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NITRIFICATION INHIBITION - A SOURCE IDENTIFICATION METHOD FOR COMBINED MUNICIPAL AND/OR INDUSTRIAL WASTEWATER TREATMENT PLANTS

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ABSTRACT

Inhibition of nitrification at combined municipal and/or industrial treatment plants can cause serve problems in regard to the future low effluent standards for ammonia nitrogen. As inhibition problems often occur only for limited periods and are caused by different substances it is very difficult to find the sources of these substances within the sewer system. Using a two step strategy based on a respiration test it is possible to locate these sources within a short time and to concentrate then on the abatement of the inhibiting wastewaters. The method of the respiration test was developed and the relationship between dilution and inhibition could be described by a new mathematical inhibition model. The method was applied at an Austrian city (Linz) with more than 50% wastewater load coming from industry during the design period for the extension of the existing plant (800 000 PE).

KEYWORDS

Large wastewater treatment plants; inhibition of nitrification; industrial effluents; source identification.

INTRODUCTION

Worldwide there is a trend towards improved wastewater treatment in order to protect receiving waters including coastal areas and inland seas. The most important indicator for advanced biological treatment efficiency is nitrification, the oxidation of ammonia to nitrate. It is also a prerequisite for biological nitrogen removal by denitrification. During recent years it turned out that different locations, especially big cities with all kinds of industrial wastewaters, are faced with the problem of inhibition of nitrification. In most cases inhibition occurs, not at a constant level, but appears only for several days, weeks or months. Often it is a very difficult task to find the sources of the inhibiting wastewaters. This paper deals with a method to start up the search for these sources by a screenig test. The case study which was the starting point of this investigation is included in order to demonstrate its practical aspect.

BACKGROUND (case study)

In 1991 in Austria a new amendment of the water act resulted in stricter requirements for waste water treatment efficiency. Based on this act, the city of Linz (200,000 inh.) with large industrial wastewater discharges to the sewer system is forced to extend its wastewater treatment plant in regard to nutrient removal.
As industrial wastewater contributes more than 50 % of the organic load of the existing wastewater treatment plant it was decided to run pilot plant investigations.
During these pilot plant investigations complete inhibition of nitrifcation has been observed. In order to find the sources of the inhibiting discharges a strategy and an easy method were developed to determine the sources of wastewaters inhibiting nitrification.

The city of Linz operates a wastewater treatment plant which treats the sewage of the city, several neighbouring small communities and the wastewaters from many small and large industries, e.g. a steel mill and a chemical works. The existing plant consists of two primary settlers (1), four aeration tanks (40000 m^3) (2), and eight secondary clarifiers (3) (fig. 1). The actual loading conditions are shown in table 1.

Table 1 Actual loading conditions

	total concentrations (mg/l)	total load (t/d)	industrial load (t/d)
COD	270	40	20
BOD	120	16	11
NH4-N	50	7	6

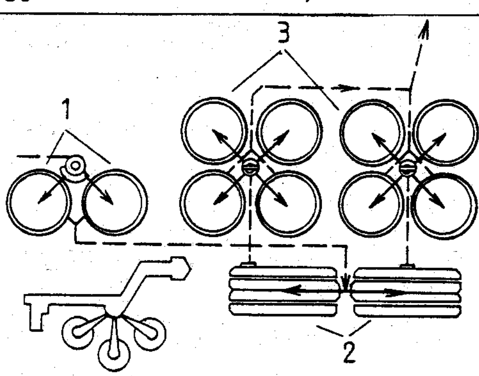

Fig. 1. Layout of the treatment plant of Linz (Austria)

This plant has been designed mainly for the removal of organic carbonaceous matter (BOD_5 loading rate of 0,5 $kg/m^3.d$). For that reason the permit to discharge contains only BOD_5 (20 mg/l) and SS. In the long term average the effluent BOD_5 meets 10 mg/l.
From time to time partial nitrification was detected. During summertime full nitrification could have been expected but never occurred. It was therefore obvious that a certain level of inhibition had to be considered for the extension of the plant. This was the main reason for the pilot plant investigations together with the denitrification problem.

