

# Effect of substrate concentration on biodegradation of 2,4-dichlorophenol using modified rotating biological contactors

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**Abstract** 2,4-Dichlorophenol used in the manufacture of pesticides, germicides, resins, seed disinfectants and antiseptics, if disposed untreated causes greater havoc for land and aquatic environment. In all the earlier works, 2,4-dichlorophenol has been fed along with easily biodegradable substrate, glucose as one of the constituents. A modified 4-stage RBC was used for the biodegradation studies of 2,4-dichlorophenol. The micro organisms attached to the disks were specially acclimatised to the extent that the 2,4-dichlorophenol alone serves as the sole carbon source supporting their metabolic activities. The RBC was operated at 12 rpm. The toxic substrate removal studies were carried out in the hydraulic loading rates ranging from 0.005 m<sup>3</sup>/m<sup>2</sup>/d to 0.035 m<sup>3</sup>/m<sup>2</sup>/d and organic loading rates from 0.35 g/m<sup>2</sup>/d to 6.15 g/m<sup>2</sup>/d. A correlation plot between 2,4-dichlorophenol removal and organic loading rate is presented. A mathematical model is proposed using regression analysis.

degraded herbicides, saw mills, wood waste incinerators and hazardous waste sites. DCP can cause death, respiratory failure, bone marrow atrophy and skin damage in animals and confers a bad taste and odour to food and water. Literature is available for biodegradation of different type of wastes ranging from domestic sewage, to industrial wastes like distillery, tapioca, sugar cane etc; using RBC technology. But very few reports are available for biological degradation of toxic phenolic waste waters using RBC. Huang et al. (1985) studied the RBC treatment of phenol-formaldehyde resin waste waters. Pilot scale RBC's at various influent phenol levels as high as 600 mg/l were used in this study. Results showed that average phenol removal rates were about 99 percent. Tokuz (1991) used RBC for studying the biodegradation of phenolic compounds such as 2-chlorophenol, 2,4-dichlorophenol, 2,4,6-trichlorophenol; pentachlorophenol, 2-nitrophenol, diethyl phthalate and dibutyl phthalate. This study reveals that the phenolic compounds are biodegradable and RBC process is a viable process in treating phenolic waste waters. Radwan et al. (1997) used laboratory scale modified RBC for studying the organic removal of 2,4-dichlorophenol waste waters. The concentration of 2,4-dichlorophenol treated was 200 mg/l and the removal efficiency was reported as 99.2%. In this study, DCP was spiked along with the synthetic sewage containing glucose as one of the constituents.

## List of symbols

A	m <sup>2</sup>	total surface area of the discs
D	m	Submerged disc depth
F		Removal of 2,4-dichlorophenol expressed as a fraction
L	gm <sup>-2</sup> d <sup>-1</sup>	Influent organic loading
Q	m <sup>3</sup> m <sup>-2</sup> d <sup>-1</sup>	hydraulic loading rate
S	mg l <sup>-1</sup>	Influent 2,4-dichlorophenol concentration
W	rpm	rotational speed
F	hr	hydraulic detention time
T	°C	wastewater temperature

## 1

### Introduction

2,4-dichlorophenol (DCP) is used extensively in the manufacture of herbicides, pesticides, resins and wood preservatives. DCP can be found in the environment in

The advantages of RBC is that, easy adaptability for small to medium type installations, simplicity of construction, operation and maintenance, high mean cell residence time thereby achieving high degree of treatment for low reactor volume. Moreover it is trouble free to operate and maintain in tropical climates.

In the present study, a modified RBC is used for studying the biodegradation of 2,4-dichlorophenol for different organic loading rates. A mathematical model based on experimental results using multiple regression analysis has been developed.

## 2

### Materials and methods

The RBC used in this study is a laboratory model. The biofilm attachment area of the perspex support media was enhanced using netlon sheets attached to it. The RBC trough is of fibre reinforced plastic, having a central spindle (made of stainless steel) of 12 mm diameter. The diameter of each disc is 0.30 m having a 40% submergence in the liquid. The reactor has four compartments with eight number of discs in each

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compartment. The total surface area of the discs works out to 4.52 m<sup>2</sup>. The discs were rotated at 12 rpm by belt drive through a gear reduction mechanism coupled to a 0.5 HP electric motor. The schematic diagram is given in Fig. 1.

