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Effects of a pulp and paper mill on the ecology of the La Trobe River, Victoria, Australia

J. H. Harris,[1] G. Scarlett[2] & R. J. MacIntyre[3]
[1] *School of Zoology, University of New South Wales (present address: Fisheries Research Institute, NSW, Cronulla, NSW.);* [2] *Australian Paper Manufacturers, Maryvale Mill, Morwell, Victoria;* [3] *School of Zoology, University of New South Wales, Kensington, NSW*

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Abstract

Four riverine animal communities were measured to assess the impact of extensively treated wastewater from a pulp and paper mill on the lower La Trobe River in Victoria, Australia. Benthic macroinvertebrates in channel and bankside habitats were sampled using a new air-lift corer. Population density was expressed in relation to substrate volume. Other communities measured at five sites upstream and downstream of the mill's wastewater outfall were the zooplankton, and the animals associated with submerged littoral vegetation. Ten environmental variables were also measured during the two sampling periods.

A total of 50 benthic macro-invertebrate taxa were dominated by Oligochaeta, Chironomidae and Bivalvia. Benthic communities upstream and downstream of the outfall were very similar. Benthic samples showed large unexplained variation between stations and seasons, despite the similarity of stations and the stratified sampling design, but within-sample variation was small. There was some evidence that benthic faunal patchiness was associated with patterns of stream-bed scouring and deposition in periods of high flow. Littoral samples collected 28 taxa, dominated by Decapoda and Hemiptera. The benthic and littoral communities were quite distinct, with only three species common to both.

Only two of the biological and environmental variables responded to wastewater from the mill: total dissolved solids rose by 20–25% over upstream levels; and zooplankton density increased by 2–3 orders of magnitude. It was concluded that wastewater treatment had successfully avoided the major environmental problems often associated with pulp and paper mills.

Introduction

Many rivers and lakes have been grossly affected by discharges from pulp and paper mills. Biological effects have included the loss of aquatic species richness and diversity in fish and macroinvertebrate communities, fish kills and overgrowth by sewage fungus (Whitney & Spindler, 1959; Hynes, 1963; Scrimgeor, 1989). Mill effluents have caused these effects through the release of organic toxicants, oxygen depletion by reducing agents, sedimentation of wood-pulp fibres, and other changes. Walden (1976) reviewed the toxicity of such effluents, and Waldichuk (1989) also noted the presence of dioxin in invertebrates exposed to effluent from chlorine-bleached paper production.

There have been few reports of the effectiveness of attempts to avoid these environmental problems. This paper describes a study of the aquatic ecological effects of the Maryvale Mill, whose extensively treated wastewaters have been discharged into the La Trobe River in eastern Victoria since 1939.

In contrast to other Australian river systems, the La Trobe River has been studied extensively, and several aspects of its ecology have been reported (Marchant *et al.*, 1984; Metzling *et al.*, 1984; Chessman, 1985, 1986; Marchant *et al.*, 1985; Marchant, 1986; Chessman & Robinson, 1987; Scarlett & Harris, 1990). These studies and other unpublished work in the La Trobe Valley Water Resources Biological Studies by the State Electricity Commission of Victoria (SEC), plus routine monitoring of the quality of the river by the La Trobe Valley Water & Sewerage Board (LVWSB) and staff of the Maryvale Mill, have identified a series of environmental changes affecting the La Trobe River system. These changes include catchment damage due to land clearance and agriculture causing accelerated erosion and sedimentation; impoundment and altered stream flow; sewage discharge plus urban and industrial runoff; thermal pollution and salination associated with coal-fired power stations; and copper pollution possibly from the same source (Robinson, 1983). All of these changes affect the river upstream of Maryvale, so that its quality is substantially altered before it reaches the Maryvale Mill and its fauna in the lowland reaches is disturbed and depauperate (Marchant *et al.*, 1984).

Despite the considerable knowledge of the general ecology and chemistry of the La Trobe River, little biological information is available to demonstrate the effect on the river of the operations of the Maryvale Mill. Hsuan *et al.* (1983) examined physical, chemical and biological aspects of the river at Maryvale and attributed local minor changes to the mill's wastewater.

Because of the usefulness, continuity and sensitivity of biological indicators of aquatic environmental quality (Hynes, 1970; Elliott, 1977; Hellawell, 1978; Campbell, 1982; Easteal *et al.*, 1981), our study assessed the four communities that constitute the aquatic fauna of the La Trobe River near Maryvale. These communities are the benthos (the most commonly used biological indicators) of the mid-channel and bankside habitats (Marchant *et al.*, 1984), the plankton, and the animals associated with submerged vegetation in the littoral zone.

Methods

Study area

The Maryvale Mill, operated by Australian Paper Manufacturers Ltd (APM), generates about 55 Ml of wastewater daily, including most of the aqueous waste discharges from a neutral-sulphite semi-chemical pulp mill, two Kraft pulp mills, four paper machines and a chlorine-based bleach plant. An additional 15 Ml d^{-1} of more concentrated pulping waste is treated elsewhere at a specialised waste-treatment facility. Primary effluent treatment at the mill involves settlement of solids and fibre in clarifiers and is followed by secondary treatment in two mechanically oxygenated aeration lagoons. Tertiary treatment occurs in a 101 ha pond where wastewater residence time is approximately three weeks, and from which there is a continual discharge which contributes up to 20% of low flows in the La Trobe River in summer.

The location of the study area in the lowlands region of the La Trobe River drainage is shown in Fig. 1. Figure 2 shows the study area and sampling stations in relation to the Maryvale Mill and the main components of its wastewater treatment system.

In the Maryvale area the La Trobe is a mature turbid river meandering in an unstable sandy channel 12–30 m wide. Pool and riffle zones are poorly developed and the river's gradient is about 0.5 m km^{-1}. Depth is generally less than 2 m in periods of low flow. Timber debris is common in the stream bed, and provides the only prominent

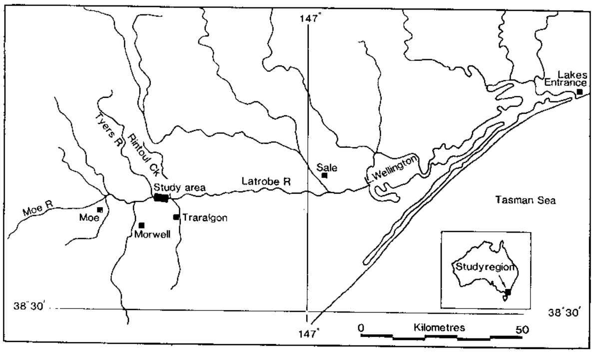

Fig. 1. Location of the study area in the lowlands of the La Trobe River drainage system in Victoria.

habitat complexity. River flow is both highly variable and poorly predictable, although winter floods and summer low-flow periods are common (Hsuan *et al.*, 1983).