In future the plant will have to meet the effluent standards shown in table 2.

Table 2 Effluent standards for municipal treatment plants in Austria (since 12.4.1991).

efficiency	effluent concentrations		
BOD > 95 %	BOD	15	mg/l
COD > 85 %	COD	75	mg/l
	TOC	25	mg/l
	NH^4-N	3	mg/l *
	NO^3-N	5	mg/l
	tot.P	1	mg/l

4 of 5 samples have to meet the standard, no sample is allowed to exceed 150 % (200 % *) of the standard.

Hence it appears that nitrogen removal becomes the priority of these standards. It is also obvious that efficient nitrogen removal will become a crucial task.
Preliminary short run laboratory tests confirmed that it is possible to nitrify and thus to denitrify too.

Pilot investigations

The pilot plants (2 activated sludge plants with 20 m^3 aeration tank volume each) were started up with half the loading rate (COD) of the existing treatment plant. Sludge retention time (SRT), pH, temperature and alkalinity were kept in an optimal range for nitrification. Within the first two months of the investigation no nitrification could be determined. After having seeded the pilot plant with nitrifying activated sludge (5 m^3) from another municipal wastewater treatment plant without major industrial contributions, nitrification disappeared within the next 3 days. This was a clear indicator that inhibition of the nitrifying microorganisms had occurred. Consequently it was decided to make a systematic investigation to find the source(s) of wastewaters inhibiting nitrification. This investigation covered the whole municipal sewer system as well as all major industrial dischargers.

Strategy of the investigation

The major goal of the strategy was to locate the dangerous discharges to the sewer system within a short time. The first step of the investigation consisted of a screening procedure for the wastewater from 15 sampling points comprising 7 sectors of the whole drainage area and 8 industrial dischargers.
Those samples showing no inhibition can be classified as causing no acute inhibition of nitrification and will therefore not be included in the second step.
Those wastewaters showing inhibition of nitrification will then be subject to step two, which is the determination of the effective concentration of the wastewater. By this test it was intended to find out whether an inhibition by the wastewaters in question can cause inhibition at the municipal treatment plant or not. By this strategy it should be possible to find a priority schedule in the source abatement. Further steps of this strategy are investigations into biodegradability of the inhibiting substances under aerobic and anoxic conditions. These will not be considered in this paper.

METHODS

Sampling

As nearly all small trade effluents are only discharged during the working day, municipal sewage samples were taken automatically from 8 am to 6 pm. The composite samples of the major industrial effluents were taken over their working time, if necessary 24 hours per day.

Determination of the inhibition by respirometry

Respiration rates or oxygen uptake rates were chosen as a method for the measurement of inhibition. In order to simulate biological wastewater treatment, activated sludge was taken from a municipal wastewater treatment plant. It was decided that the sludge used should not be acclimated to inhibiting compounds of most of the industries.
The respiration rate was measured in the apparatus shown in fig. 2. It consists of the reaction tank, a dissolved oxygen probe and a recorder. The reaction tank is closed by a conically shaped plastic stopper. The stopper of the reaction tank is equipped with an oxygen probe, a thermometer and an injector needle. During the measurement the sludge is continuosly mixed by means of a magnetic stirrer. Well aerated activated sludge is poured into the reaction tank which is carefully closed to remove air bubbles from the sludge. The stirrer is switched on and the apparatus is ready for measurement of the respiration rate.
Using activated sludge, it is necessary to determine heterotrophic and autotrophic respiration separately. In order to achieve this it is necessary to selectively inhibit the nitrifying microorganisms. For this purpose ATU is commonly used as inhibitor.

