Sampling Benthos and Substrate Materials, Down to 50 microns in Size, in Shallow Streams

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Stream bed materials, both biotic and abiotic, in the size range 50 μ–ca. 200 mm can be sampled unselectively, in shallow streams, with a simple inexpensive apparatus consisting of a box provided with an adjustable upstream inlet, and, downstream, two nets, one within the other. Collected materials are wet-sieved and the volumes of inorganic material passed by successively finer sieves are plotted as cumulative curves against a logarithmic size scale. Curves are given for materials from three contrasting habitats: a riffle, and pool, of a coastal stream, and an artificial spawning channel. Examples are also given of the densities, size distribution, and vertical stratification of invertebrates from these habitats. Applications of the method to studies on fish biology, invertebrate ecology, and geomorphology are indicated.

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THE materials that lie on the bed of a stream consist of inorganic items, such as clay and cobbles, organic detritus, and living items, such as insects and fish eggs. Irrespective of whether they are biotic or abiotic and irrespective of their densities, many of them can be regarded simply as particles of different sizes. Three groups of workers are concerned with sampling some of these materials unselectively. The fish biologist is interested in sampling eggs and alevins in spawning beds, and in assessment of silt because of its effect on their survival. The invertebrate zoologist samples streams primarily to determine invertebrate density and production, and is interested in particle-size characteristics of the substrate as they affect organisms. The hydraulic engineer and the fluvial geomorphologist measure rugosity of natural channels and use particle-size distributions of bed materials to interpret river mechanics.

Certain basic conditions of obtaining an unbiased sample seem to apply. Firstly, it is frequently impossible to push any apparatus into the stream bed, and if one succeeds, one is disturbing and dispersing material (silts, clays, and organisms) one is trying to collect. Secondly, if anything is placed on the stream bed it must cause no downward displacement of water that would disperse particles; also, it must be streamlined in form so that there is minimum resistance to flow past it. Finally, if a fine net is used to collect silts, the flow into it must be controllable so that a backwash is not generated, with consequent loss of materials.

Some techniques currently used for sampling stream benthos meet these requirements only in part (for reviews or discussions of these see, for example, Macan 1958, Cummins 1962, Southwood 1966, and Ulfstrand 1968). These aim at assessing the numbers of individuals, or biomass, of invertebrates per unit area of substrate and they delimit an area of stream bed within which the materials are disturbed;[1] the fauna is thereby dislodged into the current and so carried into a collecting net. This is the principle of samplers described by Surber (1937), Hess (1941), Waters and Knapp (1961), and Allen (1940). The Hess and Waters and Knapp cylindrical samplers are pushed into the stream bed, which limits their use to substrates with small or medium-size stones (cf. Ulfstrand 1968 p. 38). All these samplers employ a net and so impose a mesh selection on the animals collected. If the net has a mesh of, say, 350 μ or more, most of the invertebrates will pass through it; if a mesh is chosen that will retain the smallest instars of insects, then it clogs rapidly, the current through it is impeded, a backwash builds up, and the fauna is again inadequately sampled (cf. Macan 1958 p. 17; Ulfstrand 1968 p. 38).

[1]Cummins (1962) stresses the need to remove the inorganic materials along with the fauna.

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To reduce these sources of error an inexpensive sampler was designed.[2] It is not worked into the stream bed, the inflowing current can be controlled, and the animals and substrate materials are collected unselectively between 50 μ in least diameter and 200 mm or occasionally more. Within certain limitations (stated below) it therefore largely, but not entirely, meets the requirements of the geomorphologist, fish biologist, and invertebrate zoologist. The last, moreover, can investigate some factors that are major determinants of faunal densities, i.e., the particle size distribution and compactness of the substrate materials (Cummins 1962; Cummins and Lauff 1969), the amount of allochthonous plant detritus that can serve as food (Egglishaw 1964), the algal densities on the superficial stones (see discussion in Ulfstrand 1967), and the velocities and patterns of flow on these as they affect the distribution of insects that extract foods from the current, e.g., Simuliidae, Hydropsychidae.

The Sampler

Essentially the sampler is a floorless box 45 cm high (choice of dimensions is not critical) and 45 cm wide with a narrow vertical upstream inlet, and downstream, two collecting nets, one within the other (Fig. 1, 2*a*, *b*). The area of substrate enclosed is 0.18 m². The box is made of 12-mm (0.5-inch) water-proofed plywood; the converging portions of the sides are secured to the parallel sides by duralumin strips held by screws (*c*); rigidity is obtained by wooden bars across the top (*b* 2) and by a single bar, oval in section, between the converging sides (*a* 1; *b* 1). A length of curved aluminum sheet is fitted to the inlet, and is held in position by a piece of elastic tubing (Fig. 1, 2*e*). The box has a half back of plywood (*a* 2). Angle duralumin (*f*) is screwed to the sides at the back and also extends across the base; this receives the canvas-covered net frames, which are of brass wire, with the inner fitting snugly inside the outer. The canvas of the nets is about 15 cm wide (*a* 3, stippled). The inner net is a bag 55 cm long, made of three pieces of "Nitex"[3] cloth (a top, a bottom, and a continuous piece joining these) of mesh size 600 μ (30 meshes per liner inch); the outer net is made of four pieces (i.e., it is pyramidal) of 50 μ "Nitex." It is 100 cm long and tapers to a canvas sleeve. The seams of the net are strengthened by nylon tape. A canvas sleeve at the apex accepts a plastic collar secured by a hose clamp. The collar is threaded internally and a plastic collecting bucket is screwed into it (*d*). This has two large holes in its walls; these are covered with 50 μ "Nitex."

The sampler is placed on the stream bed *with the nets in place* so that the stream-lined shape

[2]A preliminary account was presented as a summary note in the Proceedings of the 50th annual conference of the Western Association of State Game and Fish Commissioners and the Western Division of the American Fisheries Society.

[3]"Nitex": trademark of Tobler, Ernst and Traber, Inc., New York, N.Y.

