Thermochimica Acta, 170 (1990) 67–80
Elsevier Science Publishers B.V., Amsterdam

THERMOGRAVIMETRIC AND IR ANALYSIS OF DIFFERENT EXTRACTS OF HUMIC SUBSTANCES

L. CAMPANELLA * and M. TOMASSETTI *

Dipartimento di Chimica, Università "La Sapienza", Piazza A. Moro 5, 00185 Roma (Italy)

A. PICCOLO

Istituto Sperimentale per lo Studio e la Difesa del Suolo, Piazza D'Azeglio 30, Firenze (Italy)

(Received 28 February 1990)

ABSTRACT

TG and IR analyses were used to evaluate the best extracting agents of humic substances from the soil, among two traditional extractive solutions and other new solutions recently proposed. The two instrumental techniques considered were found to be useful and complementary to each other in satisfactorily characterising the obtained extracts as such or after a further purification treatment.

INTRODUCTION

Thermal analysis may be profitably used for many purposes, for instance to investigate the efficiency of physical and chemical treatments, of chemical preparations, and of many analytical procedures such as precipitation, crystallisation, extraction [1,2].

This note deals with the extraction of humic substances contained in a soil and with their qualitative and quantitative characterisation, in order to evaluate the soil environmental reactivity [3].

Humic substances can be extracted from soils by a number of extractants, the most traditional being NaOH solution, under an N_2 atmosphere, and sodium pyrophosphate solution, at pH 7. Recent work [3,4] used aqueous hydrochloric mixtures of dipolar aprotic solvents to extract a selective fraction of humic substances related to the microbial dynamics of soil organic matter. In this paper thermogravimetric analysis and infrared spectrometry are used to characterise humic materials extracted with different solutions and to detect the changes brought about by the HF and HCl solution purification treatment.

---

* Authors to whom correspondence should be addressed.

0040-6031/90/$03.50

EXPERIMENTAL

Materials and methods

The soil used for this study was sampled from the surface layer of a peat developed in the ancient river bed of Lake Bientina near Lucca (Italy), classified as Dystric Histosol [5]. The soil was air dried and passed through a 2 mm sieve. Humic substances were extracted with the following five extractants: (a) acetone–HCl (0.6 N aqueous solution), (b) dimethylformamide (DMF)–HCl (0.6 N aqueous solution) and (c) dimethylsulphoxide (DMSO)–HCl (0.6 N aqueous solution), all in the ratio 8 : 2 by volume; (d) NaOH (0.5 M) under an N_2 atmosphere; (e) $Na_4P_2O_7$ (0.1 M) at pH 7. For the last two extractants, humic acids were obtained by precipitating the material at pH 1 with concentrated HCl, centrifuging at 5000 *g* dialysing (Kalle dialysing membranes) against distilled water and freeze drying. For the organic mixtures, humic extracts were obtained by directly dialysing the extracts and freeze drying. The five extracts, obtained as above, were successively purified by treatment with an HF and an HCl aqueous solution, both containing 0.5% by volume, to reduce their inorganic content [6].

All the reagents, of analytical grade, were supplied by Carlo Erba, Milan (Italy).

Apparatus

The TG and DTG curves of the solid extracts were obtained with a Mettler TG 50 thermobalance coupled with a Mettler TC 10A-TA processor system and a Swiss dot matrix printer. The heating rate was 10 ° C min^{-1}; the atmosphere was an air stream with a flow rate of 100 cm^3 min^{-1}.

Infrared spectra were recorded using a Perkin–Elmer 882 double-beam IR spectrophotometer. Samples for IR analysis were prepared by the KBr pellet technique [7].

