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EVALUATION OF BIOLOGICAL ACTIVITY OF CELLULOSE PULP BY MEANS OF THE STATIC RESPIRATION INDEX (AT_4)

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Abstract

In the countries of the European Union, work is presently being conducted on the standardisation of the limit values and test methods for the determination of the biological activity of waste. The aim of conducting the tests is to monitor the effectiveness of waste biodegradation during composting, the evaluate any decrease in the biological activity of the waste before its landfilling and control processes taking place at landfills. The evaluation of the waste's biological activity can be performed, among others, by testing respiration. One such method is AT_4 (Static Respiration Index) determination. The results of respirometric tests depict the availability of substrates for microorganisms, that is, the biodegradability.

The article describes the tests of the biological activity of the cellulose pulp, the impact of the degree of compost inoculation on the value of this parameter and the dependence on the content of organic mass and total organic carbon in the tested substrate. The measurements of the oxygen demand were made using the OxiTop® Control measuring system.

Keywords: cellulose pulp, effectiveness of waste biodegradation, AT_4, methods standardization

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1. INTRODUCTION

In order to assess the degree of biological stabilisation of waste in Poland, the determination of TOC, losses on ignition and respiration activity (AT_4) (Journal of Laws from 2012 No. 0, item 1052) are used [10]. Stabilised material may be landfilled at waste landfills, if it meets one of the following criteria:

- losses on ignition < 35 % of dry mass,
- TOC content < 20% of dry mass,
- loss of organic matter in the stabilised material in relation to the organic matter in the waste, measured by the loss on ignition or TOC content: > 40%,
- AT_4 content < 10 mg O_2/g of d.m.

Assessment of waste biodegradability may also be used for the purposes of:

- monitoring the effectiveness of the biodegradation process in the production of high quality compost from separately collected organic fractions of biodegradable municipal waste or other biodegradable waste,
- assessment of the reduction in the biological activity of solid waste stabilised in aerobic and anaerobic conditions,
- monitoring the impact of aeration of landfilled waste on the reclamation of old landfills [2, 5].

As a result of biological waste treatment, the organic fraction, being subject to biodegradation, becomes decomposed, partly mineralised and partly transformed into a new organic substance characterised by a higher stability, which may also be determined as the loss on ignition or the TOC content. Thus, the difference between the determined losses on ignition or TOC content in waste before subjecting it to biological processes, and stabilised material after the process of biological treatment, presents the total loss of organic substance as a result of mineralisation, and its specific level may constitute grounds on which to claim that the stabilised material no longer contains biodegradable fractions with gas-producing potential [9].

The indicators for the assessment of the effectiveness of aerobic and anaerobic processes, that is biochemical tests, help to determine the loss of the capacity of the waste to further decompose biologically during these processes [11].

The methods of biodegradation assessment comprise incubation of organic waste in the presence of microorganisms, which use the organic substance as the substrate for their growth. The method of determination of the respiration activity serves the purpose of assessment of the biological reactivity or the degree of maturity of the stabilised materials (composts) in the aerobic conditions. It determines the quantity of oxygen used by microorganisms during a specific time under laboratory conditions. Using this method, the oxygen demand may be specified by the measurement of the quantity of consumed oxygen or produced carbon dioxide and is expressed in g O_2/kg of dry mass of waste.

The aerobic tests may be divided into three main types. These are methods which specify the static respiration indices (e.g. SRI), dynamic respiration indices (e.g. DRI) and the speed of oxygen uptake rate (e.g. SOUR) [1, 3]. Table 1 presents selected tests conducted under aerobic conditions [4]. At present, the respiration indices are regarded as the basic parameters which serve the purpose of assessment of the compost stability, including its quality.

