present method is smaller relative to FA (C_{HA} $C_{FA}^{-1} = 0.07$) because of the pH-change only to 3–4 on treating the alkaline soil extract with cation exchange resin, giving less HA in comparison to traditional methods where the pH is lowered to 1. The humin content is larger in comparison to methods with acid treatment before humus extraction.

The proposed fractionation procedure for soil organic matter allows the separation of fractions with small and large ^{14}C specific activity without humus hydrolysis because of the usual acid treatment used to separate HA and FA. Chemical modification of FA by gel filtration is negligible, and the fractionation of FA mixture and MW determination occurs at the same time (Karpukhin & Fokin, 1970). Examples of the separation of FA from nonhumic organic soil substances on Sephadex G-10 and the fractionation of FA by MW on Sephadex G-50 are shown in Fig. 2. Fractions of higher MW are enriched with ^{14}C in comparison to low MW fractions 1 month after the labelled substance had been added to the soil. Humic substances from soil samples taken immediately and 1 week after labelled substance application have lower MW richer in ^{14}C than that in the high MW fractions. This shows that there is faster renewal of those humic fractions with lower MW than those with high MW.

Table 3 Total carbon content in the soil organic matter fractions studied.

Fractions	Total carbon content: Total amount /mg g^{-1}	Total carbon content: Percentage distribution
Soil (total)	7.5	100
Humin	4.0	53
Organo-mineral colloids	0.37	5
Humic acids	0.19	3
Non-specific substances (MW[a] < 700)	0.24	3
Fulvic acids (total)	2.69	36
FA with MW = 700–3000	1.17	16
FA with MW = 3000–10000	0.38	5
FA with MW > 10000	0.46	6
Losses	0.68	9

[a]MW, molecular weight.
Variation (10% of the value ($P \leqslant 0.01$)).