RESULTS AND DISCUSSION

TG curves of the non-purified extracted samples from acetone (a), dimethylformamide (b) and dimethylsulphoxide (c) mixtures (Figs. 1–3), after the initial loss of water (between room temperature and about 140 ° C), are characterised by a decomposition process between 160 ° C and 500 ° C occurring in two main steps, the first at about 250 ° C and the second between 300 ° C and 350 ° C. It is observed (Table 1) that, on passing from extract a to extract c, the percentage mass loss decreases for the first of the two steps, while it increases for the second step. The TG curves of the same extracts after treatment with aqueous HF and HCl solution show (Figs. 4–6), after

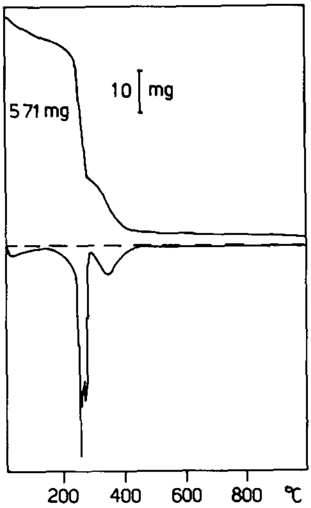

Fig. 1. TG and DTG curves of humic extract from acetone mixture, in flowing air (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}.

the initial water loss, practically only one strong decomposition step, centred at a temperature that increases on going from extract a (280 °C), to extract b (320 °C), to extract c (360 °C) (Table 2). The residue at 1000 °C corresponds to about 9% by weight for extract a, to 7% by weight for extract b and to 10% by weight for extract c; after treatment with HF and HCl solution, these values decrease to respectively 3.5%, 2% and 2.6% by weight. TG analysis shows conclusively that the extracts a, b and c are constituted of an

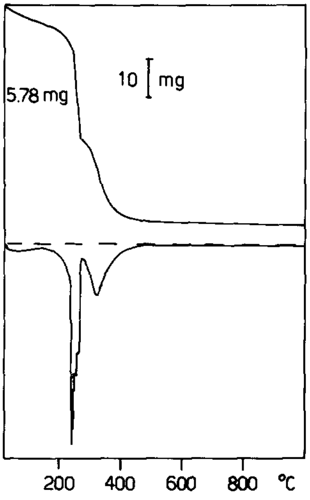

Fig. 2. TG and DTG curves of humic extract from dimethylformamide mixture, in flowing air (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}.

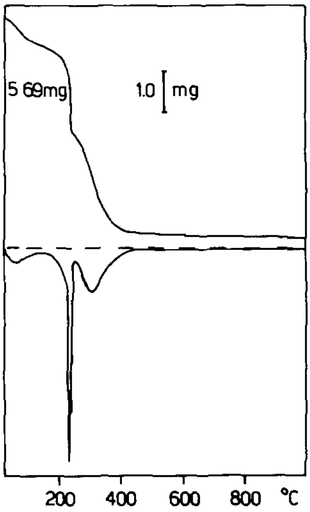

Fig. 3. TG and DTG curves of humic extract from dimethylsulphoxide mixture, in flowing air (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}.

inorganic residue of quite a modest amount (7–10%), which is reduced to 2–3% by treatment with HF and HCl solutions and an organic fraction ($\geq$ 90% by weight), decomposed in a similar way for all three extracts. Inspection of Tables 1 and 2 induces us to think that the compounds extracted in mixtures based on dipolar aprotic solvents are probably almost the same, but with different percentages of the extracted substances. All this is confirmed by the analysis of IR spectra of the dried extracts a, b, c, both treated and untreated with HF and HCl solution, and by the analysis of IR spectra of the TG residues of non-purified extracts at the end of the first TG steps and of IR spectra of the TG residues at 1000 °C (Figs. 7–9).