Table 1 Waste biodegradation assessment tests conducted in aerobic conditions

method	test	environmental conditions				sample preparation	evaluation of results	unit
		temperature °C	time	humidity	size of sample			
ASTM	D	58	4d	50%	500 g	n.d.a.	compost stability	n.d.a.
SOUR	S	30	5-6h	suspension	3-8 g	n.d.a.	n.d.a.	n.d.a.
OD20	S	30	20h	suspension	3-8 g	compost	compost stability	gO_2/h/g d.m
DSOUR	S	30	20h	without correction	3-8 g	< 9,5 mm	n.d.a.	n.d.a.
DRI	D	environmental conditions	24h	750 g/kg	n.d.a.	n.d.a.	n.d.a.	n.d.a.
RDRI	D	environmental conditions	53h	without correction	20-40 kg	50 mm	compost stability	mgO_2/kg/h
PDRI	D	environmental conditions	53h	without correction	n.d.a.	n.d.a	n.d.a.	mgO_2/kg/h
SRI	S	environmental conditions	24h	750 g/kg	n.d.a.	n.d.a.	n.d.a.	mgO_2/kg/h
Solvita	S	20-25	4h (+48 h)	saturation	to fill the vessel	removal of stones and others	compost stability	scale 1-8
DR_4	D	37	4d	50% m/m	200-250 g s.m.	BMW are not removed	performance monitoring MBT	mgO_2/kg
RI_T	S	environmental conditions	4h	40-50% m/m	250 ml	BMW crushing	compost stability	mgO_2/g/h
RI_{37}	S	37	18h	n.d.a.	250 ml	sieving < 10 mm	compost stability	mgO_2/g/h
AT_4	S	20	4d	saturation 40-50%	30-40 g	< 20 mm	biological activity, subject to landfilling regulations	mgO_2/g d.m
AT_7	S	20	7d	saturation 40-50%	30-40 g	< 20 mm		mgO_2/g d.m

S – static method, D- dynamic method, d.m. - dry mass, n.d.a. – no data available, BMW- Biodegradable Municipal Waste, MBT - Mechanical-Biological Treatment

In aerobic conditions, biodegradable substances are subject to decomposition into carbon dioxide, water and organic salts, and the energy is retained in the form of ATP (dissimulatory metabolism) [7]. The organic substances are processed into biomass ($C_5H_7O_2N$), whereby ATP is used again as the source of energy (catalytic metabolism). Both types of metabolism are bound by NADP which supplies or takes up electrons (bonded hydrogen), if needed. The consequence of the “divided” metabolism is that only part (usually from 40 to 60%) of the calculated theoretical oxygen demand (TOD) is used in the decomposition of a specific amount of substrate [7].

The amount of used oxygen may be specified manometrically, based on the measurement of the carbon dioxide produced during the biodegradation process. The carbon dioxide is bonded by the absorbent agent (e.g. sodium hydroxide, caustic soda solution, soda lime or potassium hydroxide) and does not appear in the form of free gas:

$$CO_2 + 2NaOH \rightarrow Na_2CO_3 + H_2O \qquad (1.1)$$

As a consequence, the change in pressure is assigned only to oxygen consumption. In order to be able to measure the oxygen consumption by means of a manometer, the following conditions must be fulfilled:

- a biologically active sample must be placed in a vessel which is impermeable to gas,
- a sufficiently large air space must be maintained above the sample, which will ensure the unlimited access of oxygen to the biodegradation processes,
- the measuring vessel must contain the agent that absorbs carbon dioxide, but the sample may not come into contact with the agent.

Based on equation 1, it is possible to calculate the stoichiometric amount of the absorbent agent, which must be inside the reaction vessel, needed for covering the whole measuring range. An excessively small amount of the absorbent agent leads to an underestimation of results. If the carbon dioxide was absorbed too slowly and the free CO_2 collected over the waste sample, the change in pressure will not be proportionate to oxygen consumption.

In reference to the correct assessment of the loss of the capacity of the waste for further biodegradation, in Germany, Austria and Poland, *Static respiration test* (AT_4) was considered the most appropriate parameter.

AT_4 is a parameter that determines the demand for oxygen necessary for the decomposition of waste within four days. Table 2 presents the value of AT_4 for the selected components of municipal waste [3].