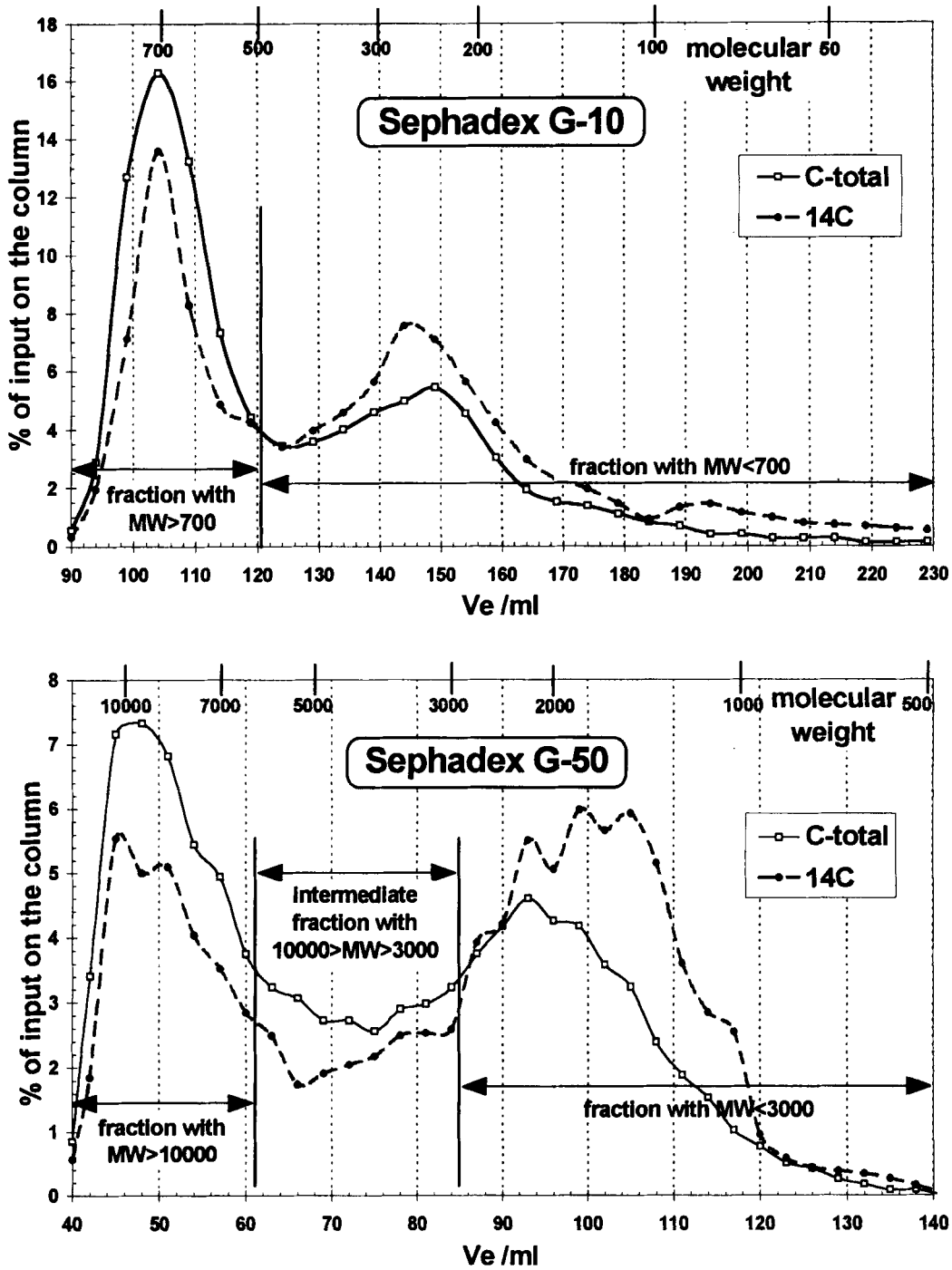

Fig. 2 Gel filtration of fulvic acids with different molecular weight (MW). Top: separation of fulvic acids from non specific organic substances with MW < 700 on Sephadex G-10 (4 month after treatment with 2-^{14}C-glycine). Bottom: fractionation of fulvic acids with different MW on Sephadex G-50 (2.5 month after treatment with 2-^{14}C-uracil).

Both isotopes, ^{14}C and ^{15}N, were detected in all organic matter fractions after addition of labelled uracil, glycine and alanine to soil. This conforms with what has been observed after the addition of labelled low molecular substances (Fokin, 1974, 1975) as well as plant (Oberländer & Roth, 1974; Fokin, 1974, 1975; Müller, 1988) and microbial residues (Martin *et al.*, 1974; Martin & Haider, 1976) to soil. Organic matter fractions extracted from soil had different ^{14}C specific activities and ^{15}N atom excesses. The content of ^{14}C and ^{15}N of humin, FA with MW > 10 000 and HA from the soils

treated with the labelled amino acid and uracil are shown in Fig. 3. The alteration of ^{14}C and ^{15}N with time in these fractions shows dynamics typical of newly included compounds in the humus.

The isotope content in the organic fraction depends on the amount of labelled substances applied to soil (Jenkinson, 1977), on the humic fraction content in the soil, and on the annual turnover rate of the fraction studied. The initial soil samples contained more ^{14}C and ^{15}N in the low MW fractions of FA than in the high MW fractions. In the later soil samples the reverse was the case (Fig. 2).

The larger quantity of humin (53% of total C) compared with FA with MW > 10 000 (6% of total C) leads to 3–5 fold more ^{15}N and ^{14}C in the humin fraction.

The content of the substances labelled with isotopes increased rapidly with a noticeable maximum followed by a rapid decrease (Fig. 3). The content and the dynamics of ^{14}C and ^{15}N in the fractions changed considerably for different organic fractions and for the different labelled substances. The maximum values were obtained within 1 month from the beginning of the experiment, in some cases within 1 week.

The content of ^{14}C and ^{15}N in organic fractions depends on the relative importance of the following main processes: the rapid incorporation of low molecular transformation products of the labelled substances in the first 1–2 weeks after they were added to soil (Cortez & Schnitzer, 1981; Müller-Wegener, 1982), and the formation of the more mobile peripheral part of humus with subsequent transformation of some part of these fragments into stable structures. These mobile fragments can be also microbiologically decomposed or hydrolysed (Fokin, 1974) 1–2.5 month after addition of the substance. In 2.5 months the decomposition of incorporated labelled fragments was very slow, and for some substances (2-^{14}C-uracil, 1-^{14}C-alanine) was similar to reported decomposition rates of humic fractions (Rodin & Basilevich, 1965; Verma *et al.*, 1975; Jenkinson & Rayner, 1977; Sørensen, 1987).