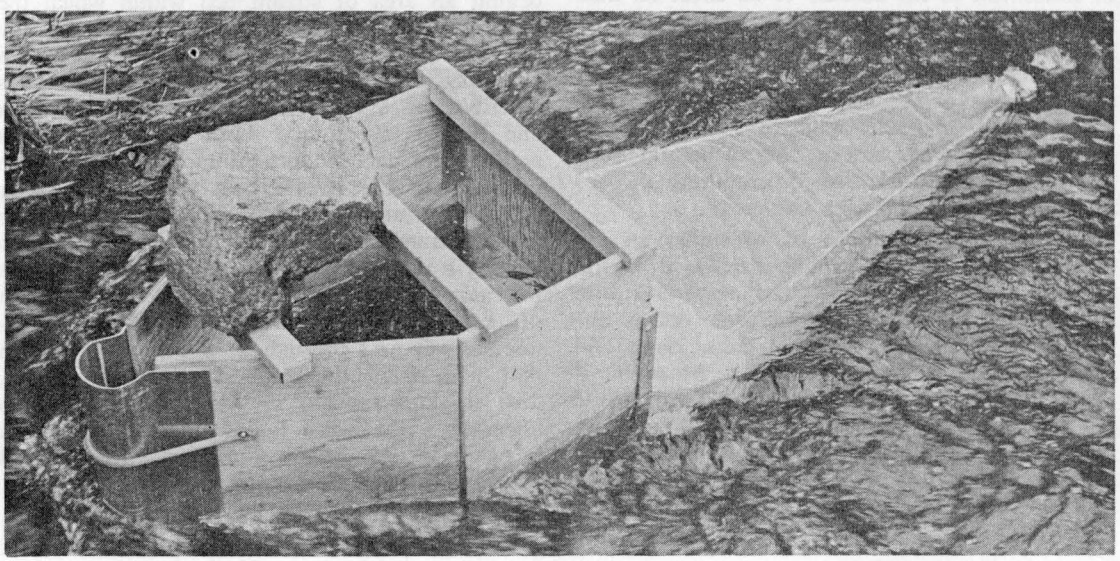

FIG. 1. Stream sampler in use.

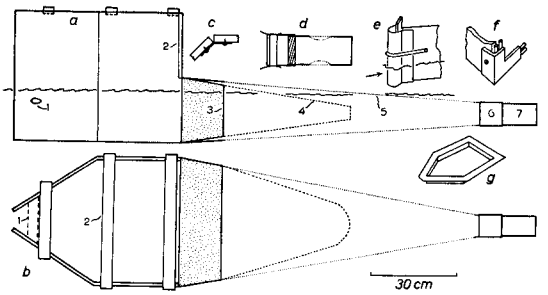

FIG. 2. Constructional details of sampler. *a*, side view; *b*, plan (the scale applies to *a* and *b* only); *c*, method of securing wooden panels together; *d*, collecting bucket; *e*, three-dimensional diagram of front of sampler in use; *f*, three-dimensional diagram of rear corner showing method of securing the net frames; *g*, three-dimensional diagram of frame for substrate materials.

of the total apparatus causes a minimum of turbulence. The forward end is lowered into the water first. Localities with one or more conspicuously obtruding large stones (>20 cm) are avoided. A shoulder of gravel is then built up around the outside if the site is a riffle, but may not be required if the site is a pool with sandy substrate. Stones are picked up downstream of the sampler, and the shoulder serves to reduce or occlude the flow of water under the side into the sampling area. This appears to make a more effective seal than a continuous strip of foam rubber around the base (Warren et al. 1964 p. 645). Loss of material below the crossbar (Fig. 2*f*) is unlikely unless the net is clogging (see below), but can be prevented by application of gravel behind the bar from outside, or of pieces of foam rubber (15 × 5 × 5 cm) below the bar from inside.

The current entering the sampler by the vertical inlet is controlled by raising or lowering the aluminum inlet cover until an adequate flow is obtained. With the flow restricted there is little turbulence within the sampler, so that the substrate becomes clearly visible through the water. When the sample is being taken, therefore, and stones are being removed or disturbed, it is possible to observe the silt particles being carried into the net and to note whether any are passing below the crossbar. Any material entering the box from beneath the sides can also be seen. (An alternative inlet cover of two short lengths of plate separated by a gap caused the water to enter some 15 cm above the substrate; no advantage resulted.)

If one person is operating the sampler, it may be necessary to anchor it with a large stone placed on the forward crossbars (Fig. 1). When a sample is taken, the cobbles, gravel, and coarse sand within the box are lifted out by hand or with a trowel. Cobbles are brushed and washed so that adhering invertebrates are carried into the net. The large inorganic materials are either transferred to a container and retained for physical analysis, or they are discarded. The remaining gravel, sand, and silts are conveyed into the net with the trowel rather than stirred up. The object is to transfer the substrate materials to the nets. In situations such as artificial spawning channels, sands may be most abundant near the surface of the bed and disturbance might allow them to settle further into the gravel. This would not occur with the lighter silts that come into suspension and are carried into the net.

The sampler works best in water less than 30 cm deep, but is usable in slightly deeper water. A higher sampler would be feasible, but the need to be able to reach to the bottom imposes a limit.

The depth of substrate from which materials can be removed varies with the substrate. A common workable depth in stream riffles is 20–30 cm, but in pools, where the substrate is mainly sand, 15 cm may be the limit. The tendency for material to slump into the sampling area increases as the depth increases. Materials are removed to leave, as far as possible, a hole with vertical sides of regular depth. In practice the hole is likely to be irregular, and judgment must be used in deciding which stones near the sides should be removed and which left. The irregularity is a source of imprecision.

The coarse inner net retains the larger animals and materials that have been carried into it, and protects the fine net that retains items down to 50 μ in least diameter. This net must be protected from abrasion from stones.

If it is of interest to record the vertical stratification of materials or invertebrates, the nets are replaced by new ones as the sampling depth increases. During the exchange of nets the opening into the sampler is occluded by the plate being pushed into contact with the substrate. Complete occlusion may not be possible.