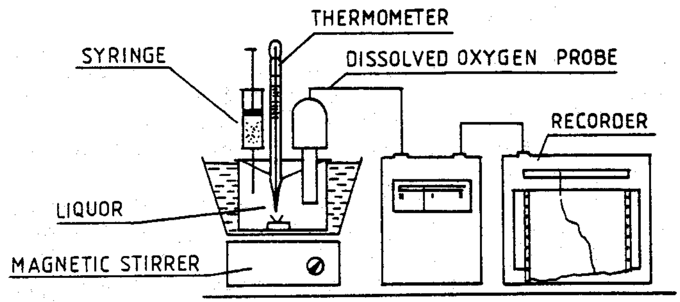

Fig. 2. Laboratory apparatus

Fig. 3 shows the DO-concentration plot of the recorder during a respiration test of nitrifiers before and after having added the wastewater sample to be tested.

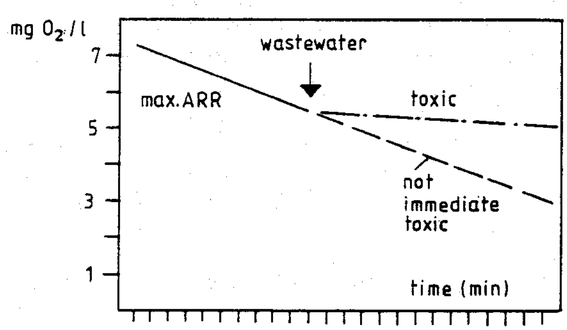

Fig. 3. Respiration rate of nitrifiers

Adding NH_4-N in surplus, nitrifying bacteria reach their maximum autotrophic respiration rate (max.ARR). If no change in the respiration rate is detected after adding the sample, the sample can be classified as causing no acute toxicity. A decrease of the respiration rate indicates an imediate inhibition of nitrifying bacteria.
Based on results from Anthonisen (1976) NH_4-N was kept between 30 and 50 mg/l and the pH between 7.5 and 8.5 to avoid ammonia toxicity. If necessary, NH_4-N was added and buffered with sodium bicarbonate. Nitrifying activated sludge from municipal treatment plants was used for all the experiments.

Screening Test

During the first step of the investigation all the samples were subject to the test of acute inhibition of nitrification.

The following procedure was used.

- Activated sludge from a municipal treatment plant without major industrial influence was used as the microbial basis (adaptation to inhibiting compounds should be avoided).
- The activated sludge was kept aerobic for some hours before it was used for the tests.
- The sludge was mixed with the different wastewater samples one by one on a volumetric basis. In any case the concentrations of the wastewater compounds in the mixture were much higher than they could be in the influent at the treatment plant, where all wastewaters are mixed together.
- After mixing the sample was aerated for 30 minutes.
- A second sample with mixture of tap water and activated sludge was subjected to the same procedure as a reference sample.
- pH was controlled during the whole procedure
- After having increased the NH_4-N concentration to about 30 mg/l the respiration test was made in order to have maximum oxygen uptake rate of the nitrifying bacteria.
- The respiration test had to be repeated with the addition of ATU in order to measure the carbonaceous respiration rate. Then it is subtracted from the value previously measured to get the maximum nitrification oxygen uptake rate.
- If the difference between the two samples (tap water and wastewater) was more than 20 percent and this was confirmed by a second test, the wastewater in question was classified as inhibiting and subject to the second step of this investigation (EC-Test).

Determination of the effective concentration (EC) of the wastewaters

Those wastewaters which caused inhibition during the screening test were subjected to the effective concentration test (EC-Test).
In this investigation the same procedure was used to measure the maximum oxygen uptake for nitrification, but the quantity of wastewater added to the activated sludge was varied in order to obtain a relationship between dilution and inhibitory effect.