Incorporation of ^{14}C into soil organic matter (OM) structures depends on the position of C in the amino acid molecule (Ugrekhelidze & Dolidze, 1987). The inclusion of nitrogen and carbon in the OM is approximately the same for ^{15}N and ^{14}C from the second position of the amino acids investigated (Fig. 3). The existence of the C-N bond in the amino acids suggest that unbroken C-N amino acid fragments are incorporated into the OM structure. This seems to contradict the fact that amino acids added to the soil are deaminated by the microbial enzymes (Tena *et al.*, 1986; Alberts *et al.*, 1987) with the subsequent utilization of inorganic N by microorganisms. This contradiction can be explained by the rapid chemical incorporation of amino acids into humic molecules (Bondietti *et al.*, 1972; Kuzyakov & Galitsa, 1993; Dashman & Stotzky, 1982) before they are decomposed into amino acids.

The amounts of the carbon from the carboxyl groups of the amino acids are much smaller than those of methylene groups because the former are completely and rapidly decarboxylated after their addition to soil (Tena *et al.*, 1986; Ugrekhelidze & Dolidze, 1987; Fokin *et al.*, 1992).

For uracil the incorporation of ^{15}N in humic fraction was greater than that of the ^{14}C (Fig. 3). This shows the difference in the transformation in soil between ^{15}N and 2-^{14}C of uracil. That is because uracil was decomposed more completely than amino acids. Complete oxidation to CO_2 of the second carbon atom from uracil by microorganisms was observed with the production of β-alanine and NH_4^+ (Alberts *et al.*, 1987). The CO_2 is lost from the soil, and the β-alanine and NH_4^+ can be incorporated in humic substances.

One year after application of the labelled substances the largest content of labelled atoms in humic fractions was obtained for the second C of the amino acids and for N of the amino acids and of uracil. The maximal incorporation of ^{14}C from amino acids was observed in the HA fraction (Fig. 3). This conforms with what has been observed after the addition of low molecular substances (Ugrekhelidze & Dolidze, 1987) and plant residues. The fact that more C is incorporated into HA than into FA contradicts results obtained by radiocarbon dating (Chichagova, 1987), perhaps because the peripheral parts of HA molecules turn over faster than those of FA, because of the greater N content of the peripheral structure of HA (Stevenson, 1982; Orlov, 1985). In this study only N-containing substances were analysed without identifying the role of carbon incorporation from sugars, lignin, cellulose, etc. On the other hand, the selectivity of low molecular substances (estimated by humification coefficients and inclusion in different humic fractions) during the humification can vary greatly. For example, the ^{14}C of amino acids was characterized by the maximal inclusion in the HA structure, whereas the incorporation of the uracil occurred only to a small extent.

The results obtained confirmed the turnover of all organic fractions in soil (Fokin, 1974; Jenkinson & Rayner, 1977; Chichagova, 1987). The annual turnover rate of the organic fractions is important in the nutrient cycle (Fokin, 1975; Chichagova, 1987). Usually the annual turnover rate of the carbon (as an integral parameter for comparing the different humic fractions and different types of humus) has been calculated to characterize the role of plants and microbial residues in the OM cycle (any specific OM fraction or all fractions). But the total carbon turnover is inadequate for characterizing the role of the specific groups of substances in the nutrient cycle. Thus the role of some substances such as amino acids, with small concentrations in soil solution, can be more important for characterizing the humus turnover than substances such as lignin and cellulose with large concentrations in soil.

The results of ^{15}N and ^{14}C inclusion were used for estimating the annual turnover rate of nitrogen in OM as well as the contribution of specific individual substances to the carbon and nitrogen cycle. The annual turnover rates of nitrogen and carbon in organic fractions were calculated by using the