IR spectra of unpurified extracts dried by heating at 140 °C show similar behaviour (i.e. the stretching band of the double bond of both olefinic conjugated systems and aromatic systems [7]), with the only exception of the large band, at about 1050 cm^{-1}, due to the Si–O stretching, which is more marked for extract a from the acetone mixture, than for extracts b and c. IR spectra of extracts purified by HF and HCl solutions and dried after heating at 140 °C show more intense bands at about 2900 cm^{-1} due to extract $-CH_2$ and $-CH_3$ stretching (more evident in the case of extract a, than in extracts b and c) and an intense band at 1715 cm^{-1}, due to C=O stretching of carboxylic groups, caused by the replacement of mixed humic–potter's clay salts by the acidic treatment; finally, the absorption of the Si–O group at about 1100–1000 cm^{-1} decreases.

IR spectra of TG residues (of unpurified extracts) at the end of the first step (i.e. heated at about 260–300 °C) are sufficiently different: the extract from the acetone mixture (a) shows almost complete disappearance of bands at 1715 cm^{-1} and at 1600 cm^{-1} and the simultaneous enhancement of the

TABLE 1

Thermal analysis of humic substances extracted by different mixtures and of untreated soil

Extracting mixture	Moisture (loss %)	pdt	First step loss (%)	pdt	Second step loss (%)	pdt	Third step loss (%)	pdt	Residue at 1000 °C (%)
Acetone–HCl (0.6 N)	7.4	24	63.9	160	23.7	295	–	–	8.6
		40		260		350			
		130		295		450			
DMF–HCl (0.6 N)	7.1	24	54.9	160	36.4	280	–	–	6.6
		70		245		320			
		140		280		480			
DMSO–HCl (0.6 N)	11.5	24	42.4	160	44.1	260	–	–	9.8
		60		230		300			
		140		260		470			
Sodium hydroxide (0.5 M)	5.0	24	20.2	160.	45.2	350	–	–	33.5
		55		325		430			
		140		350		650			
Pyrophosphate (0.1 M)	6.5	24	14.0	160	14.9	360	28.2	650	42.1
		40		280		425		950	
		140		360		580		1000	
Untreated soil	12.3	24	10.6	200	11.7	375	2.4	660	75.1
		165		320		430		990	
		200		375		660		1000	

TG in air stream (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}. The percentage losses by weight at the three different steps and of the residues at 1000 °C refer to the weight of the dry samples, while percentage losses of moisture refer to the total weight of the sample. pdt, procedural decomposition temperature.

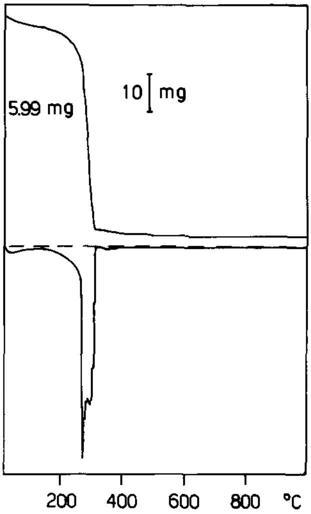

Fig. 4. TG and DTG curves of humic extract from acetone mixture purified by treatment with HF and HCl solution, in flowing air (100 cm^3 min^{-1}), heating rate 10 ° C min^{-1}.

$-CH_2$ and $-CH_3$ stretching bands; this suggests that the extract is mainly constituted of fatty unsaturated acids for which, at about 280 ° C, decarboxylation and unsaturation loss occur. On the contrary, IR examination of TG residues at about the same temperature of the extracts from DMF (b) or DMSO (c) mixtures provide evidence that in any case some decarboxylation occurs although the band at 1600 cm^{-1} is still present; this suggests that the

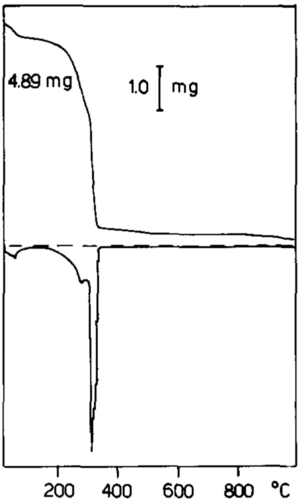

Fig. 5. TG and DTG curves of humic extract from dimethylformamide mixture purified by treatment with HF and HCl solution, in flowing air (100 cm^3 min^{-1}), heating rate 10 ° C min^{-1}.