**2.1 Start up operations**

During start up operation, the RBC was filled with sewage from I.I.T. waste water treatment plant and was operated in a batch mode until a slime layer developed on the discs. After the development of slime layer, the RBC was operated in continuous mode with the synthetic sewage having the constituents indicated in Table 1. The feed solution was prepared using tap water.

The 2,4-dichlorophenol is soluble sparingly in water and has a pKa value of 7.6. Initially a known amount of 2,4-dichlorophenol was dissolved in ethyl alcohol and fed along with the synthetic sewage in the RBC system after the development of biofilm on the discs. The toxic substrate viz. 2,4-dichlorophenol concentration was increased and subsequently the non-toxic and easily degradable substrate, glucose was withdrawn in sequential manner and also the other nutrients. This acclimatisation extends over a period of six months and at this stage the micro organisms attached to the discs were utilising the 2,4-dichlorophenol dissolved in ethyl alcohol as the sole source of carbon for their metabolic activity.

In 2,4-dichlorophenol, two number of chlorine atoms, the electron withdrawing groups are attached in second and fourth position to the phenol structure. Hence the 2,4-dichlorophenol is relatively more acidic than phenol and has a pKa value of 7.6 which facilitates its solubility in sodium bicarbonate solution.

A known quantity of 2,4-dichlorophenol was dissolved in sodium bicarbonate solution until the dissolution is

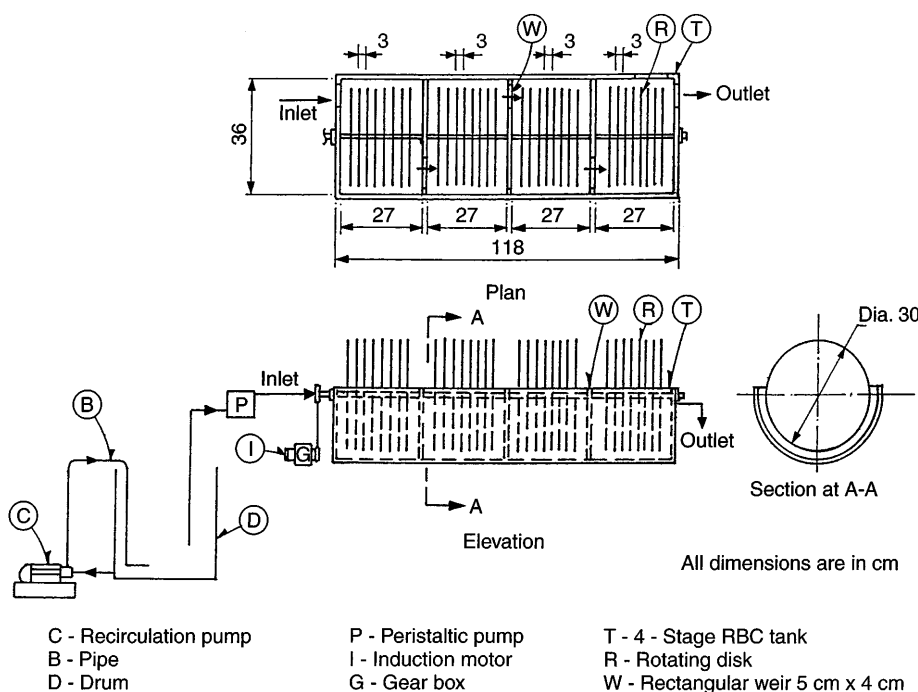
**Table 1.** Composition of Synthetic wastewater

Constituents	Quantity mg/l	Constituents	Quantity mg/l
Glucose	1000	Magnesium Sulphate	100
Di Potassium hydrogen phosphate	1070	Calcium chloride	0.75
Potassium dihydrogen phosphate	527	Ferric chloride	0.5
Urea	227		

complete and made up to the required volume with tap water in the feed tank. By this process, the usage of ethyl alcohol for dissolution of 2,4-dichlorophenol was eliminated. The micro organism attached to the discs of the RBC system utilises only 2,4-dichlorophenol as the carbon source for their metabolic activities. The pH values of the feed were found to be of 7.9 ± 0.3.