Sampling stations

Five sampling stations were selected for faunal analysis (Fig. 2). Stations 1 and 2 were controls, while Stations 3, 4 and 5 were 250 m, 600 m, and 1100 m downstream of the wastewater outfall. Sampling stations were selected for their similarity, considering streamflow velocity, substrate, amount of timber lying in the stream bed, and lack of shading by bank vegetation.

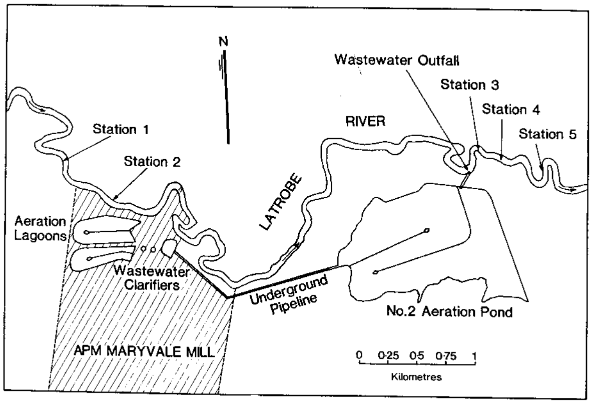

Fig. 2. Map of the study area showing the Maryvale Mill with the main components of the wastewater treatment system, and the location of sampling stations 1–5 on the La Trobe River.

Sampling methods

Measurement of environmental variables

Ten environmental characteristics were measured concurrently with faunal sampling. Temperature, dissolved oxygen (DO), pH, conductivity, suspended solids (SS), total dissolved solids (TDS), total dissolved organic solids (TDOS), total organic solids (TOS), sediment organic matter, and sediment particle size were measured.

Water-quality samples were collected from 0.5 m depth in non-backwater areas. DO, pH and conductivity were measured with suitable meters, while other water-quality measurements followed American Public Health Association procedures (APHA, 1975). The organic content of sediment was estimated in a sample collected at each station with a hand corer at a river depth of about 1 m. The sample was dried to constant mass at 60 °C then four 100 g sub-samples were ashed at 550 °C in a muffle furnace to constant mass. Preliminary digestion with HCl showed only slight loss of mass due to the presence of carbonates.

Sediment particle-size distribution was determined by sieving (Folk, 1974). Five samples were collected with a corer near the bank benthos sampling points at each station. Four samples were also collected from the river channel at each station with a 2 l pot attached to a pole. The sediment was dried at 80 °C for 2 h, then a 0.5–1 kg sub-sample was sieved for 15 min on a mechanical shake through sieves of −1.75 phi (3.35 mm), −1.0 phi (2.00 mm), −0.25 phi (1.18 mm), 0.5 phi (0.17 mm), and 1.25 phi (0.425 mm). Fractions were weighed to the nearest 0.1 g.

Benthos

A stratified random sampling procedure was used to collect replicated quantitative samples of the benthos (Elliott, 1977; Green, 1979). Experimental design was guided by the work of Marchant *et al.* (1984) in the lower La Trobe River. Their findings that the benthos existed as distinct communities in the main channel and near the river bank; that few extra taxa were encountered after examining 10 samples collected by an air-lift sampler with quadrat size of 0.02 m^2; and that this sampler generally gave mean abundance estimates with a precision (ratio of 95% confidence interval to the mean) of 30–50%; were used to fix the habitats to be sampled, quadrat size, and number of replicates. Marchant *et al.* (1984) also found that the abundance of benthos was greatest in autumn, and least in spring. Therefore two five-day sampling visits were made, in May (autumn) and November (spring), 1984.

In each visit ten samples of the benthos were taken in each of two habitat strata – 'channel' and 'bank' – at each sampling station. The bank stratum was defined as being 1–5 m from the water's edge, 0.5–0.7 m deep, and having low-medium flow velocity (<0.2 m s^{-1}). Only sandy sediments of low or moderate slope were sampled. The 'channel' stratum was over 5 m from the bank, 1–1.5 m deep, and had moderate-high flow velocity (approx. 0.2–0.5 m s^{-1}).

A new air-lift corer sampler was built to collect benthos (J. H. Harris & G. Scarlett, unpublished), in an attempt to combine the efficiency and convenience of air-lift samplers with the accuracy and precision of hand-held corers. The sampler head was a cylindrical corer giving a quadrat area = 0.02 m^2. Penetration into the sediment was limited to 200 mm by a flange. Calibration of this air-lift corer showed no significant differences in population-density estimates or species representation of benthic macro-invertebrates compared with a hand-held corer. The air-lift corer was driven by a portable air compressor (225 l m^{-1}) and its pumping efficiency was dependent on depth, but the complete particle-size distribution of the stream bed was collected from deep (1–1.5 m) and shallow (0.5–0.7 m) habitats. Substrate samples were collected in a strainer bag of 175 μm mesh on the air-lift corer, then fixed and stored in 10% (V/V) formalin. In the laboratory the samples were washed, elutriated and sorted. Washing and elutriation were carried out in a large photographic tray (440 mm × 540 mm × 50 mm), the overflow being filtered through a sieve of 200 μm mesh. The sample was covered with a saturated solution of calcium chloride to a mini-

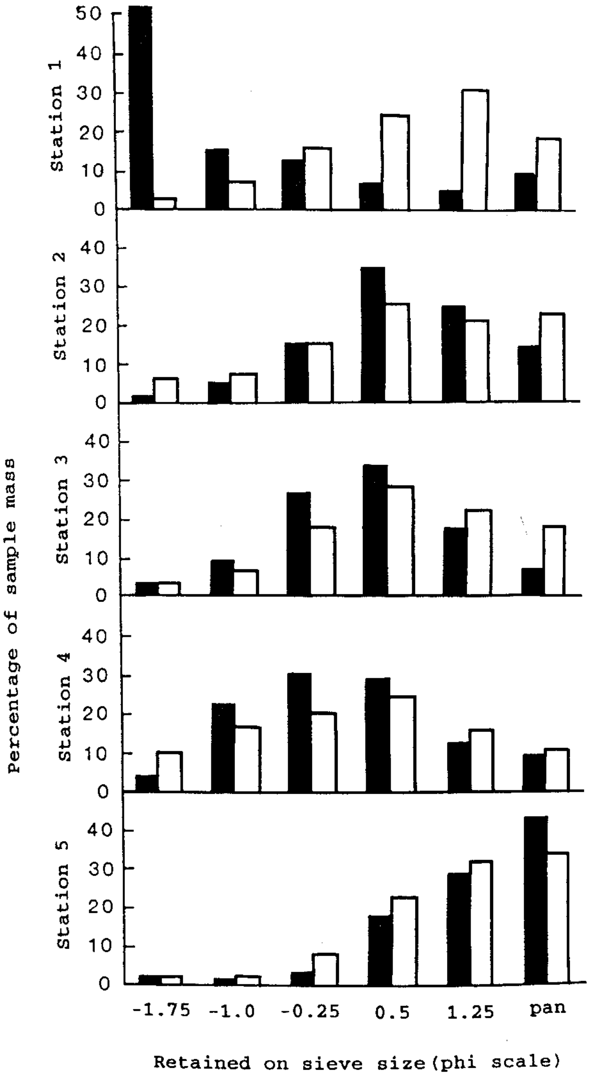

Fig. 3. Sediment particle-size distribution in bank habitats (open bars) and channel habitats (solid bars) at the five sampling stations ($n = 5$).