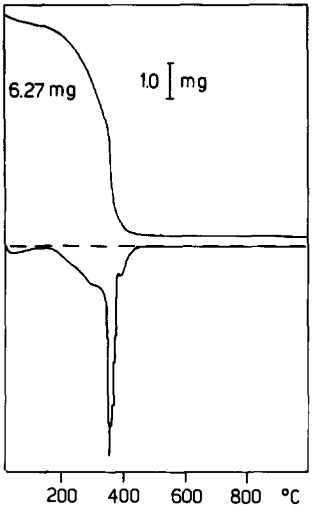

Fig. 6. TG and DTG curves of humic extract from dimethylsulphoxide mixture purified by treatment with HF and HCl solution, in flowing air (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}.

absorbing band has probably to be assigned to C=C aromatic stretching of polycyclic structures, conjugated to C=O [8] still undecomposed at about 260–280 °C.

IR results, however, agree with TG behaviour: in particular, the unpurified extract a seems to be richer in fatty unsaturated aliphatic acids and therefore the substantial mass loss occurs during the first TG step, at lower temperature, while for unpurified extracts b and c, richer in compounds with aromatic–quinonic polycondensed groups, the large mass loss in the TG curves can be observed during the second step at higher temperatures. The treatment with HF and HCl solution results in a homogenisation of the extracts, so that all the mass loss in the TG curves occurs practically in only one large step, shifted to higher temperatures, so that the content of polycyclic, quinonic and phenolic groups persisting after the purification process with HF and HCl solution is very high.

With regard to the extracts obtained with NaOH solution (d), or with pyrophosphate solution (e), the TG curves (Figs. 10 and 11) show, after the water loss, the characteristic two steps, although they are shifted to higher temperatures (about 300 °C and 430 °C respectively); the first is less well resolved in the case of the extract from NaOH solution. These TG curves differ from those of the previous extracts by the substantial content of inorganic residue (Table 1), which a large decomposition step between 900 °C and 1000 °C in the TG curve of the extract from pyrophosphate solution is also related to; in fact, the main difference between the TG curves of the two latter extracts is the higher percentage of still undecomposed material between 550 °C and 800 °C that can be observed in the TG

TABLE 2

Thermal analysis of humic substances, firstly extracted by different mixtures, then purified by HF and HCl aqueous solution

Extracting mixture; purification treatment	Moisture loss (%)	pdt	First step loss (%)	pdt	Residue at 1000 °C (%)
Acetone–HCl (0.6 N);	4.5	24	95.9	160	3.8
HF and HCl solution		40		280	
		140		400	
DMF–HCl (0.6 N);	7.8	23	93.1	150	2.1
HF and HCl solution		60		320	(4) [b]
		140		400	
DMSO–HCl (0.6 N);	5.1	24	96.3	160	2.5
HF and HCl solution		45		360	
		140		450	
Sodium hydroxide	12.3	24	39.8	160	
(0.5 M);		55		280 (A) [a]	
HF and HCl solution		140		375	
			47.3	375	
				430 (B) [a]	8.8
				650	
Pyrophosphate (0.1 M);	10.5	23	96.3	145	3.4
HF and HCl solution		60		380	
		140		490	

TG in air stream (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}. The percentage loss by weight at the first step and the percentage by weight residues at 1000 °C refers to the weight of the dry samples, while percentage loss of the moisture refers to the total weight of the sample. Pdt, procedural decomposition temperature.

[a] (A) and (B) really refer to two separate steps; they are reported in such a way to homogenize the table.