If the movement of suspended particles suggests that the fine net is clogging and some backwash is taking place, new nets must be used. Flow through the nets is facilitated if the outside of the fine net is wiped to dislodge adhering particles (this must cause some loss of particles 50 μ in size), and if stones obtruding from the stream bed below the net are removed so that the collecting bucket is not elevated.

An additional application of the sampler is its use in conjunction with an angle-iron frame that

delimits a colonizing area containing either a natural substrate or one of particles of a selected size (e.g., sand or gravel). Clean materials, surrounded by the frame (Fig. 2*g*, 3), are left in the stream for 1 week or more (Ulfstrand 1968 p. 100) so that they can be colonized by fauna. The frame has a shape corresponding to the base of the sampler, which can be placed on top of it and the invertebrates can be collected.

The cost of materials, including a spare fine net, and construction of the sampler is about $80.

Physical Analysis

Methods

The smallest particles collected (50 μ) are in the middle of the coarse-silt category on the Wentworth scale (e.g., King 1966) or the scale of the American Geophysical Union (e.g., Einstein 1964). Medium silts, fine silts, and all clays are lost. This is a limitation of the method.

The absence of very fine particles permits a uniform treatment of the inorganic materials during mechanical analysis, i.e., particles of different sizes can be separated by the same means. After manual removal from the sample, items over, say, 32 mm in minimum diameter are placed in geometrically increasing size categories and their volumes determined by displacement.

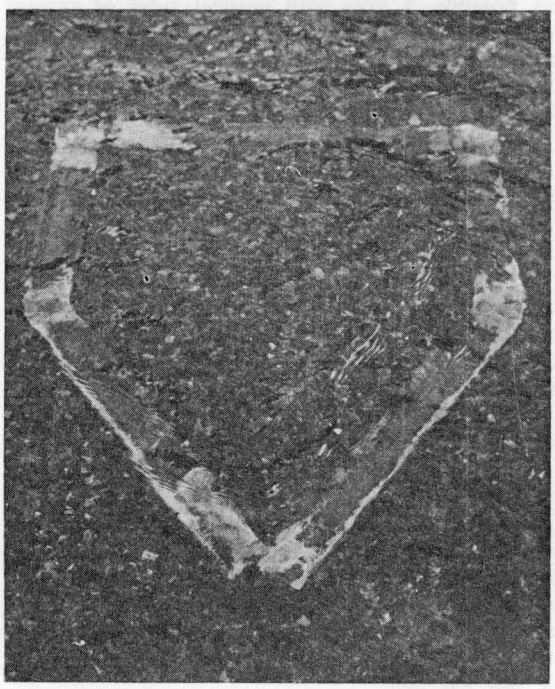

FIG. 3. Frame surrounding substrate materials in stream.

The remainder of the sample is wet-sieved, and the amounts retained on each of a series of sieves can be established as a weight or a volume. Although the measurement of volumes is less accurate than that of oven dry weights, volumes are likely to have more ecological significance and are independent of specific gravities (Morgans 1956; Cummins 1962). Volumes were therefore measured after the sieve residues had settled in water in standard cylinders for a standard time (12 hr).

Collection of a sample in the field takes about 30 min; its physical analysis (apart from 12 hr settling time) takes about 8 hr.

Results

The results are best presented in the form of cumulative curves related to a logarithmic size scale, i.e., the cumulative curve shows the percentage, by volume, of the sample consisting of particles smaller than a given size. A useful logarithmic scale is provided by phi units where phi (φ) is $-\log_2$ of the diameter in millimeters. Thus the arithmetically unequal intervals of the Wentworth scale are converted into equal intervals on the φ scale and the cumulative curve of an analysis can be plotted on equal-interval graph paper along the abscissa without changing the shape of the curve (for discussions on methods of presenting results of analysis see Inman 1952; Morgans 1956; King 1966). Here, particles of smallest size are placed to the left. Both equal-interval scales and arithmetic-probability scales are used to illustrate the examples. King (1966) discusses the advantages of the probability scale and the values used in describing a particle-size distribution.

The most useful descriptive measure of the curves, for the biologist, may be considered. One measure of the central tendency is the median diameter, or the 50% value on the cumulative curve. This can be misleading as it takes no account of the distribution of grain size on each side of the 50% value. Another, the mean, is defined by Inman (1952) as $M\varphi = 1/2\ (\varphi 16 + \varphi 84)$, but this also is unsatisfactory for polymodal distributions (see King 1966). It seems simplest and most instructive to establish from the curves those data likely to be of ecological significance. For example, the percentage by volume of the material of size 1 mm and less can be stated. This is an arbitrary but convenient measure of the fines of the fish biologist. The percentage of material of size 2–6 cm may also be stated, as it is an arbitrary size range of much good spawning gravel. These two measures are used in the examples. The percentage of other size categories can, of course, be read from the curves.

Example 1

Figure 4 shows the curve obtained from the volumetric analysis of inorganic materials taken with the sampler from a riffle of a small creek. The water depth at the site was 15 cm and the surface velocity 60 cm/sec. The substrate appeared to be suitable for spawning salmon. The sample extended 32 cm into it. Material of 1 mm or less amounted to 7% of the total sample, and material in the size range 2–6 cm amounted to 34%.

Example 2

Figure 5 shows, in contrast to the previous sample, the curve of inert materials taken from a pool (depth 21 cm, surface velocity 15 cm/sec) a short distance downstream of the riffle of example 1. The sample reached a depth of 25 cm. Materials of 1 mm or less made up 41%; materials in the size range 2–6 cm made up 15%.

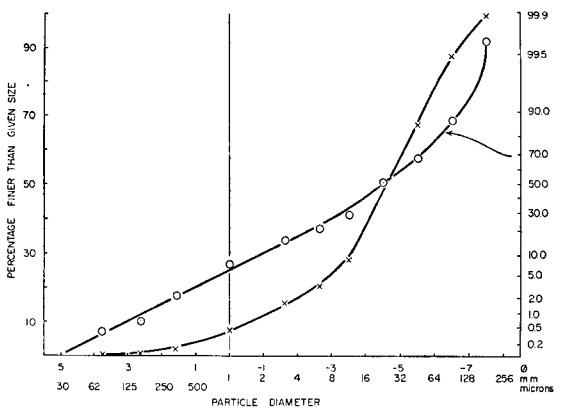

FIG. 4. Cumulative frequency curves of volumes of inorganic materials from a riffle; size scales are in phi units, millimeters, and microns.