Evaluation of the test procedure

All results are based on duplicate measurements. If the difference between mean value and one single value was more than 10 percent, the measurement was repeated.
In order to compensate for small temperature variations during the measurements the following equations were used for correction. For the heterotrophic respiration $f_T = 1.072^{(20-Ti)}$ and for the autotrophic respiration $f_T = 1.103^{(20-Ti)}$.

Theoretical background

It has been found that the Michaelis Menten kinetic theory can be used to describe the nitrification process. If a biological system is inhibited by wastewater constituents, the reaction rate will decrease. Nitrification process inhibition can be caused by different toxic mechanism.
Inhibiting compounds in industrial and municipal wastewaters can act in one of the following ways (Biochemisches Handbuch, 1964). There are different kinetics of inhibition known for the nitrification process

non-competitive inhibition

$$v_i = \frac{v_{max}}{(1+ ks/S)\ (1+ I/ki)} \qquad (1)$$

competitive inhibition

$$v_i = \frac{v_{max}}{(1+ ((ks/S)\ (1+ I/ki))} \qquad (2)$$

substrate inhibition

$$v_i = \frac{v_{max}}{(1+ (ks/S) + (S^m/ki))} \qquad (3)$$

where v_i = velocity of reaction or respiration rate (mg/L/hr)
v_{max} = maximum velocity of the reaction when saturated with substrate (mg/L/hr)
s = substrate concentration
I = inhibitor concentration
ks = half saturation constant
ki = inhibition factor
S^m = order of substrate inhibition

Each of these kinetics can be used for the description of nitrification by pure nitrifying bacteria and a defined inhibiting wastewater compound. Each of these equations (kinetics) is quantified with pure inhibitors and pure microorganism cultures. In the case of wastewater treatment there are always mixed wastewater compounds and mixed microorganism cultures in the activated sludge. This means that in practice none of these equations will be able to describe the kinetics of inhibition adaquately.

RESULTS

Screening Test

3 of 15 wastewater samples were inhibiting. One sample from the municipal sewer system caused a 35 percent inhibition and two industrial wastewaters were completely inhibiting. These were from the steel industry and the chemical works. All the other samples showed no inhibition effects.

Effective Concentrations (Toxic level diagrams)

The percentage of inhibition of biological nitrification can be defined as

$$\% I = (1-v_i/v_{max})\ 100 \qquad (4)$$

where v_i = maximum nitrification rate (autotrophic respiration) in the presence of wastewater sample

and v_{max} = maximum nitrification rate (autotrophic respiration) in the absence of wastewater sample (absence of inhibitors)

Using the non-competitive inhibition model the above equation follows

$$\% I = (1-1/(1+(ks/S)\ (1+I/ki)))\ 100 \qquad (5)$$

if the concentration of substrate "S" is much greater than ks the equation (5) can be simplified

$$\% I = (1-1/(1+I/ki))\ 100 \qquad (6)$$

This model was applied to the data derived from the EC tests. Using a non-linear regression model developed by Marquardt-Levenberg (1963).

It was found that equation (6) does not describe the relation between relative inhibition and dilution precisely enough. The correlation coefficient was between 0.85 and 0.9.
Attempts were made to extend the kinetic model using an additional parameter (kI) in order to improve the correlation coefficient. Mathematically, the relationship for "extended" non-competitive inhibition may be written as

$$\% I = (1-1/(1+(I^{kI})/ki))\ 100 \qquad (7)$$

Up to now it was not possible to find a theoretical exploration for equation (7). But as reality in these tests is very complex with different types of inhibitors and inhibiting processes, equation (7) is of practical importance because it will allow a reduction of the number of dilutions to be investigated.