[b] Per cent by weight residue at 800 °C.

curve of the extract from pyrophosphate solution. The purification by treatment with HF and HCl solution drastically reduces the amounts of these undecomposed materials (Figs. 12 and 13 and Table 2) with higher efficiency for the extract from pyrophosphate solution. This suggests that this undecomposed material mainly consists of minerals, such as potter's clay, which can be removed during the purification treatment through the formation of soluble fluorosilicic complexes [9]; see also the IR spectrum (Fig. 14).

The TG and DTG curves of purified extracts d and e show steps that are better resolved than those of the corresponding unpurified extracts. That is, in the TG and DTG curves of the purified extracts from NaOH solution two main steps at 280 °C and 430 °C can easily be identified, while the TG and

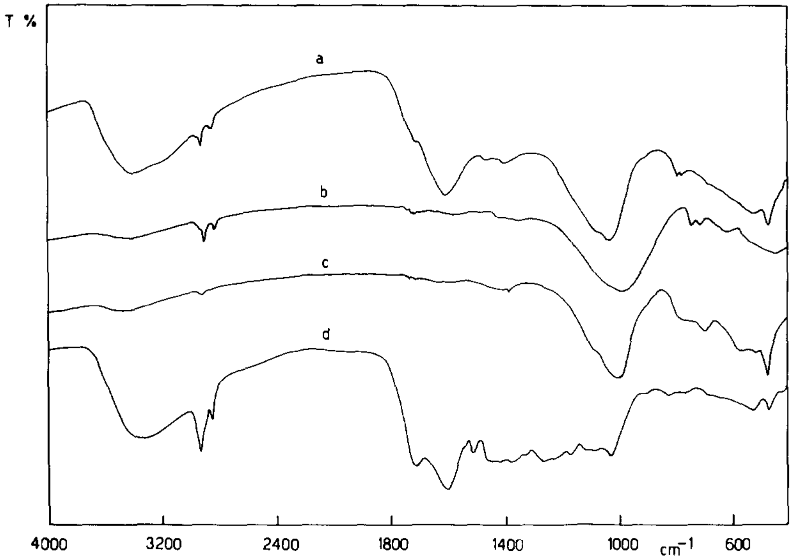

Fig. 7. IR spectra in KBr of humic extract from acetone mixture: a, dried by heating at 140 ° C; b heated at the end of the first TG step; c, heated at about 1000 ° C; d, purified by treatment with HF and HCl solution and dried by heating at 140 ° C.

DTG curves of the purified extract from pyrophosphate solution are very similar to those of purified extracts a, b and c showing only a substantial step, although at quite a higher temperature (380 ° C) (Table 2). In the IR

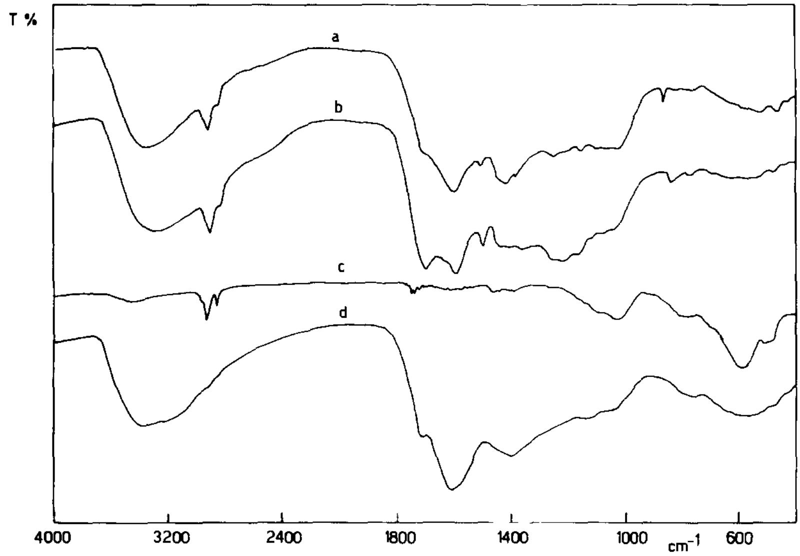

Fig. 8. IR spectra in KBr of humic extract from dimethylformamide mixture: a, dried by heating at 140 ° C; b, heated at the end of the first TG step; c, heated at about 1000 ° C; d, purified by treatment with HF and HCl solution and dried by heating at 140 ° C.