Example 3

Figure 6 shows the curve for inert materials in an artificial spawning channel. The water depth of the sampling site was 35 cm, the velocity 75 cm/sec, and the sample reached a depth of 25 cm. In the sample examined materials 1 mm or less amounted to 2% of the total volume; materials of 2–6 cm amounted to 60%.

Biological Analysis

METHODS

The objectives of faunal analysis are to estimate the numbers of invertebrates in the samples, to measure their size distribution, and to establish taxonomic categories and percentage composition.

When a sample is collected the larger stones are picked off the stream bed and brushed, within the sampler. The coarse net in the apparatus is used only to protect the fine net. Its contents are added, in due course, to those of the fine net. The benthic materials are transferred to wide-necked polyethylene jars and formalin is added. If vertical stratification is to be examined, the nets are replaced after the top 10 cm of materials have been sampled. If only the fauna is of interest the cobbles and gravel are discarded. Where moss and filamentous algae are present they would justify being kept separate from inorganic materials and being examined for their associated fauna.

The sieving procedure for physical analysis (carried out in the laboratory) is also the basis for extracting and counting the animals. When the sample is washed on the 1-mm mesh sieve (16 meshes/inch) the residue is examined, firstly with the unaided eye, and secondly with a 12× stereomicroscope, and the fauna is picked out. All fragments of leaves, twigs, insect cases, and

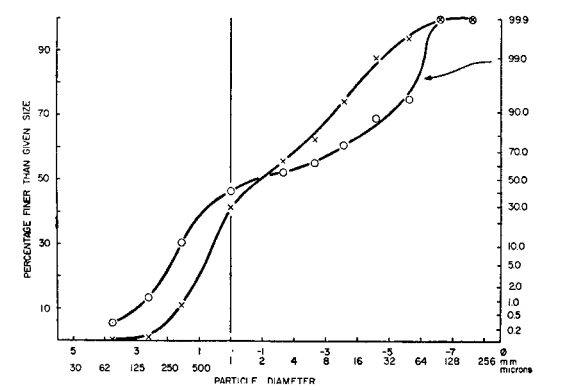

FIG. 5. Cumulative frequency curve of inorganic materials from a pool.

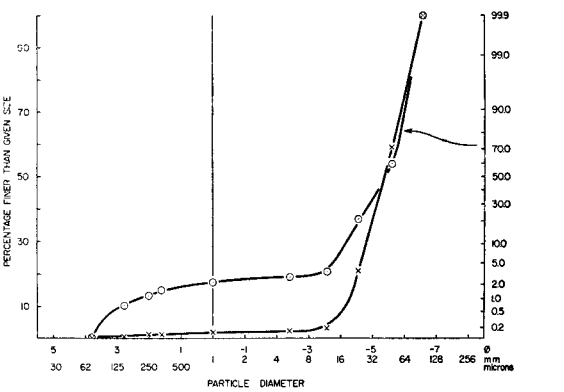

FIG. 6. Cumulative frequency curve of inorganic materials from an artificial spawning channel.

debris are examined and removed. Since there is no subsampling of the animals retained on this sieve, this step in the procedure determines that, whatever treatment is given to the animals retained on subsequent sieves, the counts have to be related to the total sample and not to an aliquot of it.

The filtrate of the 1-mm mesh sieve is washed on a 350-μ sieve, the filtrate of this on a 250-μ sieve, of this on a 150-μ sieve, of this on a 74-μ sieve, and of this on a 50-μ sieve. The residues on these sieves are then subdivided frequently to reduce the time required to count the animals present. Each residue is transferred to a 1000-ml beaker to which water is added to make the contents up to about 400 ml. Into a second beaker of the same size are placed as many upright thin-walled vials as possible. The vials used were 5 cm in height and 1.5 cm in diameter, and the beaker accepted 22; the bundle was held together by a rubber band. About 500 ml of water are added so that the vials are just submerged. The sample in the first beaker is stirred to bring it into suspension and is poured into the second beaker. If a residue is large it may be necessary to treat it in two halves in this way. After the materials have settled the water is siphoned off to a level just below the top of the vials. The band is cut and the vials are lifted out individually and arranged in a row. Those containing about average heights of material are selected; their contents are transferred to small petri dishes, and the animals are counted under a stereomicroscope capable of 50× magnification (the discrimination of small stages of some species requires the preparation of mounts). The area of the mouth of one vial was 1/45 of the cross-sectional area of the beaker so a count of animals from one or more vials could be related to the total material. The principle of subdividing samples by making them settle in a group of containers has also been used by Waters (1969). Its use here is facilitated by the homogeneity of the material resulting from the sieving.

Flotation of the animals in the vials with sugar solution did not prove advantageous, as much of the silt also became buoyant.

The usability of the settling procedure for estimating the total number of invertebrates is dependent upon the animals settling into the vials at random. Even, however, if the variance of the counts exceeds the Poisson variance on occasion, the method may still give adequate accuracy. Tables 1*a* and *b* give results from two series of sieve residues and *c* gives results of treating two filtrates of the 350-μ sieve. This second procedure is appropriate where particle-size analysis of the inorganic materials of a sample is not required; it is then possible to give a single treatment to all the material that has passed through the 350-μ sieve.

Table 1 shows that the counts tend to conform to random distributions as tested by X^2. In a Poisson distribution the accuracy of a count will be determined solely by its size. The degree of accuracy can be expressed

TABLE 1. Counts of invertebrates in sets of settling tubes; (*a*) and (*b*) are series in which the materials were washed on four sieves; (*c*) and (*d*) are series in which the materials had passed through only one coarse sieve. The counts are not significantly different from random at the 0.95 probability level, except in the (*c*) series where one count (58) causes departure from randomness.