Table 1. Organic content (mean percent loss of mass on ignition at 550 °C) of sediment collected from La Trobe River sampling stations on 15 May, 1984, ($n = 7$).

Station	1	2	3	4	5
Mean loss	0.72	1.36	0.99	0.78	1.84
Range: from	0.31	0.67	0.42	0.31	0.53
to	1.35	7.80	3.00	2.45	7.12

mum depth of 5 mm. All areas of the tray were then disturbed by hand, the tray was agitated for 10 s, then liquid with suspended animals was poured off through the sieve. Each sample was elutriated three times. The sediment left in the tray was inspected for remaining animals, then sediment volume was measured. Animals were held in 70% ethanol.

Samples were sorted into major taxonomic groups then counted in a plankton counting ring (a 220 mm diameter grooved, rotating acrylic plate) under magnification of 10–40 ×. Specimens from each species were later identified with the aid of a reference collection to provide presence/absence data for each site. In many cases, benthic macro-invertebrates were classified according to the voucher system set up by the National Museum of Victoria (NMV) (Marchant *et al.*, 1984). Staff of the LVWSB also identified some specimens. Further identifications were based on taxonomic keys produced by the museum (unpublished data); CSIRO (1970); Brinkhurst (1971); Mason (1973); Faragher (1980); McDowall (1980); Williams (1980); Nuttall (1982); Cadwallader & Backhouse (1983); and Bayly *et al.* (1967).

Littoral community

Macro-invertebrates and small fish living among the partly submerged grasses near the water's edge were sampled with a two-handled push-net (mesh size 2.5 mm, 0.8 m wide and 1 m deep, weighted with chain). Five replicate littoral samples were collected at each station. Sites to be sampled were defined as having low-medium flow velocity (<0.2 m s^{-1}), a steep bank slope ($>$ 1:4), and a continuous band of submerged grass, commonly 150–300 mm deep, near the edge. Samples were collected by spreading the net under the site then raising it to the surface while agitating the substrate and vegetation with the poles and chain. Animals collected in this 0.8 m length of habitat were fixed and stored in 10% formalin before sorting into species and counting.

Fish were also sampled specifically by i) small baited plastic funnel traps which were set near the bank in 0.5–1 m deep water for 48 h; and ii) wire funnel traps, one of which was set beside the bank at each station for five days.

Plankton

Plankton samples were collected with a plankton net (mouth opening 0.1 m^2, 200 μm mesh size) fitted with a flowmeter. Five 1 min plankton hauls were made from a boat at an estimated speed of 1 m s^{-1}. The catch was fixed with 5% formalin.

Zooplankton and drifting benthic animals were counted in a plankton ring. Large samples were first split in a plankton splitter (<7 splits), so that a minimum of about 200 animals were counted.

Table 2. Water-quality measurements in the La Trobe River and APM wastewater (APM) during biological sampling.

Date	Station	Temp °C	D.O. mgl^{-1}	pH	Cond. mSm^{-1}	S.S. mgl^{-1}	T.D.S. mgl^{-1}	T.D.O.S. mgl^{-1}	T.O.S mgl^{-1}
9/5/84	1	15.7	10.35	7.66	42.7	13.0	270	43	57
9/5/84	2	15.7	10.35	7.66	42.7	23.0	270	55	60
9/5/84	3	15.0	10.2	7.60	56.1	21.5	352	88	75
9/5/84	4	15.0	10.2	7.63	52.6	22.5	328	69	64
9/5/84	5	15.0	10.2	7.63	52.6	25.5	330	67	61
9/5/84	APM	–	6.0	7.4	125.0	41.0	956	128	–
7/11/84	1	17.5	9.2	7.25	21	19.4	144	34	35
7/11/84	2	17.2	9.2	7.35	21	19.0	140	32	27
7/11/84	3	17.0	9.5	7.40	27.5	20.0	175	30	0
7/11/84	4	17.0	9.4	7.37	26.2	19.5	154	33	32
7/11/84	5	17.0	9.3	7.32	26.2	17.5	179	39	36
7/11/84	APM	17.0	7.0	7.90	139.1	20.0	961	95	91

Table 3. Density of animals from the main taxonomic groups in samples of benthic macro-invertebrates (mean and standard error, animals per litre of sediment) ($n = 10$), plus mean benthic population density and coefficients of variation (C.V.) in the two sampling seasons and two habitat strata (Ch = channel habitat, Bk = bank) over five sampling stations.

Station	1				2				3				4				5			
Season	Autumn		Spring		Autumn		Spring		Autumn		Spring		Autumn		Spring		Autumn		Spring	
Habitat	Ch	Bk	Ch	Bk	Ch	Bk	Ch	Bk	Ch	Bk	Ch	Bk	Ch	Bk	Ch	Bk	Ch	Bk	Ch	Bk
Oligochaeta	1.8	7.6	34.6	0.8	2.0	26.1	1.7	10.3	0.7	5.2	11.0	8.0	2.2	25.1	0.3	98.6	0.2	2.0	24.5	1.2
s.e.	0.45	0.03	9.13	0.28	1.06	3.72	0.33	2.66	0.51	2.03	7.76	2.67	0.57	5.79	0.27	17.25	0.11	0.61	6.72	0.61
Chironomidae	1.8	1.0	16.2	0.5	0.8	1.0	1.1	32.1	0.5	1.5	1.1	9.1	1.2	1.7	1.1	221.6	0.1	0.5	4.9	0.5
s.e.	0.75	1.0	4.18	0.27	0.22	0.40	0.38	9.95	0.18	0.59	0.44	1.35	0.61	0.48	0.33	38.72	0.07	0.20	1.06	0.35
Mollusca	6.7	1.2	0.2	0.2	7.4	2.0	0.1	0.1	3.3	1.4	0.6	0.5	7.2	4.7	0.6	0.4	1.1	1.7	2.8	0.1
s.e.	1.58	0.41	0.12	0.16	0.81	0.62	0.06	0.14	0.63	0.58	0.22	0.50	1.20	1.16	0.18	0.27	0.33	0.66	1.1	0.14
Ephemeroptera	0	0.1	0.8	0	0	0.7	0	0.4	0.1	0.6	0	0.5	0	6.5	0	3.3	0	0	0.1	0
s.e.		0.03	0.26			0.31		0.4	0.07	0.38		0.34		1.47		0.92			0.13	
Trichoptera	0.2	0.6	0.6	0.4	0.1	0.2	0	0.7	0.1	0	0	0	0	1.2	0	2.5	0	0	0.2	0
s.e.	0.10	0.46	0.14	0.24	0.09	0.19		1.60	0.07					0.39		0.75			0.15	
Total	10.5	10.5	52.4	1.9	10.2	30.0	2.9	43.6	4.7	8.7	12.7	17.7	10.6	39.2	2.0	326.4	1.5	4.2	32.5	1.8
C.V.	67.9	61.2	63.8	69.8	49.7	43.1	45.7	90.3	49.9	80.5	197.3	60.9	48.0	55.3	82.4	52.1	72.2	77.0	77.2	147.4