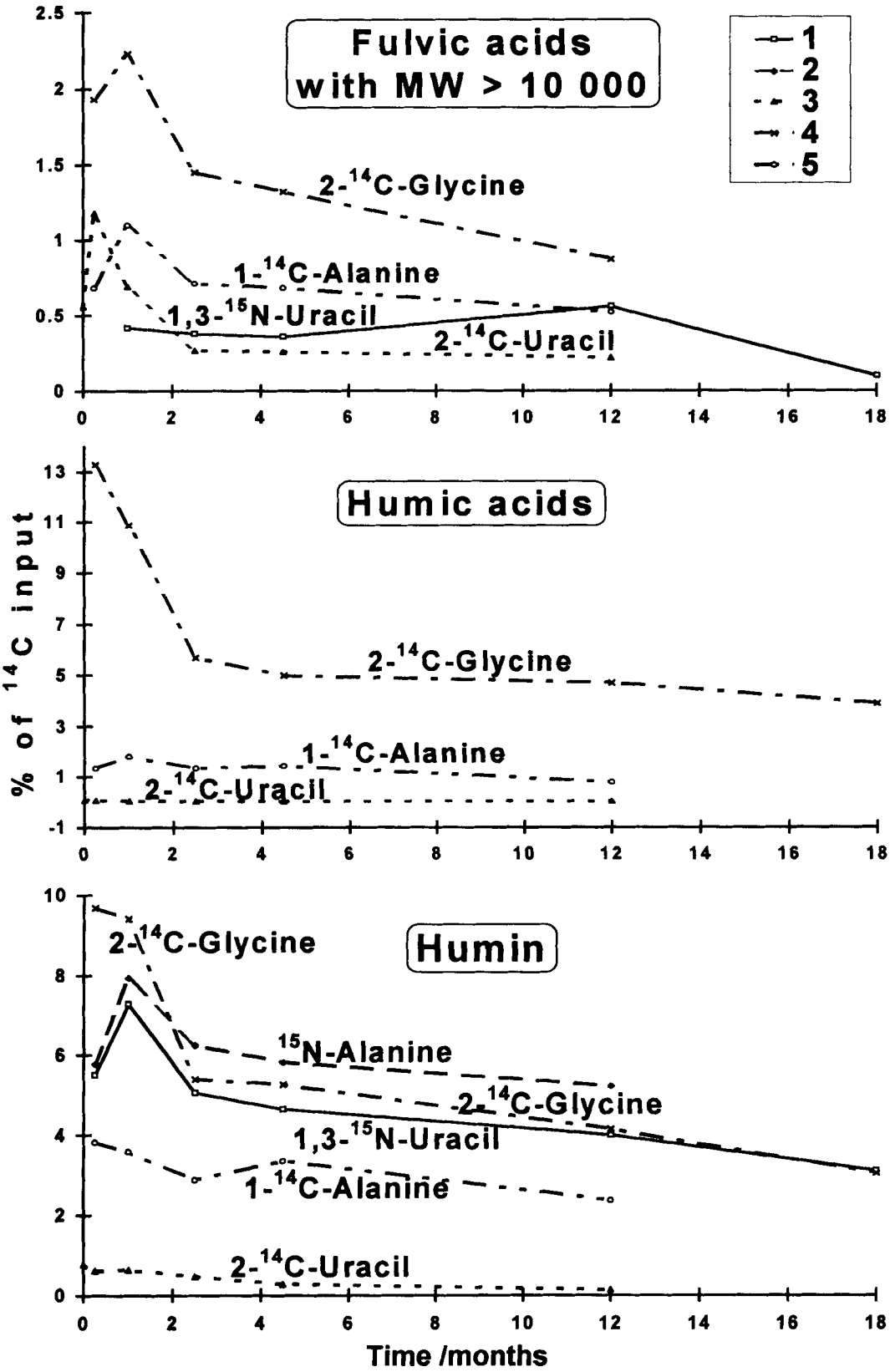

Fig. 3 Dynamics of ^{14}C and ^{15}N labelled decomposition products of uracil, glycine and alanine in fulvic acids, humic acids and humin. (1-1,3-^{15}N-uracil, 2-^{15}N-alanine, 3-2-^{14}C-uracil, 4-2-^{14}C-glycine, 5-1-^{14}C-alanine).

quantity of labelled nitrogen and carbon included into the different organic fractions 1 year after amino acid or uracil addition to the soil. The rates of microbiological decomposition of the added substances and the annual input of amino acids and nucleic bases entering the soil were also calculated. The last index was calculated taking into account the mean concentrations of amino acids and nucleic bases in soil and their microbiological decomposition rates, as follows. For amino acids: mean concentration is 5 $\mu g\ g^{-1}$ soil (Mamchenko, 1970; Umarov & Aseeva, 1971); half-life decomposition period for amino acids (calculated from of the losses of carbon from carboxyl group in natural conditions) is 2.5 days (Fokin, 1992). By assuming that the quantity of amino acids decomposed per year is equal to the amount synthesized then 30 g/100 g and 0.15 g/100 g soil of amino acids and nucleic bases (Efremov, 1988; Samko, 1983) would be synthesized per year, respectively.