Residue on sieve of mesh (μ)						$\bar{m}$	X^2
						(*a*)	
350	21	34	24	31	31	28	4.2
250	48	43	35	37	46	42	3.0
149	44	41	46	41	43	43	0.4
74	13	9	11	12	8	11	1.8
						(*b*)	
350	39	40	49	56	58	48	6.4
250	35	34	44	39	45	39	2.6
149	86	98	82	75	73	83	4.8
74	25	18	28	25	14	22	6.0

(*c*)

Total filtrate through sieve of mesh 350 μ	100	113	70	107	58	103	99	91	101	89	$\bar{m}$ 93	X^2 28.1

(*d*)

Total filtrate through sieve of mesh 350 μ	65	87	85	97	89	91	89	109	84	93	74	79	$\bar{m}$ 87	X^2 15.8

by the standard error as a percentage of the count. At 0.95 confidence limits a count of 100 will fall between 80 and 120; a count of 196 will fall between 168 and 224. The desired accuracy must be chosen and the contents of vials examined until a large enough sample size has accumulated. It is not sound practice to examine a fixed number of vials irrespective of their contents (for a discussion of estimating soil fauna see Jones 1955). If animals are, in fact, rare, a count of 100 is not possible, so a high degree of accuracy is unobtainable. Additional field samples would be required to give an accurate estimate of these.

The manner of sieving is important. Animals can be forced through sieves with jets of water, and are frequently fragmented in this way. The correct washing procedure is to agitate the sieve vertically in a large volume of standing water.

Results

Example 4

A sample taken on July 29 from the riffle examined in example 1 gave an estimated 4378 invertebrates (24,079/m²); 62 distinguishable species were present. Their size distribution (head widths) is shown in Fig. 7 as a histogram and as a cumulative percent frequency curve. It is seen that 80% of the animals have head widths of 250 μ or less; most of these are chironomids. Individual species of insects do not necessarily have their greatest numbers in the smallest size category. For example, a species of *Hydropsyche* in the sample ranged from 120 to 840 μ in head width but the mode was conspicuously at 240 μ.[4] Table 2 shows the percent composition of the taxa in the sample.

Example 5

Another sample from the riffle treated in example 1 was taken on April 15. The fauna of the top 10 cm was collected separately from that between 10 and 32 cm. The total number of invertebrates in the top layer was 5739 and in the lower layer 2400 (total number of all animals amounts to 44,765/m²). Table 2 shows that the Simuliidae account for the most obvious difference in the two layers, but other differences occur. These invertebrates were obtained from the materials analyzed physically in example 1.

Example 6

The fauna of the materials obtained in April from the pool of example 2 comprised 4903 animals (26,966/m²); 3927 invertebrates occurred in the top 10 cm and 967 between 10 and 25 cm. The proportions of Simuliidae, mayfly nymphs, and chironomid larvae are greatly different from that of example 5, and this is expected in view of the physical differences of the habitats (Fig. 4 and 5). Detailed differences are shown in Table 2.

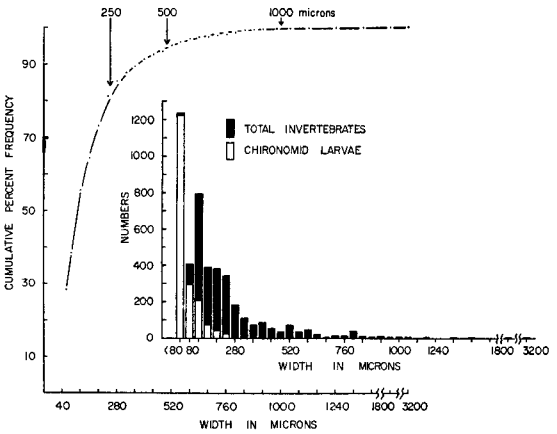

FIG. 7. The size distribution (as head widths) of insects from a riffle, expressed as a histogram (inner figure) and as a percentage cumulative frequency curve (outer figure); arrows point to sizes of meshes of sieves commonly chosen for washing benthos.

Example 7

The fauna of the materials analyzed in example 3 comprised 10,413 animals (57,271/m²) of which 7386 were in the uppermost 10 cm and 3027 were between 10 and 25 cm. Table 2 gives the compositions. The high density is presumably attributable to the low silt content, negligible sand content, high intragravel flows, and to the blanket of filamentous algae that develops on strongly lit surface gravel. This harbors a *Cricotopus* (Orthocladiinae) fauna.

Discussion

Limitations of the Method

One limitation is the depth of water in which the sampler can be used. Geomorphologists sometimes obtain samples when bed materials are temporarily exposed at river margins.

Artificial spawning channels may be slightly too deep in which to operate the apparatus, unless the water level can be temporarily lowered. The collection of fines only, however, is required from artificial channels, as the particle-size distribution of the gravel is known. It might therefore be possible to work with a high-sided sampler and to disturb the gravel with a long-handled trowel.

[4]An obvious cause of polymodal size distributions would be overlapping of generations of a species.

TABLE 2. Numbers and percentage composition of the commoner invertebrates collected. Top refers to the uppermost 10 cm of substrate, bottom to depths below 10 cm. A = adult, P = pupae. See text for descriptions of localities.