4-Amino antipyrine method has been used to determine the concentration of DCP in the waste water. 2,4-dichlorophenol being sparingly soluble in water, has to be necessarily dissolved in an organic solvent. The APHA (1971) suggest the dissolution of DCP in ethyl alcohol for preparation of stock solution. This stock solution was diluted suitably to prepare the calibration curve for estimation of DCP. This has been modified in the following manner for preparing calibration curve.

2,4-dichlorophenol has a pka of 7.6 and is soluble in sodium bicarbonate solution. Accordingly the method suggested by APHA (1971) has been modified by making use of dissolution of DCP in sodium bicarbonate solution.



**Fig. 1.** Schematic diagram of the modified rotating biological contactor unit

### 3 Results and discussion

#### 3.1 Acclimatisation of micro organisms with 2,4-dichlorophenol as the substrate

Micro organisms play a key role in the turnover of carbon in nature. The present system of treatment of 2,4-dichlorophenol is compared with the earlier works of Tokuz (1991) and Radwan (1997). The average influent concentration of 2,4-dichlorophenol used by Tokuz (1991) were in range of 7.17 to 13.87 mg/l with hydraulic loading rate in the range of 0.04 to 0.08 m<sup>3</sup> m<sup>-2</sup> d<sup>-1</sup> and the average effluent in the range from 1.57 to 2.15 mg/l. The toxic element DCP is spiked along with the synthetic sewage having non toxic element glucose as one of its constituents.

Radwan et al. (1997) in their studies used DCP which was fed into the modified RBC system along with the synthetic waste water having glucose as one of its constituents. Since, glucose is an easily biodegradable substrate and the toxic material, DCP, fed along with it would have inhibitory effect on the mixed culture. The micro organisms would be having options of utilising either glucose or DCP as its carbon source for supporting the metabolic activities.

In the present study, the micro organisms are acclimatised to utilise the DCP as the sole source of carbon to support their metabolic activities. This has helped to maintain the RBC system at a higher organic loading and the removal efficiency was almost 100%.

#### 3.2 Effectiveness of staging in RBC system

Most of the 2,4-dichlorophenol into the RBC system has been utilised by the first stage of the RBC.

For a desired effluent concentration at the end discharge, a multistage RBC system is essential. Fig. 2 is a typical representation of DCP concentration of each stage of the RBC system for various influent concentrations

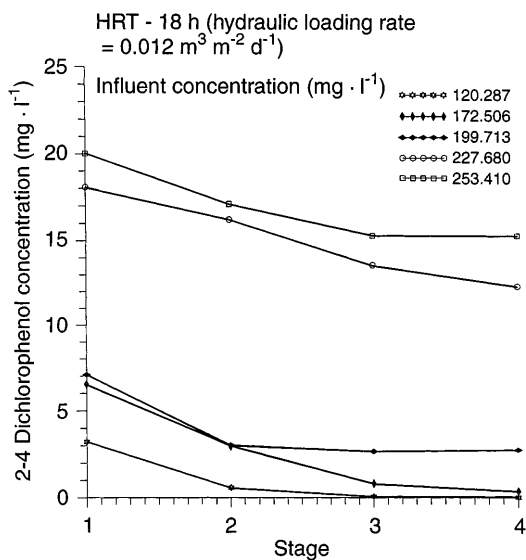


Fig. 2. Effluent concentration at different stages for various influent concentration

ranging from 120.29 mg/l to 235.41 mg/l for a hydraulic loading rate of 0.12 m<sup>3</sup> m<sup>-2</sup> d<sup>-1</sup> (hydraulic retention time of 18 h).

#### 3.3 Effect of organic loading on organic removal

Figure 3 shows the results of organic removal under different organic loading rates. Since the micro organisms have been acclimatised to accept DCP as the sole carbon source for their metabolic activities, the treatment efficiency for organic loading rate of 0.36 g m<sup>-2</sup> d<sup>-1</sup> was found to be 99.5% and for the highest loading of 6.15 g m<sup>-2</sup> d<sup>-1</sup> the treatment efficiency dropped down only to 92.22%.