With this new kinetic the correlation coefficient r^2 could be raised beyond 0.95 for the industrial wastewaters. In the following table 3 the results are shown:

Table 3 Results of the fitted "extended" non-competitive inhibition model

	kI	ki	r^2
steel industry	3.35	3.41	0.967
chemical works	3.14	528.7	0.987
steel+chemical works	2.06	49.7	0.969
Influent wwtpl	2.44	2.68×10^6	0.949
effluent wwtpl (SRT 6 days)	3.48	10.0×10^{10}	0.825
effluent pilot plant (SRT 15 days)	2.60	34.0×10^6	0.926

Table 3 shows that equation (7) fits very well for the industrial wastewaters but much less well for the wastewater treament plant influent of Linz and the biologically treated effluents, showing still strong inhibition.

Figures 4, 5, and 6 show the relationships between dilution and inhibitory wastewaters. It can be seen that the steel industry wastewater is 10 times more toxic than the chemical one. Due to dilution in the mixture of steel and chemical wastewaters, the inhibition effect of the chemical works wastewater dominates.
Fig. 5 demonstrates that different SRT do not have much influence on the reduction of inhibition. Line 5 shows the inhibition effect of the wastewater treatment plant effluent and line 6 the inhibition effect of the pilot plant effluent. Wastewater treatment plant SRT is about 6 days and pilot plant SRT is about 15 days (at 15°C).