	Example 4 Riffle July 29	Example 5 Riffle April 15		Example 6 Pool April 15		Example 7 Spawning channel July 14	
	Total	Top	Bottom	Top	Bottom	Top	Bottom
No. per sample	4378	5739	2400	3927	976	7386	3027
No. per m²	24079	31564	13200	21598	5368	40623	16648
Collembola							
Folsomia sp.	–	–	–	–	–	–	<0.5
Smynthurides sp.	–	–	<0.5	<0.5	–	–	–
Ephemeroptera							
Baetis parvus Dodds	6.6	<0.5	<0.5	2.6	1.8	<0.5	<0.5
Baetis intermedius Dodds	7.8	3.1	<0.5	<0.5	<0.5	<0.5	–
Ameletus sp.	–	–	–	0.5	<0.5	–	–
Ephemerella inermis Eaton	–	<0.5	–	<0.5	<0.5	–	<0.5
Ephemerella spp.	1.5	0.5	<0.5	–	–	<0.5	<0.5
Paraleptophlebia sp.	0.9	–	<0.5	5.5	9.5	0.6	1.3
Iron sp.	–	1.6	<0.5	–	–	–	–
Rithrogena sp.	11.6	<0.5	<0.5	–	–	–	–
Cinygmula sp.	0.9	1.1	<0.5	2.4	2.3	–	–
Cinygma sp.	<0.5	–	–	–	–	–	<0.5
Plecoptera							
Chloroperlinae	1.0	3.8	2.2	2.3	3.6	0.9	<0.5
Isoperla spp.	<0.5	4.0	6.6	7.9	9.3	–	<0.5
Nemoura spp.	<0.5	–	–	–	–	11.8	7.2
Trichoptera							
Hydropsyche sp. 1	–	–	–	–	–	4.6	1.6
sp. 2	–	–	–	–	–	3.4	0.8
sp. (1?)	4.3	0.8	<0.5	<0.5	–	–	–
sp. 2 (P)	–	–	–	–	–	–	<0.5
Cheumatopsyche sp.	–	–	–	–	–	1.5	0.7
Glossosoma sp.	8.8	<0.5	<0.5	<0.5	–	–	–
Wormaldia sp.	–	–	–	–	–	<0.5	<0.5
Rhyacophila spp.	<0.5	<0.5	<0.5	<0.5	–	<0.5	–
Limnephilidae spp.	<0.5	<0.5	<0.5	1.6	1.5	<0.5	–
Oxyethira sp. (P)	–	–	–	–	–	–	<0.5
Other Hydroptilidae	–	–	–	–	–	–	<0.5
Other Hydroptilidae (P)	–	–	–	–	–	–	<0.5
Diptera							
Tanypodinae	0.9	<0.5	1.8	13.2	10.7	9.7	13.0
Diamesinae	<0.5	–	–	–	–	–	–
Orthocladiinae	16.8	9.1	16.2	20.3	13.1	43.2	49.8
Orthocladiinae (P)	–	<0.5	<0.5	<0.5	<0.5	1.0	–
Corynoneura spp.	3.2	3.6	8.0	20.5	11.5	2.6	6.3
Thienemanniella spp.	3.3	2.7	3.6	5.1	7.9	0.9	–
Chironomini	3.2	16.6	34.2	0.5	–	3.3	–
Tanytarsini	11.0	1.0	2.0	11.9	20.7	4.4	4.7
Tanytarsini (P)	–	–	<0.5	–	–	–	–
Ceratopogonidae	0.5	<0.5	2.1	<0.5	<0.5	–	–
Antocha sp.	<0.5	<0.5	–	–	–	–	<0.5

(*Continued*)

TABLE 2. Numbers and percentage composition of the commoner invertebrates collected. Top refers to the uppermost 10 cm of substrate, bottom to depths below 10 cm. A = adult, P = pupae. See text for descriptions of localities. (*Concluded*)

	Example 4 Riffle July 29	Example 5 Riffle April 15		Example 6 Pool April 15		Example 7 Spawning channel July 14	
	Total	Top	Bottom	Top	Bottom	Top	Bottom
Hexatoma sp.	<0.5	<0.5	<0.5	–	–	–	–
Limnophila sp.	–	–	<0.5	–	–	–	–
Other Tipulidae	<0.5	–	<0.5	<0.5	<0.5	–	–
Tipulidae (P)	–	<0.5	–	–	–	–	–
Prosimulium sp.	<0.5	34.8	0.7	<0.5	–	–	–
Prosimulium sp. (P)	–	0.8	–	–	–	–	–
Simulium spp.	–	10.6	<0.5	–	–	2.3	<0.5
Dixidae	–	–	–	–	–	<0.5	–
Blepharoceridae	–	<0.5	<0.5	–	–	–	–
Chrysops sp.	–	–	–	<0.5	–	–	–
Empididae	<0.5	<0.5	–	–	–	<0.5	<0.5
Coleoptera							
Zaitzevia sp. (A)	<0.5	<0.5	–	–	–	–	–
Zaitzevia spp.	2.1	1.3	3.9	<0.5	<0.5	–	–
Narpus sp.	0.7	<0.5	<0.5	–	–	–	–
Oligochaeta							
Nais spp.	<0.5	<0.5	1.4	<0.5	–	<0.5	2.0
Pristina sp.	<0.5	–	–	–	4.1	<0.5	–
Slavina sp.	–	–	–	–	–	1.0	1.0
Lumbriculus sp.	0.8	<0.5	<0.5	–	–	<0.5	<0.5
Eisienella sp.	–	–	–	–	–	–	<0.5
Other Oligochaeta	–	<0.5	0.6	–	–	–	–
Turbellaria							
Cura foremanii (Girard)	<0.5	–	<0.5	–	–	0.7	3.6
Phagocata sp.	–	–	–	<0.5	–	–	–
Acari							
Torrenticola spp.	2.3	<0.5	7.0	<0.5	<0.5	<0.5	<0.5
Unionicola sp.	–	<0.5	–	–	–	–	–
Ljania sp.	<0.5	<0.5	<0.5	<0.5	–	–	–
Hygrobates sp.	–	–	<0.5	–	–	–	–
Lebertia sp.	–	–	–	<0.5	–	–	–
Tyrrellia sp.	–	–	–	–	–	<0.5	<0.5
Sperchon spp.	<0.5	<0.5	–	–	–	–	–
Hydrozoetes sp.	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5
Malaconothrus sp.	4.6	<0.5	–	–	–	–	<0.5
Trimalaconothrus sp.	1.0	–	<0.5	–	–	–	<0.5
Mollusca							
Physa sp.	–	–	–	–	<0.5	0.6	1.1
Planorbidae	–	–	–	<0.5	–	–	<0.5
Sphaeriidae	–	–	–	<0.5	<0.5	–	<0.5

Another limitation is the depth into the substrate to which sampling can penetrate. This might be increased if the sides of the box were extended downwards as the sample is being taken. Metal brackets could be fitted to the inside to accept pieces of wood; these could be lowered to prevent slumping of materials into the sampling area. They would be useful when materials were being

taken from pools, but they could not be operated if large stones were present.

