

# BOD

AR\_BOD\_soils+solids.8, earthworms\_lab\_01\_E

## Application report Respiration activity of earthworms



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**Manometric determination  
of the respiration activity  
of earthworms with the  
OxiTop® Control  
measurement system**

1st edition, July 1999

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**Note:** This report was made by using OxiTop® Control. All measuring procedures can easily be transferred to the OxiTop®-IDS system.

## Area of application

Determination of the respiratory activity of earthworms and other soil dwelling organisms in the fields of application of ecotoxicology and animal physiology

## Measurement principle

Manometric measurement of oxygen consumption with simultaneous absorption of CO<sub>2</sub> in caustic soda solution and titrimetric determination of the absorbed amount of carbon dioxide.

## Material

OxiTop® Control measuring heads (WTW, Weilheim, Germany)

Oxitop®OC 110 Controller (WTW, Weilheim, Germany)

ACHAT OC PC Communication Software (WTW, Weilheim, Germany)

Data transmission cable, type AK 540/B for RS 232 (WTW, Weilheim, Germany)

Measuring vessel MG 1.0 with lid-locking device DV/MG (WTW, Weilheim, Germany)

Temperature-controllable room or thermostat cabinet in variants TS606/2 ....TS606-G4/Var (WTW, Weilheim, Germany)

Personal computer, minimum configuration: 80486 processor, 16 MB RAM, RS232 interface

Windows 3.1 or 3.11 operating system

EXCEL® software (Microsoft, USA)

Vaseline

Laboratory scales (reading accuracy: min. 0.1 g)

Measuring container (50 mL)

Volumetric pipette, 50 mL

Measuring cylinder, 50 mL

Caustic soda solution (1 mol/L)

Earthworms

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## Conducting the measurement

Con-sec. no.	Workstep	explanations, comments, notes
1	Put 200 g soil (50% $Wk_{max}$ ) into each of two 1 L measuring vessels.	
2	<p>In one of the two preparations - depending on the question - place 1 to 4 earthworms which have been previously rinsed with tap water, freed of excess water with blotting paper and weighed.</p> <p>The second preparation is used for registering the endogenous respiration without the addition of earthworms.</p>	
3	Place each of the 2 absorption vessels with 50 mL caustic soda solution (1 M) in the holder of each lid.	

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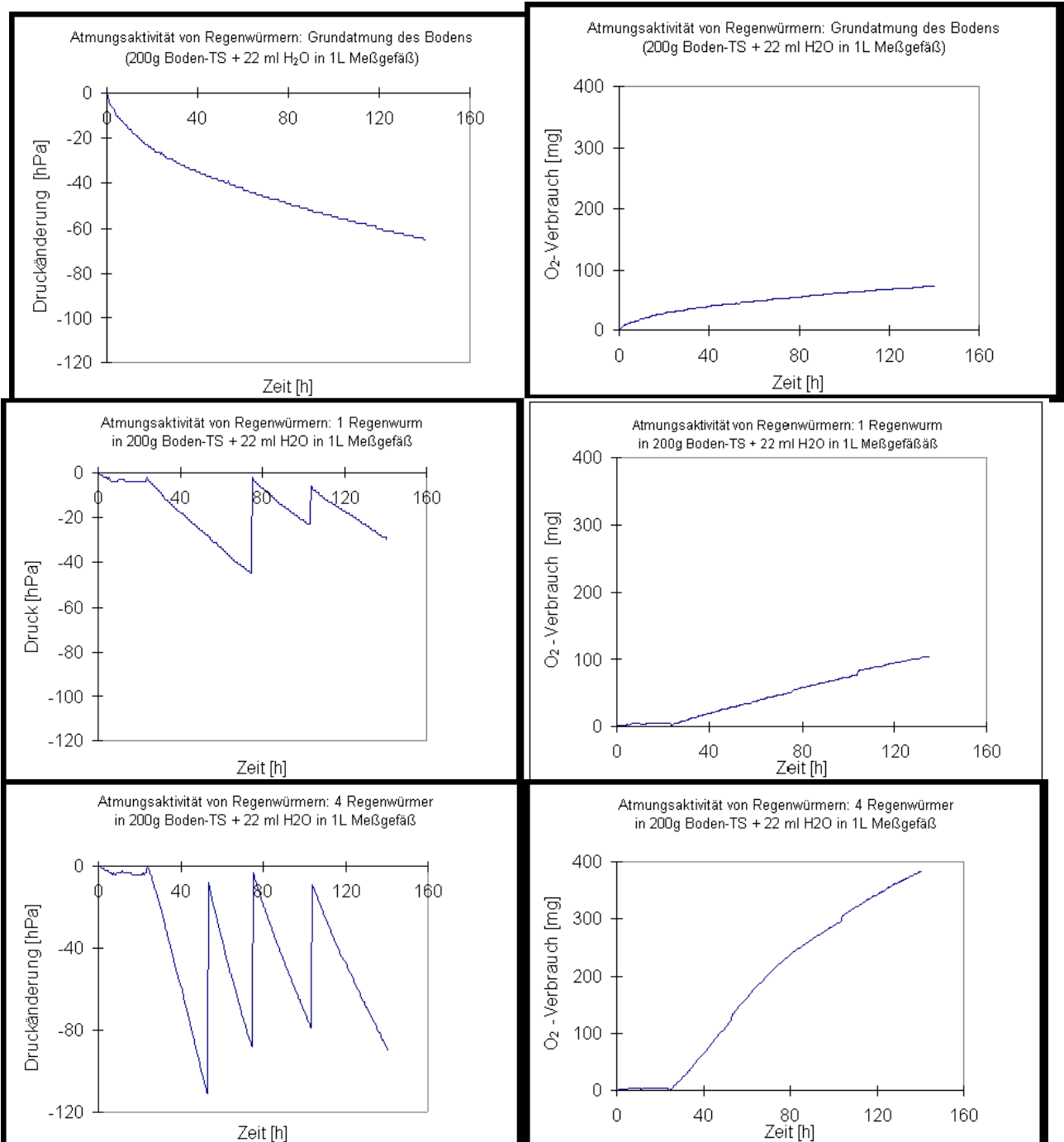
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Con-sec. no.	Workstep	explanations, comments, notes
4	Put on the lid and affix with 4 clamps (4x 90°)	
5	Insert rubber sleeve	Do not grease this joint!
6	Screw on the Oxitop®-C measuring head	Do not use force!
7	Place the test preparation into the constant temperature room or thermostat cabinet	For many applications, particularly in the case of short-time measurements, it is advisable to preheat all materials/components in order to avoid incorrect measurements due to temperature adjustment
8	Start recording the measurements with the OC110 controller (Pressure p mode)	The total measurement time depends on various factors. Measurement times of 7 days are recommended for natural soils; however, a general recommendation cannot be given.  360 measurements are taken at equidistant time intervals throughout the entire measuring period
9	To prevent oxygen-limiting measurement conditions from occurring, the measurement data of the measuring heads are transferred to the controller at regular intervals. If a defined pressure value or the set warning pressure is undercut by -100 hPa, for example, while processing the concerned measuring vessels/samples, by ventilating, addition of nutritive solution, sampling, refilling of the CO <sub>2</sub> absorber etc., for instance, call up the instantaneous value before the start and after the end of the action and save in the controller (max 10 instantaneous values M01... M10)	In the manometric/respirometric determination of the respiratory activity, the partial oxygen pressure in the sealed measuring vessel decreases during the measurement. If a minimum partial oxygen pressure is maintained, the biological activity of the microorganisms is not affected.  The Oxitop® Control measurement system documents the entire pressure development or the oxygen consumption as a graphic function.
10	If the pressure range mentioned above is undercut, the measuring vessel should be opened and the caustic soda solution replaced.	This procedure can be repeated as often as necessary during the specified time period or via 10 storable instantaneous values without having to restart the measuring head.
11	At the end of the measurement the data are read into a PC using the ACHAT OC software and prepared with EXCEL for presentation of the data.	To determine the specific respiratory rates of the creatures, the basal respiration of the soil must be subtracted from the corresponding measurement values.