Table 2. AT_4 index values for the selected components of municipal waste

Component of municipal waste	AT_4 (gO_2/kg d.m.)
cellulose	84.9
newspapers	76.6
corrugated paper	12.5
grass	119.0
twigs	57.1
plants	137.0
meat	150.0
cotton	12.9
wool	17.0
disposable nappies	86.1
compost	26.3

Not only does the AT_4 index depict the loss of the total mass organic matter as a result of mineralisation processes, but also a decrease in the capacity of the organic matter to further decompose in the aerobic process.

The article describes the analysis of the biological activity of cellulose pulp, using the AT_4 respiration test, the impact of the degree of compost inoculation on the value of this parameter and its dependence on the content of organic matter and TOC in the tested substrate.

2. METHODOLOGY AND SCOPE OF STUDIES

The waste from paper production (cellulose pulp, photo.1) delivered to the Waste Disposal Plant in Gorzów Wlkp. from Arctic Paper Kostrzyn S.A. where uncoated graphic paper is produced, was used in the biological activity tests. The raw materials used for the production of the paper include: cellulose, calcium carbonate, water, starch and chemical agents. The annual throughput of the paper production plant is approximately 260,000 tonnes.

Municipal compost from the mechanical-biological treatment plant for municipal solid waste and from the separately collected green waste and biowaste treatment plant (composting plant) in Chróścik (photo 1) were used to inoculate the samples of cellulose pulp. The municipal waste treatment technology covers the division of the stream of municipal waste into organic waste, raw material waste, waste with energy properties, as well as separation of the hazardous waste fraction and ballast. It also allows the composting of the biofraction, compost purification as well as treatment and conditioning of secondary raw materials. The first fraction separated at the plant is the so called subscreen fraction with a diameter of 20 mm, which is used as the transfer material at the landfill. The remaining part of the waste is divided into organic fraction, also called biofraction, and oversize fraction (>80 mm). The bio-fraction is cleansed from minor elements such as: plastics, glass and batteries, and then it is directed to the composting hall together with the contaminated waste cardboard. The obtained organic matter is subjected to composting by means of piles aerated for a period ranging from 8 to 12 weeks (depending on the season of the year). The finished compost is cleansed on the compost purification line which consists of a drum screen, ballistic table, cyclone and belt conveyor system. The treated compost reaches the finished compost warehouse, where it is stored for a period of 1-2 months.

Photo 1. Test substrates: cellulose pulp and compost

The biological activity of the cellulose pulp was analysed using the AT_4 test. In order to carry out the respirometric test, a set including the OxiTop® OC 110 controller and MG 2.5 measuring vessels was used. The OxiTop apparatus consists of reaction vessels, adapters with fixing clamps and measuring heads. The stabilised material samples are placed in reaction vessels, which are closed tightly using an adapter with a measuring head fixed outside which contains an electronic vacuum pressure sensor. Inside each vessel, there is an attachment secured to the adapter, on which the CO_2 absorbent (photo 2) is placed. The measurement was performed at a temperature of 20±1°C for samples with a humidity ranging from 40 to 50%.

Photo 2. Oxi-Top set

- Despite the fact that the AT_4 parameter presents the 4-day demand for oxygen by microorganisms, the test itself must be carried out longer – typically for 7 days. This is related to the potential occurrence of the so called lag-phase during the test. 4 phases of microorganism development can be distinguished [8]:
- Phase 1 - Primary inhibition phase, also called the resting or adaptation phase. It occurs during the transfer of the bacterial colony into a new environment and during changes in the environmental conditions (as, for instance, at sample defrosting).
- Phase 2 - Intensive development phase, also called the logarithmic growth phase. The cells divide intensively, taking advantage of the favourable conditions.
- Phase 3 - Equilibrium (stationary) phase, in which equilibrium is established between the newly formed cells and the dead cells. The quantity of available nutrients is limited then by the number of cells.
- Phase 4 - Death (decline) phase. This occurs when the sources of nutrients begin to deplete and/or the concentration of metabolism products increases to a level which is harmful to the bacteria themselves.