Table 4. Three-way analysis of variance for transformed numbers of benthic macro-invertebrates per litre of sediment among stations, seasons and habitats.

Source	DF	SS	Mean S	F	Prob
Stations	4	1.2358	0.3090	8.92	0.0001
Seasons	1	0.2528	0.2528	7.30	0.0075
Habitats	1	0.0694	0.0694	2.00	NS
Station × season	4	0.3252	0.0813	2.35	NS
Station × habitat	4	1.2036	0.3009	8.69	0.0001
Habitat × season	1	0.3452	0.3452	9.97	0.019
Station × season × habitat	4	1.6588	0.4147	11.98	0.0001
Error	180	6.2314	0.0346		
Total	199	11.3222			

Large amounts of fibrous material – mostly of plant origin – made counting difficult in samples that were not split.

Results

Environmental variables

Sediment particle size. The distributions of sediment-particle sizes in bank and channel habitats are shown in Fig. 3. Distribution patterns were similar in bank habitats at each station. They were dominated by medium-coarse sands, with 10–34% of finer sand and silt particles. Sediment particle-size distributions were less uniform among channel habitats. Station 1 had a higher proportion of gravel, while Station 5 had a predominantly medium-fine sand sediment. Stations 2, 3 and 4 were similar to the bank habitats, with coarse sand dominant.

Organic matter. The organic content of sediment, listed in Table 1, was low at all five stations. Values varied, with occasional high readings where concentrations of particles of charcoal, brown coal and other organic matter were collected from small hollows in the stream bed. The mill's wastewater outfall had no apparent effect on the organic content of sediments.

Table 5. The percentage composition, by total number of individuals, of the major benthic invertebrate groups collected in the two habitat strata of the lower La Trobe River (columns headed APM). Results of Marchant *et al.* (1984) from the same area are listed for comparison (columns headed NMV). (Rounding-off of data has removed some rarely occurring groups).

	Channel		Bank	
	APM	NMW	APM	NMV
Oligochaeta	53.6	26.6	44.3	59.5
Crustacea	0	0	0	0.2
Plecoptera	0	0	0	0.2
Ephemeroptera	0.6	31.9	3.5	8.6
Trichoptera	0.6	14.7	1.6	11.0
Coleoptera	0	0.1	0	0
Chironomidae	20.5	18.7	45.9	16.5
Other Diptera	0	0.6	0	0.2
Other Insecta	0	0.1	0	1.9
Hydracarina	0	0.3	0	0
Molluscs	24.7	6.8	4.7	1.7
Others	0	0	0	0.1

Water quality. Table 2 shows water-quality analyses at the five sampling stations and the wastewater outfall. The wastewater varied from control river values in DO, pH, SS, TOS, and TDOS, without creating any marked change in measurements in river stations below the outfall, except an increase (20–25%) in TDS (and therefore conductivity) in downstream stations. This change was previously detailed by Hsuan *et al.* (1983).

Table 6. Benthic macro-invertebrate taxa collected in channel and bank habitats at the five sampling stations in autumn (Aut) and spring (Spr). 'P' indicates presence in samples.

Station	1		2		3		4		5	
Season	Aut	Spr	Aut	Spr	Aut	Spr	Aut	Spr	Aut	Spr
Oligochaeta										
Tubifilidae										
Limnodrilus ?hoffmeisteri Claparede	P	P		P	P	P	P	P	P	
Aulodrilus sp 1			P							
Aulodrilus sp 2			P		P					
Tubificidae sp 3			P				p			
Branchiura sowerbyi Beddard	p				P	P	P			
Chaetogaster sp 1		P				P				
Telmatrodrilus multiprostatus Brinkhurst		P		P						
Naididae										
Pristina longiseta Ehrenberg	P									
Pristina ?aequiseta Bourne	P	P	P	P		P		P		
Pristina nr. proboscoidea Beddard			P				P			
Pristina sp 5			P				P			
Pristina sp 6							P			
Pristina idrensis Sperber		P		P				P		P
Nais elinquis Muller		P		P		P		P		P
Nais communis Piquet		P		P		P		P		P
Nais sp		P						P		
Lumbriculidae										
Lumbriculus ?variegatus (Muller)				P	P	P				P
Haplotaxidae										
Haplotaxidae sp 4			P						P	
Megascolecidae										
?Megascolecidae sp									P	
Insecta										
Chironomidae										
nr. Saetheria (915E)	P	P	P	P	P		P	P	P	P
?Parachironomus sp 3 (LTCS2)			P		P		P		P	
Cricotopus sp		P	P	P	P	P		P		P
?Calopsectra sp 1 (22E)						P	P	P		
Parakiefferiella sp 1 (12BE)	P	P	P	P		P	P	P		P
Cryptochironomus grisiedorsum (13E) Kieffer		P		P		P	P	P	P	
nr. Cordites sp 1 (9E)	P	P		P		P		P		P
Nanocladius sp 1 (93E)	P	P		P				P		
Polypedilum tonnoiri Freeman						P			P	
Rheotanytarsus MMBW sp 1		P	P	P	P			P		
Thienemaniella sp 1 (10E)		P				P		P		P
Reocricotopus sp 1 (2E)								P		
Microspectra sp 1 (50E)		P						P		P
Polypedilum ovesitrophus (Skruse)								P		P
Cladotanytarsus sp 1 (122E)								P		
LTCS 15		P		P		P		P		
Simuliidae larvae		P						P		P

Table 6. (Continued)