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## Examples of measurement results



**Figure 1:**

Results of the respiratory measurement of earthworms in soil samples at 20°C. The fresh weight of the earthworm in the individual test was 4.58 g, the total weight of the 4 earthworms in the preparation shown below was 18.65 G. Left: Progression of the measurement recording with the OxiTop measurement system; Right: Oxygen consumption calculated from the measurements. What currently cannot be explained are the delays of the oxygen depletion in the first 30 hours that

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occurred in both measurement preparations (any leakage of the measuring vessels has been excluded). The respiratory rates are listed in Table 1.

**Table 1:**

Respiratory rates of the earthworms at 20°C, determined with the Oxitop® measurement system in 200 g of soil. At two points in time (t = 60 and t = 120 h) the respective respiratory rates were determined graphically by applying the tangents. The respective values of the pure soil were deducted from the numerical values which were obtained with the worms. The value obtained was divided by the appropriate fresh mass of the earthworms.

Measurement preparation	Mass Organisms [g FS]	Time of measurement [h]	Respiratory rate [mg O <sub>2</sub> /d]	Corrected respiratory rate [mg O <sub>2</sub> /d]	Specific respiration activity of the worms [mg O <sub>2</sub> /(d*g FS)]
Soil	-	60	11,4	-	-
Soil	-	120	5,7	-	-
1 worm	4,58	60	25,0	13,6	3,0
1 worm	4,58	120	22,6	16,9	3,7
4 worms	18,65	60	114,0	102,6	5,5
4 worms	18,65	120	49,8	44,1	2,4

The measuring procedure described in this application is currently undergoing further examination with regard to ecotoxicological questions.

## Bibliography

- [1] AR\_BOD\_soils+solids, Platen, H., Wirtz, A. (1998), Analytical applications No.1: Measurement of the respiration activity of soils with the OxiTop®-Contol measurement system: Basic principles and process characteristics, Fachhochschule Gießen-Friedberg, Wiesenstraße 14, 35390 Gießen. 1st edition
- [2] AR\_BOD\_soils+solids, Platen, H., Wirtz, A. (1998), Analytical applications No.2: Measurement of the respiration activity of soils with the OxiTop®-Contol measurement system: Standard test preparation, Fachhochschule Gießen-Friedberg, Wiesenstraße 14, 35390 Gießen. 1st edition

## Note

The information contained in our application reports is only intended as a basic description of how to proceed when using our measurement systems. In isolated instances or if there are special general conditions on the user side, exceptional properties of the respective sample can, however, lead to a change in the execution of the procedure or require supplementary measures and may, in rare cases, lead to a described procedure being unsuitable for the intended application.

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In addition, exceptional properties of the respective sample such as special general conditions can also lead to different measurement results.

The application reports have been prepared with the greatest possible care. Nevertheless, no responsibility can be accepted for the correctness of this information.

The current version of our general terms of business applies.

Any further questions? Please contact our Customer Care Center:

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