During the AT_4 measurement, the impact of the first phase and partly the second phase, when the amount of microorganisms is too small to accomplish the full “metabolic efficiency”, is responsible for the lag-phase. An assumption has been made that the lag-phase ends when the mean oxygen demand after 3 hours reaches 25% of the 3-hour mean values, occurring at the time of a maximum increase in the oxygen demand, determined for the first four days. The oxygen mass used during the lag-phase is deducted from the oxygen mass taken up during the whole test (lag-phase + 4 days). The oxygen mass from the lag-phase may not exceed 210% of its total demand during the first 4 days.

During the studies, respirometric tests with various degrees of inoculation of the cellulose pulp with compost (40, 60, 70, 80, 90% of dry mass per sample) and tests for the compost (control tests) were performed. Two repetitions per each sample were made.

Photo. 3 Selected cellulose pulp (P) samples inoculated with compost (K)

AT_4, humidity, total nitrogen, total phosphorus, losses on ignition and TOC were determined in the studied waste.
Upon determination of the primary moisture at the stage of sample preparation, before the analysis itself, the sample was moistened accordingly. The appropriate moistening of the sample has a significant impact on the obtained results. An excessively small quantity of water does not ensure optimal conditions for the development of microorganisms to process the given material. An excessive water content, on the other hand, causes the pores inside the studied material to become clogged which prevents the air from free circulation.
The basis for calculations of the AT_4 parameter is the ideal gas law. It follows from the general gas equation that each pressure change is related to a change in the quantity of substance at the constant vessel volume and constant temperature [6]:

$$\Delta p = \Delta n \cdot R' \cdot T \cdot V^{-1} \tag{2.1}$$

where:
Δn – pressure difference
R' – gas constant for CO_2 in normal conditions
T – temperature
V – volume of the measuring vessel

The quantity of substance is a result of the m/M quotient, therefore, the equation takes the following form [6]:

$$\Delta p = \Delta m \cdot R' \cdot T \cdot V^{-1} \cdot M_R^{-1} \tag{2.2}$$

where:
M_R – molar mass of substance.

Equations 2.1 and 2.2 present the correlation between the pressure change and the oxygen consumption.
During measurement, the test vessel should be located in a place characterised by a constant temperature. Changes in the temperature lead to changes in pressure, which gives incorrect the measurements of oxygen consumption. At an initial pressure equal to 1000 hPa and temperature of 20°C (293 K), an increase in temperature by 1°C causes an increase in pressure to 1003 hPa. With the measuring system sensitivity equal to 1 hPa, this means an error which cannot be ignored.
The biological activity (respiration) of waste samples has been calculated on the basis of the following equation:

$$AT4 = \frac{M_R(O_2)}{R \cdot T} \cdot \frac{V_{fr}}{m_{Bt}} \cdot \Delta p \tag{2.3}$$

where:
AT4 - biological activity of a sample (g O_2/kg of dry mass),
M_r (O_2) - molar mass of oxygen: 32000 (mg/mol),
V_{fr} - free gas volume (dm^3),
R - general gas constant: 83.14 ($dm^3 \cdot hPa \cdot (K \cdot mol)^1$),
T - measurement temperature (K),
m_{Bt} - dry mass of substrates in a sample (kg of dry mass),
Δp - pressure drop during measurement (hPa).

The free gas volume is calculated from the difference between all the components present in the reaction vessel and the total volume. The free gas volume has been calculated in accordance with the following formula:

$$V_{fr} = V_{ges} - V_{AG} - V_{AM} - V_{Bf} \tag{2.4}$$

where:
V_{fr} - free gas volume,
V_{ges} - total volume of substrate space in the measuring vessel closed with a cover (without the sample, without the absorption agent, without the absorbent agent),
V_{AG} - volume of the vessel for the absorption agent,
V_{AM} - volume of the absorption agent,
V_{Bf} - volume of the wet sample.

The dry mass of the studied substrate was calculated according to the following formula:

$$m_{Bt} = m_{Bf} \cdot \frac{TS}{100\%} \tag{2.5}$$

where:
m_{Bt} - mass of dry substrate (kg),
m_{Bf} - mass of wet substrate (kg)
TS - content of dry mass in a sample (%).