Station	1		2		3		4		5	
Season	Aut	Spr	Aut	Spr	Aut	Spr	Aut	Spr	Aut	Spr
Trichoptera										
Asmicridea edwardsi (McLachlan)	P	P	P		P			P		
Cheumatopsyche sp 2	P		P		P		P		P	
Ecnomus sp 2	P	P	P		P		P	P		
Cheumatopsyche sp 1		P								P
Ephemeroptera										
Tasmanocaenis tonnoiri Lestage			P		P		P			
Baetis sp 5		P		P		P		P		P
Atalonella sp 2		P								
Empididae sp 3							P	P		
Odonata										
Hemigomphus sp		P				P				
Plecoptera										
Trinotoperla nivata Kimmins	P									
Leptoperla neboissi McLellan		P		P						
Leptoperla primitiva McLellan		P								
Mollusca										
Corbiculina australis (Deshayes)	P	P	P	P	P	P	P	P	P	P
Chidaria										
Cordylophora sp					P	P				
Total species – Seasons	12	27	18	18	15	18	16	27	8	16
– Stations	39		36		33		43		24	

Benthos

The volume of the substrate sample collected by the air-lift corer (mean = 1.12 1, S.E. = 0.09) was dependent on the hydraulic efficiency of the pumping action, which is determined by water depth (J. H. Harris & G. Scarlett, unpublished). Benthic macro-invertebrates were grouped for analysis into six groups: Oligochaeta, Chironomidae, Bivalvia, Trichoptera, Ephemeroptera, and 'other taxa'. Nematodes were common in samples, but, since they were capable of passing through the 200 μm mesh sieve, they were not included in counts. Fragmented colonies of the cnidarian genus *Cordylophora* were occasionally found, but, since counts could not be made, they were not included in the density data. Fragments of other animals were counted if they included the head. Planktonic microcrustacea – particularly *Daphnia carinata* King – were collected occasionally, but were also excluded from counts. Only 13 macro-invertebrate specimens outside the five main groups were collected in the benthos, and the 'other taxa' classification was not included in calculations. Table 3 summarises the occurrence and population density of the main animal groups. The benthos was dominated by oligochaetes, chironomids and bivalves.

Densities of benthic macro-invertebrates were analysed by three-way analysis of variance with sampling stations, seasons, and habitat strata as main effects. Numbers of macro-invertebrates

Table 7. Taxa caught in littoral samples in autumn (Aut) and spring (Spr), with total numbers of animals.

Station		1		2		3		4		5	
Season		Aut	Spr	Aut	Spr	Aut	Spr	Aut	Spr	Aut	Spr
Crustacea	*Paratya australiensis* Kemp	44	20	31	26	8	67	9	50	99	25
Gastropoda	*Physastra* sp	12		6			2	1	5	3	
Plecoptera	*Leptoperla primitiva* McLellan		1	2		2	5		2	2	
	Dinotoperla serricaudata Kimmins								2		1
	Leptoperla sp				1			1			
Trichoptera	*Anisocentropus* sp 1		1								
	Ecnomus sp 2					1		3		1	
Hemiptera	*Paranisops* sp							1			
	Agraptocorixa sp					8	6	8			
	Micronecta sp		1	1	6		2		1	1	17
	Sigara sp				7	1	10			3	11
	Gerridae sp		1	19	5				1		
	Diaprepocoris sp		1								
	Veliidae sp		3		1		1				
	Hebridae sp				1						
	Mesoveliidae sp						1				
	Anisops sp								1		
Coleoptera	*Notriolus maculatus* (Carter)					1					
	Spercheidae sp		1								1
	Gyrinidae sp						1				
	Dytiscidae sp						1				
Ephemeroptera	*Baetis* sp 5		6		11		7		10		7
Odonata	*Ischnura heterosticta* (Burmeister)	1	1	2		1				1	
	Hemicordulia sp									1	
	Austrogomphus sp		1							1	
Teleostei	*Retropinna semoni* Steindachner	4		3				1			
	Gambusia holbrooki (Girard)	6		2		3		2		9	
	Cyprinus carpio L.				1		1		4		
Total animals		67	37	66	59	25	104	26	76	120	62
Total species		5	11	8	9	8	12	8	9	9	6

were corrected for substrate sample volume to give population density in animals per litre of sediment collected.

Because these population density data were skewed, the counts were transformed by Taylor's exact variance-stabilising function, calculated from the regression of sample variance on the mean (Elliott, 1977), to enable the use of parametric statistics (Green, 1979; Downing, 1979). Each observation (x) is transformed (y) by the formula $y = x^{1-0.5b}$ where b is the slope of the regression line. Since $b = 1.945$ in this case, the function gave a transformed count of each observation:

$$y = x^{0.03}.$$

Analysis of variance showed highly significant differences in population density between sampling stations and seasons (Table 4). No differences were found between the channel and bank habitat strata. The analysis also showed large first and second-order interaction mean squares, but there was no discernible pattern in these interactions. The 'within-sample' variance was low. Table 3 shows the mean density of macro-invertebrates per litre of sediment in the various stations, seasons and habitat strata. The considerable variation between samples did not follow any pattern: high mean density occurred in autumn in the bank habitats at Stations 2 and 4, while in spring, high densities occurred in channel habitats at stations 1 and 5, and again in bank habitats at Stations 2 and 4. Very low mean densities also occurred in an unordered manner. Following analysis of variance, a Student-Newman-Keuls procedure to compare the means showed no significant differences in benthic population density between Stations 1, 2, 3 and 4, but Station 5 had significantly fewer animals than the group of Stations 1–4. Table 5 shows the percentage composition of the major benthic groups compared with the results of Marchant *et al.* (1984).

The occurrence of benthic species at the five stations in each season is shown as presence/absence data in Table 6. Taxonomic difficulties with some of the faunal groups are evident from