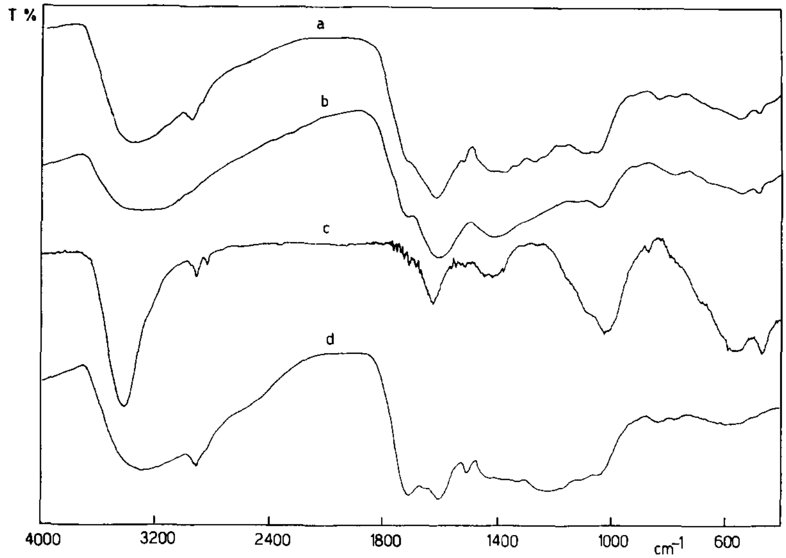

Fig. 9. IR spectra in KBr of humic extract from dimethylsulphoxide mixture: a, dried by heating at 140 °C; b, heated at the end of the first TG step; c, heated at about 1000 °C; d, purified by treatment with HF and HCl solution and dried by heating at 140 °C.

spectra of TG residues at about 300 °C of unpurified extracts d and e (Figs. 14 and 15), the disappearance of the bands near 1430 cm^{-1} due to the "C–H deformation" of methylenic and methylic groups and to "deformation" of the C–O and O–H bonds of alcoholic and polysaccharic groups and that of the band of 1100 cm^{-1} (primary alcohols) can be observed. All this is less

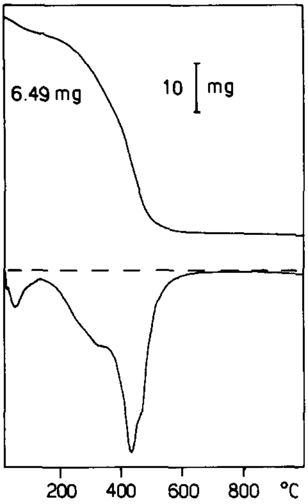

Fig. 10. TG and DTG curves of humic extract from NaOH solution, in flowing air (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}.

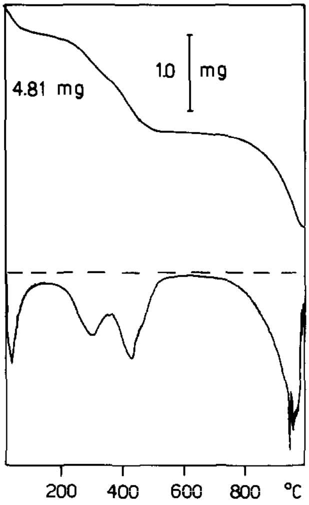

Fig. 11. TG and DTG curves of humic extract from pyrophosphate solution, in flowing air (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}.