#### 3.4 Scanning Electron Micrograph studies

Figures 4–9 are the photographic version of the micro organism found in the RBC system at various levels at different combination of the substrates.

Figure 4 shows the growth of mixed culture growing utilising the mixed substrate of glucose, alcohol and DCP in the feed. The photographs indicate the presence of fibrous structure. Compared to this, such filamentous fibrous structures were absent when the glucose concentration was reduced to zero level and the mixed culture was utilising alcohol and DCP as carbon source. This is exhibited in Fig. 5.

Figures 6 to 9 show the organisms acclimatised to DCP and using it as the sole carbon source. The micro organisms were found to resemble the genus *Arthrobacter* which have the tendency to exist in rod as well as in coccoid shape depending upon the substrate concentration.

### 4 Mathematical model based on experimental results

A linear regression model with the parameters, influent organic loading (*L*), influent substrate concentration (*S*), hydraulic flow rate (*Q*), retention time (*θ*), rotation speed (*W*), effective surface area (*A*), submerged disc depth (*D*)

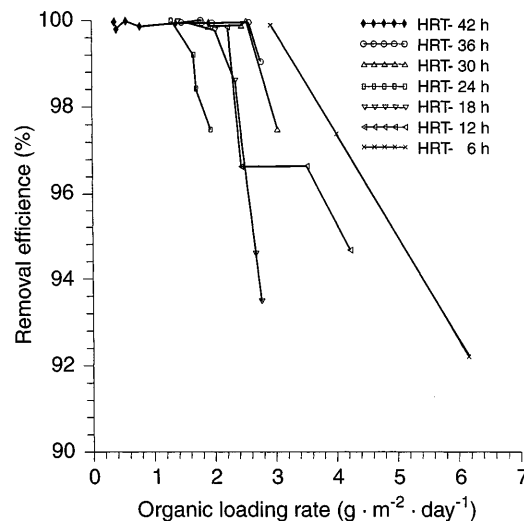


Fig. 3. Removal efficiency vs organic loading of 2,4-dichlorophenol g m<sup>-2</sup> d<sup>-1</sup>