3. RESULTS OF STUDIES AND DISCUSSION

Before preparation of the samples for the AT_4 respiration test, the cellulose pulp and compost were subjected to tests. The results of these tests have been listed in table 3. The cellulose pulp was characterised by a high percentage of organic matter (96.2%) and pH= 8.55. A smaller percentage of organic matter was found in compost (34%), while the content of total nitrogen and total phosphorus was significantly higher.

Table 3. Physical and chemical characteristics of analysed substrates

substrates	pH	dry matter	organic matter	$N_{Tot.}$	$P_{Tot.}$	TOC
	-	% d.m.				
compost	6.98	85.00	33.90	0.86	0.196	12.6
cellulose pulp	8.55	39.30	96.20	0.11	0.013	43.4

The characteristics of the chemical composition of the cellulose pulp and compost mixtures, with inoculation degrees amounting to 40, 60, 70, 80 and 90% (in relation to the dry mass of waste in a sample) have been presented in table 4. The pH value measured in the obtained mixtures ranged from 7.65 to 7.82.

The proper course of the biodegradation is determined by an adequate percentage of organic carbon and nitrogen in the substrates. In the prepared mixtures, as the share of cellulose pulp in the sample increased, the content of organic carbon also increased and the content of total nitrogen and total phosphorus decreased. This affected the change in the C:N quotient in the range between 20:1 and 76:1, and C:P quotient in the range between 523:1 and 259:1. It is assumed that the optimal percentage of carbon and nitrogen for running the process of the aerobic degradation of organic substances contained in waste is expressed by the C:N quotient with a value of about 25 - 35 and C:P quotient with a value of about 100. Therefore, an assumption may be made that the proper conditions for running the biochemical degradation of organic substances, taking into account the content of organic carbon and total nitrogen, were obtained in samples where the percentage of cellulose pulp amounted to 20 and 30% respectively. By subjecting the content of phosphorus in the samples to analysis, values which differed significantly from the optimal ranges were obtained.

Table 4. Chemical composition of mixtures: cellulose pulp + compost

mixtures	organic matter	TOC	$N_{Tot.}$	C:N	$P_{Tot.}$	C:P
	% d.m.					
cellulose pulp 10% + compost 90%	40.13	15.68	0.79	20	0.03	523
cellulose pulp 20% + compost 80%	46.36	18.76	0.71	26	0.05	375
cellulose pulp 30% + compost 70%	52.59	21.84	0.64	34	0.07	312
cellulose pulp 40% + compost 60%	58.82	24.92	0.56	45	0.09	277
cellulose pulp 60% + compost 40%	71.28	31.08	0.41	76	0.12	259

Table 5 lists the values of the AT_4 parameters obtained for the studied samples and the respiration index calculated for the cellulose pulp after taking into consideration (deducing) the respiration activity of compost. The AT_4 determined in the compost control sample amounted to 14 gO_2/kg d. m.

Table 5 Value of the AT_4 parameter in the studied samples

mixtures	AT_4	The respiration index AT_4 calculated for the cellulose pulp after taking into consideration the respiration activity of compost
	average value [gO_2/kg d.m.]	
compost	14.0	-
cellulose pulp 10% + compost 90%	20.6	64.6
cellulose pulp 20% + compost 80%	22.9	58.5
cellulose pulp 30% + compost 70%	22.2	42.4
cellulose pulp 40% + compost 60%	22.9	36.5
cellulose pulp 60% + compost 40%	23.7	30.2

The value of the AT_4 parameter in the samples changed in a range between 20.6 and 23.7 gO_2/kg d.m. As the degree of the inoculation of the samples with compost decreased, the calculated AT_4 of the cellulose pulp decreased from 64.6 to 30.2 gO_2/kg d.m. after taking into consideration the respiration activity of the compost. The difference between the extreme values amounts to 113%. It may be presumed that the unfavourable share of nitrogen and phosphorus compounds and the increasing content of persistent carbon in the samples (cellulose) influenced the decreasing activity of microorganisms, which resulted in the low intensity of respiration (fig. 1).