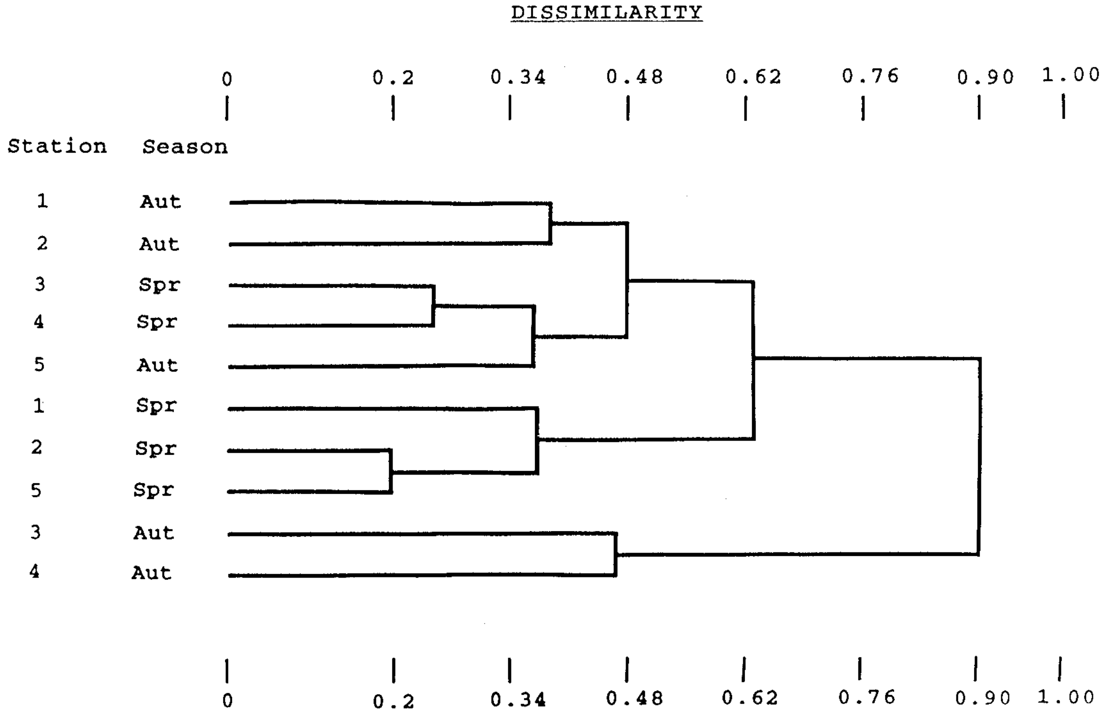

Fig. 4. Dendrogram from classification analysis of littoral samples from the La Trobe River.

Table 8. Species representation in La Trobe River plankton samples in autumn (Aut) and spring (Spr). 'P' indicates presence.

	1		2		3		4		5	
	Aut	Spr	Aut	Spr	Aut	Spr	Aut	Spr	Aut	Spr
Microcrustacea										
Daphnia carinata King	P	P	P	P	P	P	P	P	P	P
Mesocyclops albicans Smith	P	P	P	P	P	P	P	P	P	P
Boeckella triarticulata (Thomson)		P		P		P		P		P
Ostracoda*		P		P	P	P				
Cyclopoid copepod*					P					
Drift animals										
Oligochaeta	P	P	P	P	P	P	P	P	P	P
Ephemeroptera	P	P	P	P	P		P		P	P
Trichoptera	P		P	P	P		P	P	P	P
Chironomidae	P	P		P	P	P	P			P
Paratya australiensis Kemp**		P		P				P		P
Hydracarina*		P			P					
Hemiptera**				P						
Teleostei**				P						

* Single individual
** Larval forms

Tables 6 and 7, where a number of oligochaetes and hemipterans, in particular, are provisionally identified. Similar numbers of species were found at each station, and there were few upstream/downstream differences in the representation of the species. Of the total of 50 species identified, seven occurred at upstream sites but not downstream, while nine species identified from downstream sites were not found upstream of the outfall. Station 4 had most benthic species (43), and Station 5 had least (24). Both of these stations were downstream of the outfall. Species representation was very similar in bank and channel habitats at each station.

Littoral community

The larger animals that constituted the littoral community were more mobile than the benthic species, and were capable of actively avoiding adverse conditions. Twenty-eight animal taxa were found. Table 7 shows that similar numbers of species were recorded from all stations. The fauna was quite distinct from the benthos, and only three species, *Leptoperla primitiva* McLellan, *Ecnomus* sp 2 and *Baetis* sp 5, were recorded from both communities.

Total numbers of littoral animals collected at each station are recorded by major taxonomic groups in Table 7. Catches were dominated by the decapod shrimp (*Paratya australiensis* Kemp) and various Hemiptera. Population density data were again skewed. Because the exponent b in the variance-to-mean regression ($b = 2.14$) showed that the data approximated a log-normal distribution, a logarithmic transformation was used, $y = \text{Log}_{10}x$ (Elliott, 1977). The transformed counts were then analysed by a two-way analysis of variance with replication (Sokal & Rohlf, 1969). There were no differences in the densities of littoral animals between either stations or seasons (sampling stations $F_{(4)} = 1.6851$, $P > 0.10$; seasons $F_{(1)} = 2.2208$, $P > 0.05$; stations × seasons $F_{(4)} = 4.5472$, $P < 0.001$).

Classification analysis of the littoral data using the Bray-Curtis procedure (Belbin, 1989) (Fig. 4) showed three main groups of samples: A) stations 1, 2 and 5 in Autumn plus stations 3 and 4 in spring; B) stations 1, 2 and 5 in spring; and C) stations 3 and 4 in Autumn. The pattern of association showed no relation to the wastewater outfall.

Only those fish caught by push-net sampling were recorded in the analyses. The few fish caught in traps were included in Table 7. Large carp, *Cyprinus carpio* L., were caught in wire funnel traps in the spring sample at Stations 2, 3 and 4. Their stomach contents were dominated by zooplankton. Anglers frequented the area immediately below the outfall pipe. Trapping of small fish was also highly variable. A number of Australian smelt, *Retropinna semoni* Steindachner, and gambusia, *Gambusia holbrooki* (Girard), were caught in the plastic traps in the autumn sample, but none was caught in spring.

Plankton

Two distinct groups of animals were taken in plankton hauls: they were microcrustaceans which dominated the catches (almost exclusively *D. carinata*, *Mesocyclops albicans* Smith, and *Boeckella triarticulata* (Thomson)); and drifting benthic larvae, which were much less abundant that the true zooplankton. The species, and their representation in samples, are shown in Table 8. Those present, and their proportional representation, were similar at all stations. Figure 5 shows a plot of the mean abundance of plankton against sampling station. Plankton density downstream of the wastewater outfall was two orders of magnitude higher than upstream at the control stations. In autumn and spring at Stations 1 and 2 there were mean catches of 2 and 72 animals per cubic metre, while at Stations 3–5, the means rose to 715 in autumn and 3173 in spring. Amounts of fibrous plant material were similar in hauls above and below the outfall. It appeared that much of this material came from sewage discharged upstream, plus decomposing timber in the river bed.