evident in the IR spectra of the extracts from pyrophosphate solution; however, in these spectra the presence of the Si–O group is more evident (see bands at about 1100–1000 cm^{-1}). All these IR spectra, and those of TG residues at higher temperatures, show also the band at 1600 cm^{-1} of C=C aromatic stretching, conjugated by C=O and O–H groups, that demonstrate that polycyclic aromatic structures are still present in these TG residues. Lastly, the IR spectra of TG residues at 1000 °C show that only traces of

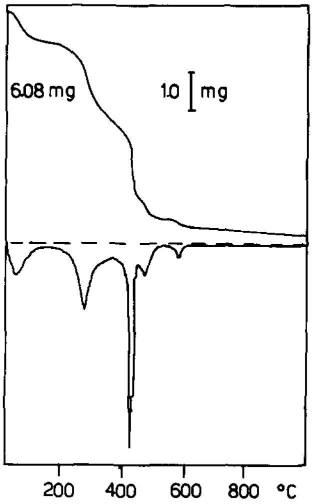

Fig. 12. TG and DTG curves of humic extract from NaOH solution purified by treatment with HF and HCl solution, in flowing air (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}.

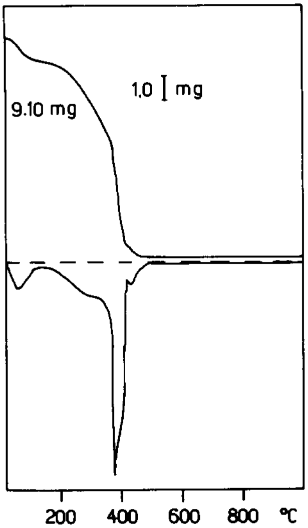

Fig. 13. TG and DTG curves of humic extract from pyrophosphate solution purified by treatment with HF and HCl solution, in flowing air (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}.

silicates are still present, as confirmed by X-ray powder spectra. Finally, it can be noted that TG techniques rapidly and easily reveal the high molecular complexity of the extracts from NaOH or pyrophosphate solution, in comparison with extracts a, b and c obtained with dipolar aprotic mixtures. Moreover, it can be noted that the TG curves of extract e (Fig. 11) obtained

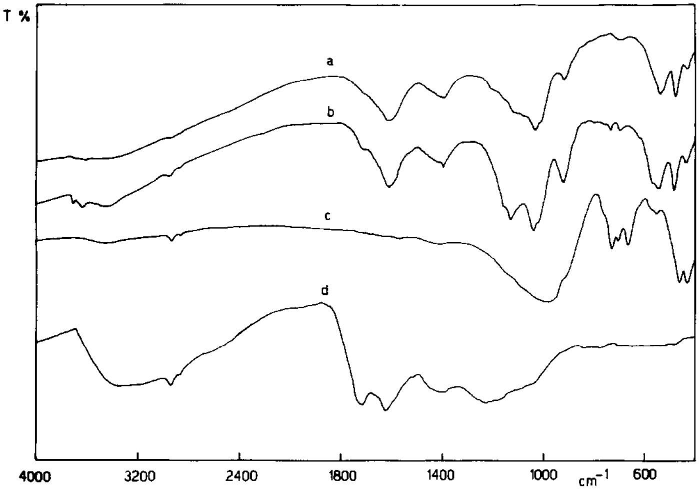

Fig. 14. IR spectra in KBr of humic extract from pyrophosphate solution: a, dried by heating at 140 °C; b, heated at the end of the first TG step; c, heated at about 1000 °C; d, purified by treatment with HF and HCl solution and dried by heating at 140 °C.