The area of substrate sampled is constant, but the volume of materials removed varies with depth and irregularity of the cavity. This must be taken into account in comparisons of samples or strata.

The total loss of fine silts and clays is an irreducible limitation. The fine net could be made slightly finer, say, to 30 μ mesh size, but this is well outside the clay range. Furthermore, clogging difficulties would occur. Perhaps the assessment of fine silt and clay might be approached by attempting to relate bed materials to suspended load, a subject about which little is known. If fine silts and clay occur in the absence of gravel and cobbles they can, of course, be sampled with corers.

Applications

Fish biologists are concerned with assessing silt and describing quantitatively the physical characteristics of spawning bed materials. Various methods have been described that involve working a device into the substrate (Hatch 1957; McNeil and Ahnell 1964; Rukhlov 1969; Ryan 1970), but have the drawback of causing losses of the silts. The measurement of materials down to 50 μ in size is probably adequate for describing the characteristics of habitats that sustain salmon. (Situations could exist where an exceptional geology or man's influence introduced to a stream bed large quantities of clay; the sampler could not assess these.) Artificial spawning channels have exceptional particle-size characteristics. As they age they accumulate silt and it becomes necessary to clean them. There is need for detailed studies relating the field measurement of fines to fry production. The influence of filamentous algae that develop as extensive mats on the gravel is insufficiently examined, as is the possible role of invertebrates as predators of fish eggs and alevins. The sampler could be used for study of these problems.

A well-tried method for recovering salmon eggs and alevins from gravel exists (McNeil 1964), but has drawbacks in coarse gravels (Merrell 1965). The present sampler should be effective for collecting eggs, provided a single coarse net is used. It is unlikely, however, that the sampler would be useful for collecting alevins, which would quickly move through the gravel. At most, if used promptly, with a net of appropriate mesh, it might provide an index of alevin density.

Living invertebrates can pass through meshes that are smaller than their maximum body width; in dipterous larvae, for example, the diameter of the head capsule determines whether a larva will be retained or not (Jonasson 1955). In general the maximum widths of head capsule of common stream invertebrates fall between 60 μ for newly hatched chironomid larvae to about 5 mm for large Plecoptera (much larger aquatic insects exist but they are not abundant). The younger stages tend to predominate in a natural community. Consequently, even a mesh of 116 μ (Anderson 1967) could allow 50% of the fauna to pass through if the community contained high proportions of chironomid larvae, mayfly nymphs, and stonefly nymphs. The sampler can, of course, be used with a single net of 200- or 250-μ mesh. This would allow perhaps 70–80% of the animals to pass through, but would be adequate for many purposes, such as the estimation of biomass and for general faunistic surveys.

A source of inaccuracy in estimating stream invertebrates may be the depth to which the fauna penetrates. Most samplers retrieve animals from the top 10 to 15 cm of the substrate only, but significant numbers may live below this. Evidence that invertebrates may occur down to 30 cm or more has been obtained (Coleman and Hynes 1970) from the colonization, up to a period of 28 days, of sand and gravel placed in perforated cylinders and buried in the substrate. That higher flows through the substrate than those in the undisturbed state may have occurred was suggested by the initial presence of *Simulium* below 22 cm and by the continued presence of Hydropsychidae (cf. example 4). There seems no reason to doubt that wherever adequate intragravel flows occur invertebrates will be found. Experimental studies show that a clean homogeneous gravel bed with its surface in humps and depressions can have surface water penetrating to a depth of 46 cm (Cooper 1965 p. 21).

It is not possible to make, with the sampler, precise analysis of the vertical stratification of fauna in coarse materials in a stream; nevertheless, subdivision into three layers would be possible. Examination of the fauna at different localities, and at different depths in one locality, in relation both to particle sizes and to quantities of dead organic material, is therefore possible. It is unlikely that the abundance of animals can be accounted for solely in terms of one of these factors (see Ulfstrand 1967; Cummins and Lauff 1969; Egglishaw 1969).

It is not possible to compare the densities of animals found in the examples given with those obtained by other workers. The factor of size selection by other methods, apart from other differences, makes comparison impossible.

One generalization to emerge from the size-frequency distribution is that the times when most insects are present in streams are when most hatching is taking place. Thus, the claim that aquatic insect are scarce at certain times of the year (e.g., late autumn in temperate localities) is likely to be founded on size selection. It is possible that more insects occur then than at any other time of year, but they are present mainly as small instars.

Estimation of production of communities of stream invertebrates is very difficult as many species of different sizes with varied life histories, frequently living in a heterogeneous environment subject to instability, have to be dealt with. Its calculation is dependent, however, on repeated estimates of stocks. Comprehensive data of the complete size distribution of an invertebrate at various stages through its life cycle can be obtained with the sampler and permit direct calculation of rates of production for the population. An accurate measurement of invertebrate stock in riffles is also a requirement for calculating the theoretical capacity of riffles to produce invertebrate drift (see the treatment in Ulfstrand 1968), frequently the main food of the fishes.

The paucity of the hydrologist's methods of sampling bed material is apparent from texts on applied hydrology (e.g., Chow 1964), which mention only grabs and bucket-type samplers. One method, for coarse materials, that provides a representative sample of a whole reach of a stream, is to pick up 100 individual particles from the bed at random. Small sizes are not included, and the stream must be wadeable (Wolman 1954). The sampler described here would seem to offer the geomorphologist the advantage of being able to collect materials down to 50 μ and to select the depth down to which they should be taken. Sizes too large for the sampler can, of course, be recorded independently. The significance of size distributions of bed materials for geomorphologists is discussed by Leopold et al. (1964).

Finally it has been assumed that no biases occur in the sampling procedure of sufficient magnitude to invalidate the results obtained, i.e., that no significant amounts of material are lost from the sampler or gained by it from the outside. This assumption is based on close observation while samples were being taken. Nevertheless, it could be argued that in the absence of any experimental assessment of the efficiency of the sampler it is not possible to say anything about bias. Presumably any errors are most likely to be caused by losses; additions of silt or animals from outside would be small in relation to the total material collected. Losses are likely to be of small particle sizes and therefore of greatest consequence in the estimation of invertebrates.