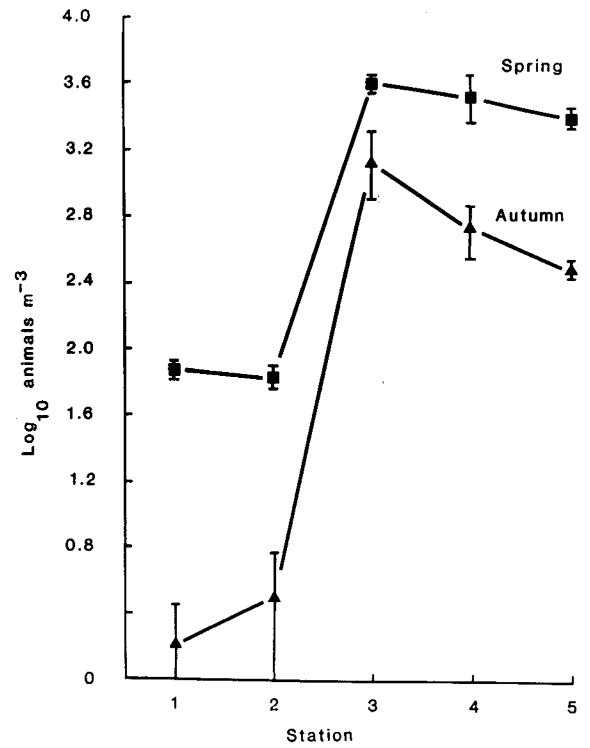

Fig. 5. Mean numbers of animals (± 1 s.d.) per cubic metre of water (log scale) in plankton samples in autumn and spring ($n = 5$).

Discussion

Environmental variables

As Cummins (1962) observed, '... substrate particle size is a common denominator in benthic ecology', and fine-particle organic sedimentation suppresses benthic communities downstream of some North American pulp mills (Blosser, 1984; Hynes, 1970). Our sediment organic analyses showed low but somewhat variable values for all five stations (Table 1), so either the quality of the wastewater discharged from the Maryvale Mill is better than that discharged from some North American mills (at least in settleable organic content), as would be expected after extensive effluent treatment, or the La Trobe River is more able to flush organic material away. The latter seems unlikely.

The dominance of large substrate-particle sizes in channel habitats at Station 1 could not be related to any consistent differences in the benthic community. The greater concentrations of finer particles in both channel and bank habitats at Station 5, however, is probably implicated in the lower benthic population densities and species richness at that site.

The analysis of only two samples of water quality in our study served merely as a check on the results of weekly and fortnightly analyses carried out by Australian Paper Manufacturers' laboratories and those conducted by the La Trobe Valley Water & Sewerage Board. The results support previous studies (Marchant *et al.*, 1984). Wastewater discharge elevated the level of TDS and conductivity by 20–25% (Hsuan *et al.*, 1983), but had little impact on the other physical and chemical characteristics of water quality (Table 2). Although Marchant *et al.* concluded that elevated TDS and conductivity were either the principal form of disturbance, or indicators of some unknown factors affecting benthic communities in the lower La Trobe River, there was no evidence to support this view in our biological samples from above and below the Maryvale Mill outfall. The levels of TDS and conductivity are within commonly recorded ranges for Australian lowland streams (DeDekker & Williams, 1986). Marchant *et al.* also noted that 'Discharges from the APM pulp mill appeared to have little additional impact on water quality'.

Benthos

Estimates of benthic population density showed surprisingly large variation among samples, but this was unrelated to the wastewater outfall. While there was relatively little within-sample variation in the ten sampling units collected from each habitat stratum at each station and season, major differences in density were found between stations and between seasons. These differences occurred despite efforts to minimise environmental variation between sampling areas, which were stratified and chosen for their similarity of locality, stream flow, sediment type and morphology. Thus we conclude that the variability observed in our benthos data does not arise from variation in animal density within particular sampling stations, seasons and strata. Instead, there are real, but unexplained, benthic community aggregations producing density differences between stations and seasons.

The occurrence of species was similar in benthic communities upstream and downstream of the wastewater outfall. Species richness was also very similar, with 41 species recorded from the two upstream sites, and 43 species downstream. Mean benthic population density was no different below the outfall from upstream. Thus, although there were sharp differences between sites and seasons in the density of animals, these differences were not related to the wastewater outfall. Benthic communities upstream and downstream of the outfall were very similar.

The cause of the marked variability in the population density of benthos in the lower La Trobe River was not obvious from our data. There is some evidence that it was associated with the pattern of stream-bed scouring and deposition in times of high flow which were not accounted for in the selection of sampling areas. River discharge was generally low and stable during the four months preceding the autumn sampling, averaging approximately 500 Ml d^{-1}, although a brief peak flow of 3500 Ml d^{-1} occurred three to four weeks before the autumn visit (La Trobe Valley Water & Sewerage Board, unpublished data). By contrast, river discharge was high in the three months preceding the spring sampling. A series of floods passed down the river, four of which reached peak discharges of 5000–15000 Ml d^{-1}. Table 3 shows that benthic population density in the autumn samples varied little between sampling stations and habitat strata relative to the spring samples. Sample means ranged from 1.5–39.2 animals per litre, and the coefficient of variation (CV) among the means of the ten autumn samples was 93.6%. In spring samples collected after repeated flooding, however, variation between stations and strata was much larger: sample means ranged from 1.8–326 animals per litre, and the magnitude of variation was twice as great (CV = 200.5%). The main source of increased variation in spring was a sharp increase in the number of chironomids and oligochaetes in all ten sampling units in the channel stratum at Station 1 and in the bank stratum at Station 4. The other taxa maintained their usual low densities.

Published data show the general impact of stream communities of scouring by floods; for example Allen (1951) recorded marked decreases in animal abundance caused by flooding in the Horokiwi stream. Sagar (1986) found that the abundance of benthic invertebrates in the Rakaia River was inversely related to antecedent discharge. But there are few studies showing the response of benthic communities to local variation in stream-bed scouring intensity associated with hydraulic patterns during floods, although Statzner & Higler (1986), after reviewing studies of faunistic stream zonation, concluded that 'distinct changes in species assemblage are often linked to changes in parameters associated with stream hydraulics'.

Flooding can influence benthic communities directly through increasing the rates of dislodgement and drift of animals. This is due to the shear stress of turbulent flow causing scouring of stream beds (Smith, 1975), especially when sediment particles are small and the substrate uncompacted and unstable, as in the La Trobe River. Robinson (1983) found a correlation between animal density and particle size of mid-channel sediments in the La Trobe River, suggesting that the critical erosion velocity required to mobilise sediments is often exceeded, leading to instability of the substrate inhabited by benthic animals. Flooding can also have an indirect effect by redistributing stream-bed food resources. In the lower La Trobe these resources are mainly associated with dead timber (Hynes, 1970; Chessman, 1986) and algae (Chessman, 1985). Flooding not only has a negative direct effect, as in swiftly flowing turbulent areas where drift rates and population depletion are increased. There can also be a positive local effect because complex hydraulic patterns during floods also produce backwater areas and smaller localised areas of quieter flow. It is reasonable to postulate that drifting animals are able to settle and re-enter the substrate in such quiet areas, thus producing locally aggregated population distributions such as those recorded in our spring samples after flooding. Food resources can also be locally concentrated in the same way, as shown by the variability of organic matter distribution (Table 1). Subsequent periods of low streamflow would permit these locally aggregated animals to disperse into the more uniform population densities seen in autumn samples. In addition to these observations on flow-related benthic community aggregations, which have been supported in continued sampling (Scarlett & Harris, 1990), B. C. Chessman & D. Robinson (pers. comm.) showed that prolonged drought, while it led to a marked deterioration in water quality, caused little alteration in the macroinvertebrate fauna. It appears that physical disturbance can have a more powerful effect on benthic animals living in the unstable sandy substrate of rivers like the lower La Trobe than does water-quality variation within the presently experienced ranges.