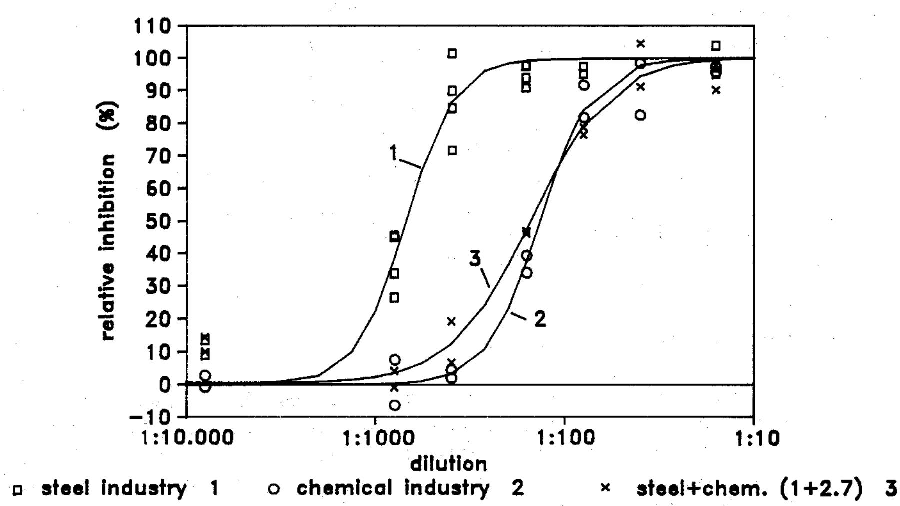

Fig. 4. Inhibition vs dilution

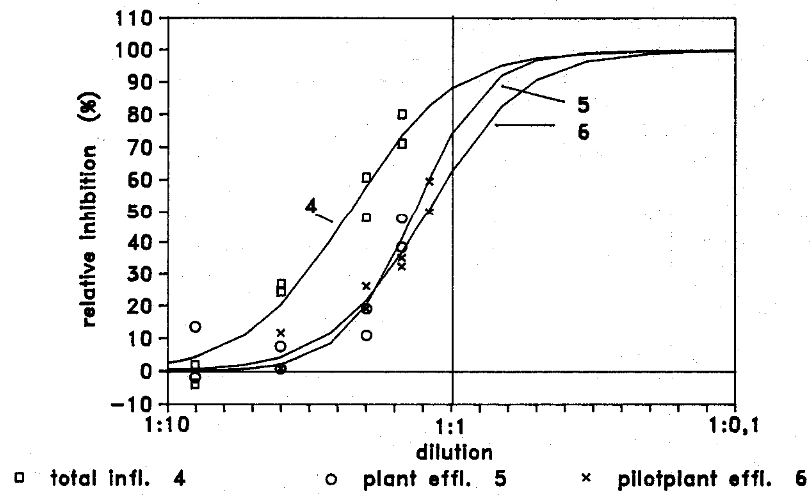

Fig. 5. Inhibition vs dilution

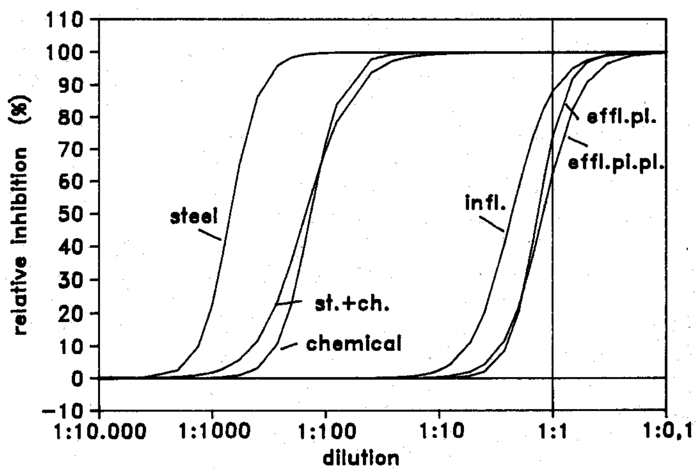

Fig. 6. Inhibition vs dilution

In figure 7 it was tried to demonstrate that for both industrial waste-waters and their combination the actual concentration in the influent to the treatment plant caused an inhibition well over 90 %. If a 10 % inhibition of the nitrifiers could be accepted, the factor of dilution would have to be increased to more than 10 which is absolutely impossible. The only solution of the problem is source abatement or pretreatment.

From the practical point of view any inhibition of nitrification beyond 10 or 15 % should be avoided for reliability. For the design, inhibition of nitrifiers can be compensated for to some extent by increased sludge age (SRT) but the economic constraints and stability of effluent quality allow only low values of inhibition. The actual situation with more than 90 percent inhibition cannot be accepted - it has to be avoided.

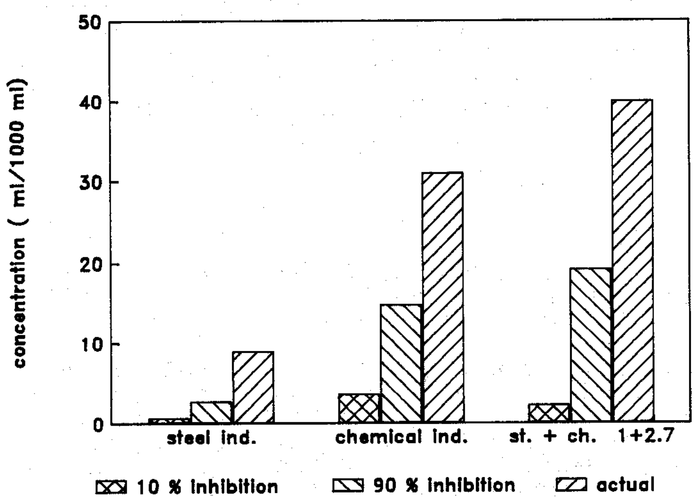

Fig. 7. Actual concentration vs 10% and 90% inhibition concentration

CONCLUSION

The investigation described enabled the decision makers of the city of Linz (Austria) to define the actual number and sources of inhibitory wastewaters and a first estimation of their inhibitory effects. Within one month it was possible to decide how to proceed in order to solve the problem.

It turned out that only 2 of 15 different wastewater sources contain dangerous concentrations of inhibitory substances. Only 2 of 8 industrial effluents were inhibiting. Future activity can concentrate on these two big factories. It was further possible to conclude that a major part of the inhibiting substances will not be biodegradable as, even after biological treatment, the inhibitory effect was preserved to a large extent.

Various mathematical inhibition models from the literature did not fit the results. The introduction of a new parameter into the non-competitive inhibition model improved the description of the relationship between inhibition and dilution (effective concentration).