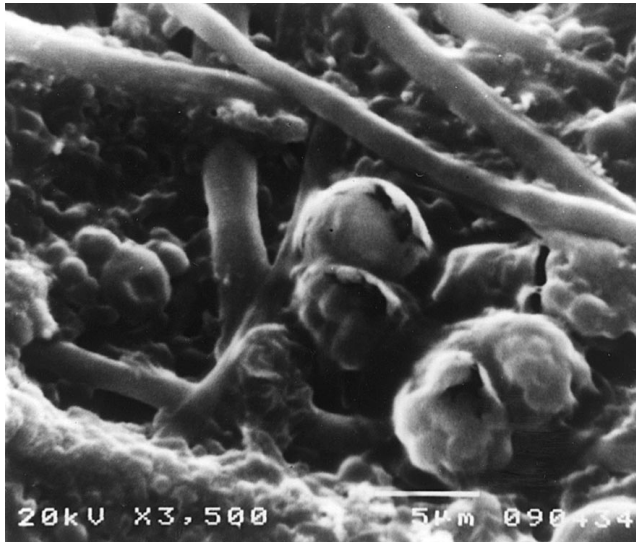


Fig. 4. Mixed culture acclimatised to glucose, alcohol and 2,4-dichlorophenol

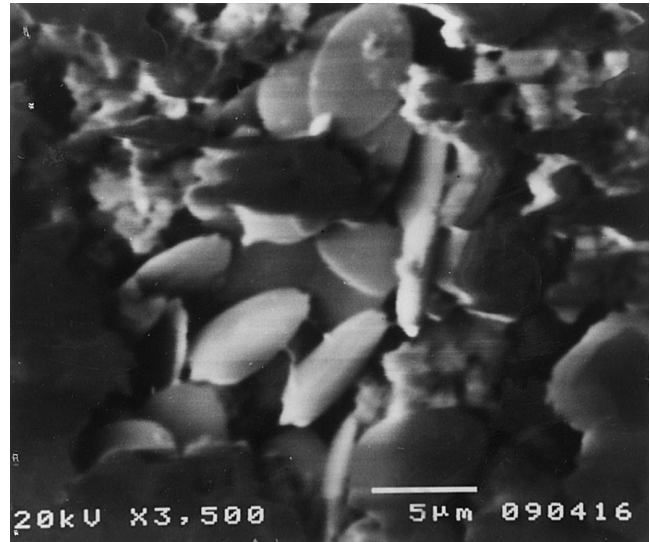


Fig. 6. Mixed culture in stage I RBC acclimatised to 2,4-dichlorophenol

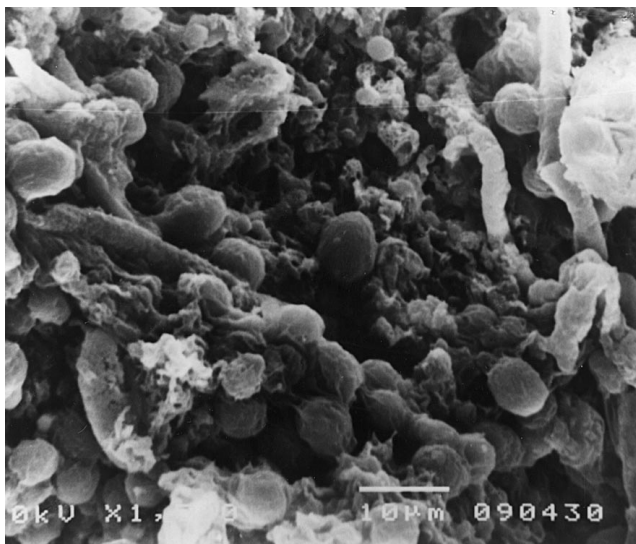


Fig. 5. Mixed culture acclimatised to alcohol and 2,4-dichlorophenol

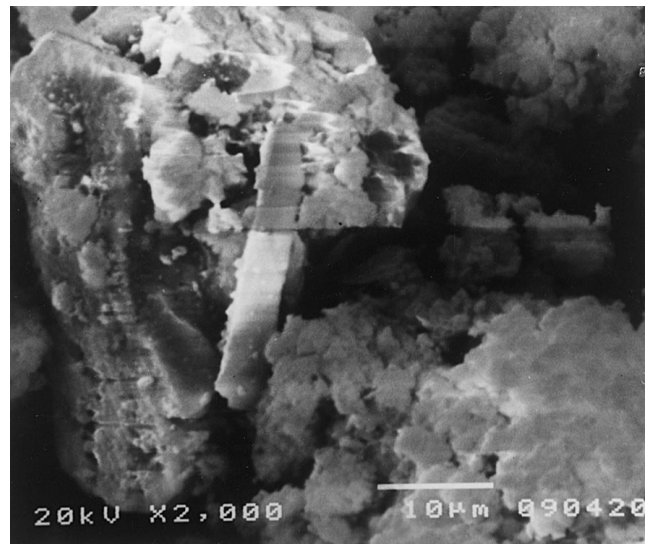


Fig. 7. Mixed culture in stage II RBC acclimatised to 2,4-dichlorophenol

and liquid temperature ( $T$ ) is attempted. The removal efficiency expressed as fraction ( $F$ ) is given by:

$$F = A_0 + A_1 * L + A_2 * S + A_3 * Q + A_4 * \theta + A_5 * W + A_6 * A + A_7 * D + A_6 * T.$$

Wu et al. (1980) modified the equation by omitting  $t$ ,  $D$ ,  $A$  and  $T$  because they do not significantly contribute to the regression. In the present case  $A$  and  $W$  are constant and  $T$  is assumed constant.

We can write:

$$F = A_0 + A_1 * L + A_2 * S + A_3 * Q + A_4 * \theta.$$

The partial regression co-efficients in the equation were determined by conducting the regression analysis of the experimental data and the following correlation was obtained:

$$F = 1.076 - 0.121 L - 0.0003 S - 0.383 Q - 0.0067 \theta,$$

(number of data points = 16).

The observed and predicted data of removal efficiency for 2,4-dichlorophenol waste in RBC is presented in Fig. 10. The deviation between the observed value and predicted value is not more than 1%.