Nevertheless, the results of benthos sampling do not suggest that any ecological effect of the Maryvale Mill is being masked by high variability. Population densities, and numbers of individuals in major taxa (Table 3), and also numbers and representation of species (Table 6) were similar upstream and downstream of the Maryvale Mill wastewater outfall. While the few representatives of the sensitive stonefly taxa in the benthos were collected above the wastewater outfall, the littoral samples collected more stoneflies downstream.

The benthos study supported many of the findings of Marchant *et al.* (1984) at their Site 8, which was a few kilometres downstream of the Maryvale study area. The overall mean population density of 31.2 animals per litre of sediment was similar to the estimate recalculated from the data of Marchant *et al.* of 18–76 animals per litre, using their estimate of sampling depth of 50–100 mm. Species representation and relative abundance of major taxa were also similar (Table 5). However, the greater number of samples (12 vs 2) by Marchant *et al.* collected more rare species, and a total of 79 taxa, whereas only 50 benthic taxa were identified in our study. Furthermore, the study of Marchant *et al.* yielded more of the sensitive Ephemeroptera and Trichoptera, while our samples were dominated by the resistant Oligochaeta and Chironomidae. This apparent shift suggests a possible change in the river's ecology during this two-year period, although the few kilometres separating the two study sites prevents direct comparisons. Habitat-quality data provide no insight into the cause of such a change. The difference could be an artefact of sampling, related to a shallower, wider and less-controlled area of substrate sampled by the air-lifter of Marchant *et al.*, compared to the air-lift corer (J. H. Harris & G. Scarlett, unpublished). Greater losses by fragmentation of fragile chironomids and oligochaetes in the earlier study is another possible cause. Subsequent sampling (Scarlett & Harris, 1990) has found local concentrations of Ephemeroptera and Trichoptera in the study area.

Marchant *et al.* (1984) in their extensive survey considered that bank and channel habitats had different faunas. Our study does not support this conclusion in the river reach around Maryvale as the species composition was similar in both habitats and there was no consistent difference in mean population densities between the two habitats.

Littoral community

Measurements of the littoral community did not show any significant differences; both the numbers of species and the total number of animals at each station remained similar between stations and seasons. Multivariate analysis (Fig. 4) showed associations indicating some seasonal grouping of samples, but there was no suggestion of associations related to the location of the wastewater outfall. The littoral data showed no effect from the Maryvale Mill's wastewater.

It was noteworthy that small fishes (*Retropinna semoni* and *Gambusia holbrooki*) were common in push-net samples in autumn, but absent in spring. While reported pulses of copper pollution in the river (Robinson, 1983) could be relevant, this seasonal change reflects the usual patterns of abundance of these fish which are more abundant in summer (McDowall, 1980; Cadwallader & Backhouse, 1983). Frequent catches in both seasons of the gastropod *Physastra* sp., which is highly sensitive to copper ions, support the latter explanation. The failure to catch carp in autumn is difficult to explain. Carp distribution in spring was positively influenced by wastewater zooplankton, on which the fish were feeding; fish were more frequently observed downstream of the mill's outfall.

Environmental variability in the littoral sampling sites was difficult to control, and the habitat depth, slope, and density of vegetation cover all varied considerably. Nevertheless, the 'within-sample' variation in the analysis of variance was acceptably low (MS = 0.3 × subgroups MS) and littoral sampling provided a useful check on the conclusions drawn from the benthic data, especially as it examined a distinctly different and more mobile community. Part of the variation in composition of the two communities could be due to the different methods of sampling. The littoral community might be expected to be a good indicator of noxious influences as it contained, among the 28 taxa collected, such relatively sensitive animals as stoneflies, mayflies, dragonflies and fishes, as well as the more tolerant shrimp and bugs. The lack of discernible effect on this community suggests that such noxious influences were absent from the mill's wastewater.

Plankton

Wastewater from the Maryvale Mill markedly changed the river's plankton community. Downstream of the outfall draining the large shallow lagoon in which aerobic wastewater treatment is completed (No. 2 Aeration Lagoon, Fig. 2), zooplankton density in the river increased by 2–3 orders of magnitude. This biomass substantially increased the food available for riverine fish and other planktivores. Since riverine plankton is usually sparse (Hynes, 1970), and because the species composition of the zooplankton community was the same above and below the outfall, it is assumed that zooplankton above Maryvale are also derived from productive impoundments upstream, such as Lake Narracan. Only *B. triarticulata* did not increase in numbers downstream of Station 3. Plankton samples contained many drifting benthic animals, usually larvae. In shallow stations the disturbance caused by the boat's motor may contribute to these catches, but comparable 'drift' catches were present at each station.

Conclusion

The Maryvale Mill affects conditions in the La Trobe River through an increase in TDS and conductivity of 20–25% over upstream levels. The increase in TDS and conductivity was not associated with any detectable change in the fauna other than an increase in the abundance of zooplankton. The absence of any adverse ecological effect on the four aquatic animal communities that were measured must, however, be seen in the context of the many modifications of the river upstream of Maryvale, which have affected its ecology to the extent that the lowland reaches are significantly disturbed (Marchant *et al.*, 1984). Our study has emphasised the need for a better understanding of the effects on benthic communities of local patterns of hydraulic scouring and deposition during floods, and the need to control such variability to maximise the power of biomonitoring studies.

Whitney & Spindler (1959), Hynes (1963, 1970), Walden (1976), Scrimgeor (1989), and Waldichuk (1989) describe the effects of pulp and paper mills which lacked the tertiary-treatment system used to treat the Maryvale Mill effluent, and which have caused gross ecological changes in rivers and lakes through the release of organic toxicants, oxygen depletion, or sedimentation of wood-pulp fibres. Such changes, and consequent losses of biodiversity and faunal production, have not been found in the La Trobe River below the Maryvale Mill. We conclude that effluent treatment at the Maryvale Mill has successfully avoided the major environmental problems often associated with pulp and paper mills.

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