Bias must be considered with reference to the purpose of the study; consistent bias might not be a serious disadvantage. Assessment of bias would require careful experimental studies in flumes with known contents of silt (e.g., the flume figured in Cooper 1964 p. 38). In view of this it should be borne in mind, when an investigation is being carried out in which great accuracy is necessary, that, even if the sampler gives results superior to those obtained by other methods, possible sources of bias have not been experimentally assessed.

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ALLEN, K. R. 1940. Studies on the biology of the early stages of the salmon (*Salmo salar*). 1. Growth in the River Eden. J. Anim. Ecol. 9: 1–23.

ANDERSON, N. H. 1967. Biology and downstream drift of some Oregon Trichoptera. Can. Entomol. 99: 507–521.

CHOW, V. T. [ed.] 1964. Handbook of applied hydrology. McGraw-Hill Book Company, Inc., New York, N.Y. 1418 p.

COLEMAN, M. J., AND H. B. N. HYNES. 1970. The vertical distribution of the invertebrate fauna in the bed of a stream. Limnol. Oceanogr. 15: 31–40.

COOPER, A. C. 1965. The effect of transported stream sediments on the survival of sockeye and pink salmon eggs and alevin. Int. Pac. Salmon Fish. Comm. Bull. 18: 1–71.

CUMMINS, K. W. 1962. An evaluation of some techniques for the collection and analysis of benthic samples with special emphasis on lotic waters. Amer. Midland Natur. 67: 477–504.

CUMMINS, K. W., AND G. H. LAUFF. 1969. The influence of substrate particle size on the microdistribution of stream macrobenthos. Hydrobiologia 34: 145–181.

EGGLISHAW, H. J. 1964. The distributional relationship between the bottom fauna and plant detritus in streams. J. Anim. Ecol. 33: 463–476.

1969. The distribution of benthic invertebrates on substrate in fast-flowing streams. J. Anim. Ecol. 38: 19–33.

EINSTEIN, H. A. 1964. River sedimentation. *Cited in* Sect. 17, 2 of V. T. Chow, 1964.

HATCH, R. W. 1957. Success of natural spawning of rainbow trout in the Finger Lakes Region of New York. J. N.Y. Fish Game 4: 69–87.

HESS, A. D. 1941. New limnological sampling equipment. Limnol. Soc. Amer. Spec. Publ. 6: 5 p.

INMAN, D. L. 1952. Measures for describing the size distribution of sediments. J. Sediment. Petrol. 22: 125–145.

JÓNASSON, P. M. 1955. The efficiency of sieving techniques for sampling freshwater bottom fauna. Oikos 6: 183–207.

JONES, F. G. W. 1955. Quantitative methods for the estimation of cyst-forming nematodes (*Heterodera* spp.) in soil. *In* D. K. McE. Kevan [ed.] Soil zoology. London Butterworths Scientific Publics, London. xiv + 512 p.

KING, C. A. M. 1966. Techniques in geomorphology. Arnold Ltd., London. ix + 342 p.

LEOPOLD, L. B., M. G. WOLMAN, AND J. P. MILLER. 1964. Fluvial processes in geomorphology. Freeman, San Francisco, California. viii + 522 p.

MACAN, T. T. 1958. Methods of sampling the bottom fauna in stony streams. Mitt. Int. Ver. Limnol. 8: 1–21.

MCNEIL, W. J. 1964. A method of measuring mortality of pink salmon eggs and larvae. U.S. Fish. Wildlife Serv. Fish. Bull. 63: 575–588.

MCNEIL, W. J., AND W. H. AHNELL. 1964. Success of pink salmon spawning relative to size of spawning bed materials. U.S. Fish. Wildlife Serv. Spec. Sci. Rep. Fish. 469: 15 p.

MERRELL, T. R. [ed.] 1965. Workshop on methods in spawning ground studies of Pacific salmon: summary of proceedings. Alaska Dep. Fish and Game Auditorium, Biological Laboratory, Auke Bay, Alaska. 24 p.

MORGANS, J. E. C. 1956. Notes on the analysis of shallow-water soft substrata. J. Anim. Ecol. 25: 367–387.

RUKHLOV, F. N. 1969. Materials characterizing the texture of bottom material in the spawning grounds and redds of the pink salmon (*Oncorhynchus gorbuscha* (Walbaum)) and the autumn chum (*Oncorhynchus keta* (Walbaum)) on Sakhalin. Probl. Ichthyol. (Vop. Ikhtiol.) English-language ed. 9: 635–644.

RYAN, P. 1970. Design and operation of an *in situ* frozen core gravel sampler. Technical Rept. 12. Dep. Fish. Forest. Fish. Serv. Pacific Reg., Vancouver, B.C.

SOUTHWOOD, T. R. E. 1966. Ecological methods with particular reference to the study of insect populations. Methuen, London. xvii + 391 p.

SURBER, E. W. 1937. Rainbow trout and bottom fauna production in one mile of stream. Trans. Amer. Fish. Soc. 66: 193–202.

ULFSTRAND, S. 1967. Microdistribution of benthic species (Ephemeroptera, Plecoptera, Trichoptera, Diptera: Simuliidae) in Lapland streams. Oikos 18: 293–310.

1968. Benthic animal communities in Lapland streams. Oikos Suppl. 10: 120.

WARREN, C. E., J. H. WALES, G. E. DAVIS, AND P. DOUDOROFF. 1964. Trout production in an experimental stream enriched with sucrose. J. Wildlife Manage. 28: 617–660.

WATERS, T. F. 1969. Subsampler for dividing large samples of stream invertebrate drift. Limnol. Oceanogr. 14: 813–815.

WATERS, T. F., AND R. J. KNAPP. 1961. An improved stream bottom fauna sampler. Trans. Amer. Fish. Soc. 90: 225–226.

WOLMAN, M. G. 1954. A method of sampling coarse riverbed material. Trans. Amer. Geophys. Union 35: 951–956.