## 5 Conclusions

1. The RBC system successfully treated 2,4-dichlorophenol waste waters. The treatment efficiency was as high as 99.95% at lower organic loading rate of  $0.363 \text{ g m}^{-2} \text{ d}^{-1}$  to 92.22% at higher organic loading rate of  $6.15 \text{ g m}^{-2} \text{ d}^{-1}$ .

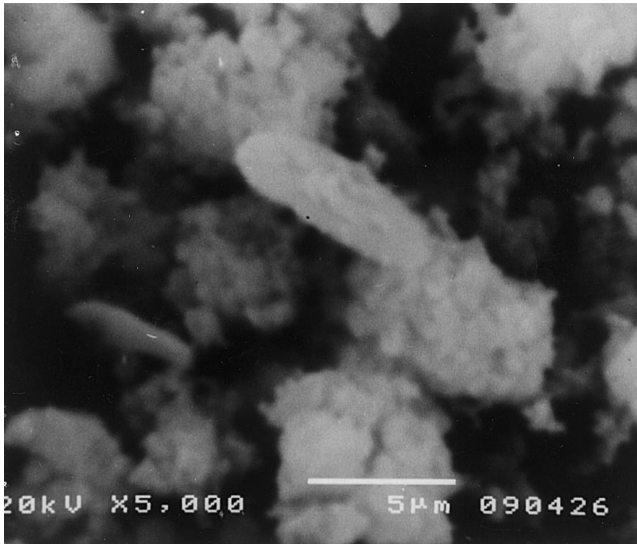


Fig. 8. Mixed culture in stage III RBC acclimatised to 2,4-dichlorophenol

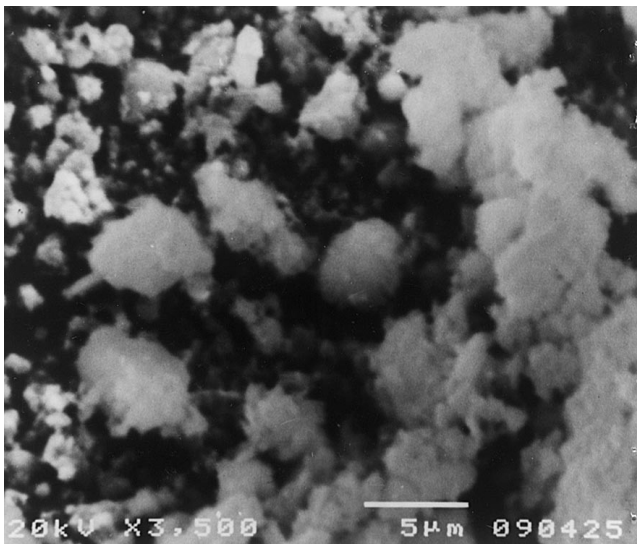


Fig. 9. Mixed culture in Stage IV RBC acclimatised to 2,4-dichlorophenol

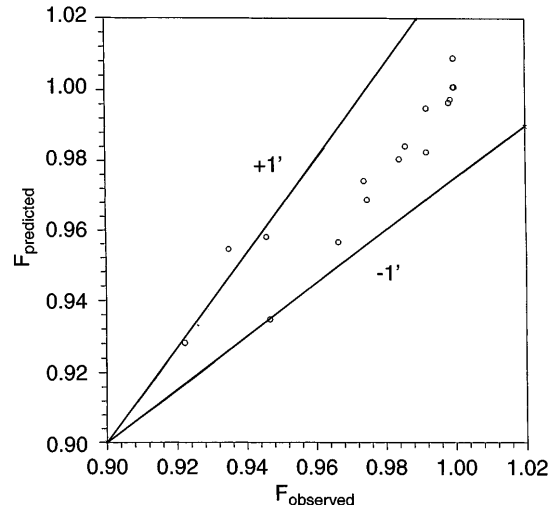


Fig. 10. Observed and predicted data of removal efficiency for 2,4-dichlorophenolic waste in RBC

2. The mixed culture even though takes a longer time for acclimatisation, once acclimatised, accept DCP alone as carbon source to support the metabolic activities.
3. Most of the DCP degradation occurs in the first stage. For achieving a polished effluent of desired effluent concentration for safe discharge into land or aquatic environment, it is desirable to have multistage RBC system.
4. Mathematical model based on experimental data was derived and good agreement has been found.

#### References

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