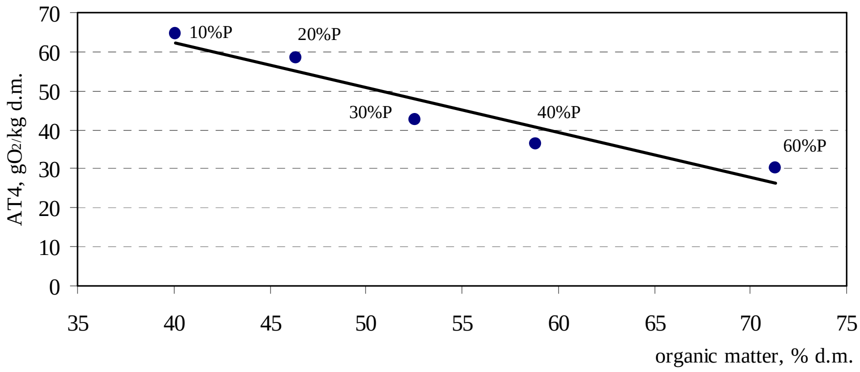

Fig. 1 The relationship of the AT_4 parameter and the percentage of organic matter (cellulose pulp) in samples

4. SUMMARY AND CONCLUSIONS

The quantity of waste produced in Poland is constantly growing, and the main method of its treatment is still landfilling. In order to fulfil the obligations resulting from membership in the European Union, it is necessary to take practical actions to limit the quantity of waste deposited at landfill sites. This requires rational waste management and proper treatment. Waste treatment should have a positive impact on the environment protection and the generated products must be safe for the environment.

During the studies of the respiration activity of the cellulose pulp, the following conclusions have been drawn:

1. The conditions for performing the test, such as the C:N:P quotient and the degree of compost inoculation are of crucial importance for obtaining reliable results;
2. During the conducted studies, the approximately optimal conditions were obtained for tests in the samples with a degree of compost inoculation of 20 and 30% respectively: C:N - from 26:1 to 34:1;
3. In the compost control sample, the determined AT_4 was 14 gO_2/kg d.m., which proves the sufficient activity of microorganisms for the initiation of the mineralization of the organic carbon included in the analysed cellulose pulp;
4. For the cellulose pulp, depending on the degree of the inoculation of the samples with compost, the values of the AT_4 parameter were obtained in a broad range, that is, between 64.6 and 30.2 gO_2/kg d.m.;
5. It may be presumed that those factors which limited the speed of the biochemical processes in the samples with a cellulose pulp content above 30% included an unfavourable share of nitrogen and phosphorus compounds and an insufficient population of microorganisms.

Data obtained from literature indicate that there is no single method which may be used at the same time to monitor the process and determine the stability of the compost sample. Despite the routine performance of respirometric tests, further studies aimed at better understanding of the waste biological activity are necessary.

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OCENA AKTYWNOŚCI BIOLOGICZNEJ PULPY CELULOZOWEJ TESTEM RESPIRACYJNYM AT_4

Streszczenie

W krajach UE prowadzone są obecnie prace nad ujednoliceniem wartości granicznych oraz metod testowych oznaczania aktywności biologicznej odpadów. Celem prowadzenia testów jest monitoring efektywności biologicznego rozkładu odpadów podczas kompostowania, ocena zmniejszenia aktywności biologicznej odpadów przed ich składowaniem, kontrola procesów zachodzących na składowiskach. Ocenę aktywności

biologicznej odpadów można przeprowadzić m.in. poprzez badanie respiracji. Jedną z takich metod jest oznaczenie AT4 (*Static Respiration Index*). Wyniki badań respirometrycznych obrazują dostępność substratów dla mikroorganizmów, czyli podatność na biodegradację.
W artykule opisano badania aktywności biologicznej pulpy celulozowej testem AT_4, wpływ stopnia zaczepienia kompostem na wartość tego parametru oraz zależność od zawartości masy organicznej i OWO w badanym substracie. Pomiarów zapotrzebowania na tlen dokonano przy pomocy systemu pomiarowego OxiTop® Control.

Słowa kluczowe: pulpa celulozowa, aktywność mikrobiologiczna, AT_4, metodyka oznaczania

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