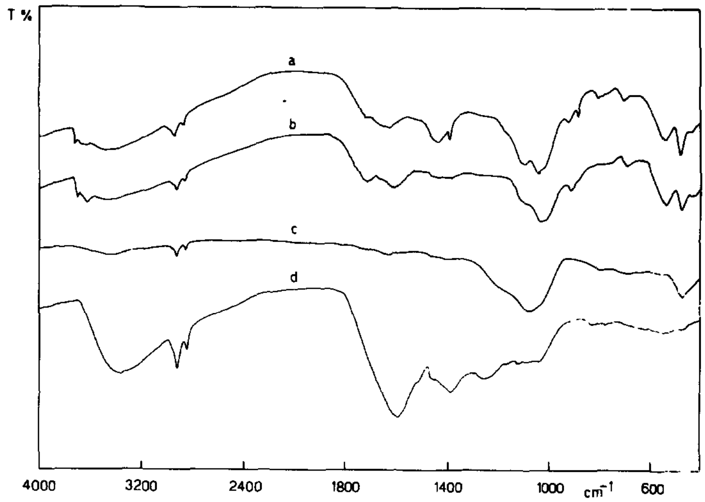

Fig. 15. IR spectra in KBr of humic extract from NaOH solution: a, dried by heating at 140 °C; b, heated at the end of the first TG step; c, heated at about 1000 °C; d, purified by treatment with HF and HCl solution and dried by heating at 140 °C.

by pyrophosphate solution and of the untreated soil (Fig. 16) are not very different and this immediately suggests that this kind of extraction is less efficient in performing a separation of the different fractions of the humic substances. A similar remark can be made on observing the behaviour of TG curves of unpurified and purified extracts from NaOH solution.

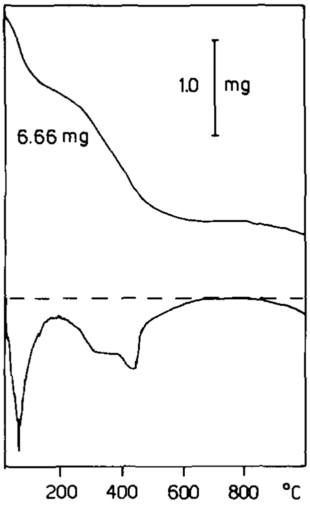

Fig. 16. TG and DTG curves of untreated soil, in flowing air (100 cm^3 min^{-1}), heating rate 10 °C min^{-1}.

CONCLUSIONS

TG techniques coupled to IR analysis resulted in the ability to differentiate sufficiently and to characterise humic substances extracted from the soil by different extracting agents.

The extracts from dipolar aprotic solvents (acetone, DMF or DMSO) mixtures, especially after purification with HF and HCl solution, were very poor in inorganic matter and contained different percentages of aliphatic chains of fatty unsaturated acids, highest for the extract from the acetone mixture and lowest for the extract from the DMSO mixture, while, the contrary is observed for the content of aromatic, quinonic and phenolic groups. These compounds are more evident in the extracts with NaOH or pyrophosphate solutions.

Pyrophosphate solutions used as extracting agents resulted in a selectivity intermediate between those of dipolar aprotic mixtures and of NaOH solution, the latter extracting humic substances with greater heterogeneous molecular complexity.

ACKNOWLEDGEMENT

This work was supported by the National Research Council (CNR) of Italy.

REFERENCES

1 C. Duval, Inorganic Thermogravimetric Analysis, Elsevier, New York, 1953.
2 L. Campanella and M. Tomassetti, Inquinamento, (Milano) (7–8) (1987) 81.
3 A. Piccolo, Soil Sci., 146, (1988) 418.
4 A. Piccolo and A. Mirabella, Sci. Total Environ., 62 (1987) 39.
5 D. Magaldi, D. Bidini, C. Calzolari and G. Rodolfi, Ann. Ist. Sper. Stud. Dif. Suolo (Florence), 14 (1983) 21.
6 A. Piccolo, Ann. Ist. Sper. Stud. Dif. Suolo (Florence), 17 (1986) 189.
7 A. Piccolo and F.J. Stevenson, Geoderma, 27 (1982) 195.
8 L.J. Bellamy, Infrared Spectra of Complex Molecules, Wiley, New York, 1975.
9 F.J. Stevenson, Humus Chemistry: Genesis, Compositions and Reactions, Wiley-Interscience, New York 1982.