The annual turnover rate was calculated as follows:

$$t_r = \frac{1}{t_{mr}} = \frac{H_c \times H}{c \times t},$$

where t_r is the turnover rate, t_{mr} is the mean residence time of isotopic label from the substance investigated in the organic fraction, H_c is the fraction of the label incorporated in the organic fraction (humification coefficient), t is the time in years after addition (1 year in this experiment), M is the atom mass renewed in 1 year in the fraction investigated (in this experiment: mass of ^{14}C or ^{15}N labelled substance incorporated in humic fraction after 1 year), and c is the total atom concentration (C or N) in the investigated organic fraction.

We assumed that the humification coefficient of amino acids and the nucleic bases not studied in the experiment was approximately the same as for glycine, alanine or uracil. The calculated turnover rate of carbon in the humic fraction was compared to the mean residence times (mrt) for Podzoluvisol soils determined by radiocarbon dating (Jenkinson & Rayner, 1977; Chichagova, 1987). The quantity t_{mr}^{-1} in the equation, represents the annual turnover rate of the organic fractions. It is the reciprocal of the mean residence time (mrt). The results obtained are presented in Table 4.

Nitrogen inputs to the soil in native ecosystems are mainly due to proteins and nucleic acids, therefore the overall turnover of the added amino acids and nucleic bases may be assumed to correspond to the overall turnover of N in soil.

On the results of C and N turnover of humic fractions due to the added amino acids and nucleic bases (Tab. 4) it is possible to make the following conclusions.

The overall turnover of N in the organic structure is two to three times faster than that of C (C turnover was calculated using radiocarbon mrt). This is due to the faster turnover of peripheral parts of humic molecules (Jenkinson & Rayner, 1977) in which the content of N is larger than that of central part of the molecule (Orlov, 1985). The rate of turnover of amino acids in the humus is about twice that of nucleic bases and other N containing organic substances.

The differences in turnover rates between amino acid and nucleic bases may explain the selective participation of amino acids in the humification and the formation of organic structures. This is a result of the larger concentration of amino acids in the soil (Umarov & Aseeva, 1971; Aseeva *et al.*, 1978; Samko, 1983; Cortez & Billes, 1987; Efremov, 1988), as well as the slow microbiological transformation of nucleic bases compared to that of amino acids (Fokin *et al.*, 1992).

The renewal of HA is the most rapid, and the turnover of the humin is slowest. This fact contradicts the radiocarbon dating results for some Podzoluvisols. Amino acids play the most important role in the turnover of peripheral parts of HA molecules. The incorporation of the amino acids in the molecules of the humin and of the FA and the turnover of these two fractions due to amino acids are small in comparison to the turnover of HA.

We could detect no significant differences in turnover rates of FA fractions with different molecular masses. The nucleic bases play an important role in the turnover of FA.

The position of a C atom in the molecule of the substances investigated may be responsible for different turnover rates and incorporation processes. For example, the participation in the OM renovation by C in the pyrimidine ring of uracil, or in the carboxyl or methylene groups of amino acids gives the following comparative contribution of 1 : 100 : 400.

Table 4 Literature values of mean residence times (mrt—second column) and present results of carbon and nitrogen annual turnover in soil organic fractions due to amino acids and nucleic bases.

			Parts of C or N annual turnover / $year^{-1}$; calculated due to:				
Fractions	mrt / years	mrt	2C-Nucleic bases	2C-Amino acids	1C-Amino acids	N-Nucleic bases	N-Amino acids
Humus (total)	300	3×10^{-3}	2×10^{-6}	5×10^{-4}	2×10^{-4}	—	—
Humin	500	2×10^{-3}	7×10^{-8}	2×10^{-4}	10^{-4}	10^{-4}	7×10^{-3}
Organo-mineral colloids	—	—	10^{-6}	3×10^{-4}	5×10^{-5}	4×10^{-5}	10^{-2}
Humic acids	300	3×10^{-3}	10^{-7}	2×10^{-3}	6×10^{-4}	10^{-4}	2×10^{-2}
Fulvic acids (total)	100	10^{-2}	2×10^{-6}	8×10^{-4}	2×10^{-4}	—	—
MW = 700–3000	—	—	3×10^{-6}	7×10^{-4}	2×10^{-4}	10^{-4}	—
MW > 10000	—	—	10^{-6}	6×10^{-4}	2×10^{-4}	10^{-4}	—

These results indicate the necessity to differentiate the role of various molecules fragments as well as the position of atoms in the molecule in OM turnover.

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