The investigation described in this paper represents the first steps towards the solution of the problem. Using the respiration tests combined with biological degradation procedures it will be possible to determine whether some of the inhibitory wastewaters contain biodegradable inhibitory substances. It has then to be investigated whether they are biodegradeable under aerobic and anoxic conditions and whether the wastewaters exert only inhibition (reversable) or toxicity (irreversable). Using the methods described it is possible to investigate the different wastewaters in each industrial production process in order to find the important sources. The measurements must be combined with chemical analysis and a literature review, but this is the cheapest and easiest way to determine how to proceed in solving the problem. Inhibition of the carbon degradation will also be detected at the same time. It is obvious that the procedure demonstrated in this paper gives important but limited information e.g. on long-term inhibition by accumulation processes. On the other hand the respiration test can easily be modified for the detection of more detailed information on specific inhibitory effects.

REFERENCES

ANTHONISEN, A.C., et.al. (1976). Inhibition of nitrification by ammonia and nitrous acid. Journal WPCF, 48, No.5, 835-852

Biochemisches Taschenbuch, (1964). 2. Teil, 2. Auflage, Hrsg. RAUEN, Springer Verlag.

GEE, C.S, SUIDAN, M.T., PFEFFER, J.T. (1990). Modeling of nitrification under substrate-inhibiting conditions. Envir. Engin., 116, 18-31.

HOCKENBURY, M.R., GRADY, C.P.L. jr. (1977). Inhibition of nitrification-effects of selected organic compounds. Journal WPCF, 768-777

LEWANDOWSKI, Z., JANTA, K., MAZIERSKI, J., (1984). Inhibition coefficient (ki) determination in activated sludge. Water Res., 19, No. 5, pp 671-674.

MARQUARTD, D. W., (1963). J.Soc.Ind.Appl.Math., 11, 431.

MARTIN, G., RICHARD, Y., (1982). Elements of inhibition of nitrifying Bacteria. Wat.Sci.Tech., 14 (6/7) 781-794

MATSCHE, N., (1990). Toxizität und Abbaubarkeit. Wiener Mitteilungen, 81, 2. Auflage

MIDDELDORF, J., (1990). Methoden zur Messung der Nitrifikationshemmung durch Abwässer. Abwassertechnik, 3, 21-22.

NEUFELD, R.D., HILL, A.J., ADEKOYA D.O., (1980). Phenol and free ammonia inhibition to nitrosomonas activity. Water Research, 14, 1695-1703.

NOWAK, O., SVARDAL, K., (1990). Nitrifikation und Denitrifikation Wiener Mitteilungen, 81, 2. Auflage.

PAGGA, U., (1981). Der Kurzzeitatmungstest - eine einfache Methode zur Bestimmung der Atmungsaktivität von Belebtschlamm. Vom Wasser, 57, 263-275.

PANTEA-KISER, L., WUKASCH, R.F., ALLEMAN, J.E., (1990). The effect of inhibitory compounds on biological nitrification. 44th Purdue Industrial Waste Conference Proceedings, 465-474.

RICHARDSON, M., (1985). Nitrification inhibition in the treatment of sewage. In: Royal Society of Chemistry, Burlington House London W 1V OBN.

STENSEL, H.D., McDOWELL, C.S., RITTER, E.D., (1976). An automated biological nitrification toxicity test. Journal WPCF, 48, No.10, 2343-2350.

WAGNER, R., KAYSER, G., (1990). Laboruntersuchungen zur Hemmung der Nitrifikation durch spezielle Inhaltsstoffe industrieller und gewerblicher Abwässer. gwf, Wasser,Abwasser, 4, 165-177.

WOOD, L.B., HURLEY, B.J., MATTHEWS, P.J. (1981). Some observations on the biochemistry and inhibition of nitrification. Water Research, 15, 543-551.